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Research Article

Camel Milk Quality and Bacterial Contamination along Market Chain in Wajir and Garissa Counties of Kenya

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Abstract

This study endeavoured to establish the physical characteristics and bacterial contamination of camel milk along market chain in North-Eastern Province of Kenya. The objective of the study was to identify various points of milk contamination with a view of developing intervention strategies that can improve milk quality leading to improved productivity for pastoralists in the region. The investigation was done on various marketed camel milk samples collected from two counties: Garissa and Wajir. This was considered important because milk is a very nutritious substance that readily supports growth of microorganisms; which is more encouraged when the conditions are hot. Parameters used to assess the physical characteristics included organoleptic tests, measurement of specific gravity, determination of pH, and alcohol test. Bacteriological parameters included: Total coliform count, Total viable bacterial count, and Resazurin test. The study showed that.289 samples [75.26%] had gross dirt/particulate matter including grass/leaves, sand/soil particles and/or black charcoal particle. Thirty four [34] samples [8.85%] had an abnormal yellowish colour. Formation of flakes in the Alcohol test was recorded in 128 samples [33.33%] indicating they were either acidic, mastitic or colostrum milk. This explains the high values of more than 107 cfu/ml of TCC and TVBC observed in most of the samples [80%] in the present study. Escherichia coli O157:H7 was identified from one of the samples that were serotyped with Escherichia coli antisera O157 and H7. This organism can cause severe disease in humans. Other bacterial microorganisms isolated from the milk samples alongside the coliforms included: Staphylococcus species [90.10% = 346 samples], Streptococcus species [84.90% = 326 samples] and Bacillus species [45.83% = 176 samples]. Of the 346 Staphylococcus species isolated, 91 [23.70%] were coagulase-positiveThis could have been due to inadequate washing of milk containers and poor personal hygiene of the milkers, as a result of there being inaccessibility of soap and insufficient clean water, as reported in another study. Such contaminated milk spoils easily and is a prelude to both clinical and subclinical, which results in reduced milk production; both causing economic loss to the farmer.This information is expected to be of benefit to policy-makers in their efforts towards improving milk quality and safety.

INTRODUCTION

The camel population in Kenya is estimated to be at about one million, of which more than half are reared in North-Eastern Province. Camel milk, in pastoral areas, is produced in areas with several challenges that include environments that are hot, dusty and distant, with scarcity of water, transport and infrastructural facilities [1]. Most camel milk in pastoral areas is thus kept and transported at high ambient temperatures due to lack of refrigeration facilities. These conditions make the milk spoil fast and unsafe; i.e. capable of causing food-borne diseases. Most of the milk is collected by retailers who then bulk it for sale. The fact that, currently, there is wide demand for camel milk – it is marketed in urban areas like Garissa, Wajirand Eastleigh, Nairobi has necessitated the need to test for its quality, so that measures can be put in place to make sure that the sold milk is safe for human consumption. Testing for possible adulteration of the milk is also necessary as this will contribute towards lowering of the milk quality. This study endeavoured to establish the physical characteristics and bacterial contamination of camel milk produced in the two counties. This was found to be important because milk is a very nutritious substance that readily supports growth of microorganisms; which is more encouraged when the conditions are hot.

MATERIALS AND METHODS

Study area

The study was done in Garissa and Wajir counties of North-Eastern Province, They lie in the Arid and Semi-Arid Lands [ASAL] of Kenya. The rainfall pattern is erratic and unreliable; it is always less than 600 mm annually. Temperature ranges between 22^0 C and 42^0 C. The districts are flat, covered by trees and shrubs with grass undergrowth. Water sources are rivers [permanent and seasonal], pans, boreholes, dams and shallow wells. The mainstream activity of the two districts is livestock keeping. The animals are kept under pastoralist system. They include cattle, sheep, goats, camels, donkeys and poultry. Nomadic pastoralist communities living in ASAL regions largely depend on milk produced by camels which contribute 80% of the household needs [2,3, 4].

Wajir County lies between latitudes 3^o 6" N and 0^o20"N and longitudes 39^0 E and 41^0E. It borders the republic of Somalia to the East, Garissa County to the South, Isiolo to the South-West, Marsabit to the West, Moyale is to the Northwest, Ethiopia to the North and Mandera to the North-East. The county covers an area of 56,501km^2, divided into 13 administrative divisions. The

county's population is 533,537 persons [1999 Kenyan census] with annual growth rate of 3.7%. The main form of land use is nomadic pastoralism, which is the most efficient method of exploiting the range land. Incidence of insecurity as a result of banditry in the area is quite high because of the porous borders. Locations that were selected for sampling were conveniently chosen; they included those that had large populations of camels. These were: Griftu,Khorl-Haral,Tarbajand Wajir-Bor.

Garissa County has a population of 329,939 [1999 Kenyan census]. It has an area of 44,952 Km^2. It is located near 0^0 27`25" S, $39^0$ 39`30"E and has a population of 65,881, according to 1999 Census [Populations of local authorities with towns-1999]. Tana River flows through the county. Most of the inhabitants of Garissa are ethnic Somalis. Locations that were selected for sampling were also conveniently chosen; they were: Korakora, Kulan, and Damajale.

Sampling

A total of 384 samples of milk produced by locally-kept camels were collectedfrom 30 marketing centres where 15-20 samples per site were collected in the two districts. Volumes of 200 to 300 mlsof bulk camel milk [from producers or hawkers] were collected into labelled sterile bottles and kept in an ice box, transported to laboratory for bacteriological culture and identification, which was done either immediately or after keeping them for not more than 24 hours in a refrigerator.

Study design

The study was cross-sectional. The collected milk samples from various milk outlets were investigated using various parameters including: assessment of the physical characteristics of milk andassessment of milk quality [bacteriological carriage]. Assessment of physical characteristics of milk included: carrying-out of organoleptic tests, determination of specific gravity, pH of the milk, and doing alcohol test [6]. Organoleptic tests involved physical observation of the milk for gross dirt, colour, consistency and smell for odours. Assessment of milk quality [bacteriological carriage] was done by determining the Total Viable Bacterial Count [TVBC] and Total Coliform Count [TCC] and by doing Resazurin test, which is used to determine the microbial load in milk [5,6]. Characterisation of the isolated bacteria, including *E. coli* O157:H7, was also done,

Assessment of physical characteristics

Gross dirt assessment: The sample of the milk collected [200 – 300 ml] was filtered /sieved using a clean sterile sieve or clean filter paper into a clean container [glass flask] and the gross dirt recovered was noted and recorded.A positive sample was denoted by the recovery of gross-dirt which included pieces of grass/leaves, soil or sand particles, charcoal particles [black] or any other particulate matter.

Colour assessment: The colour of the milk sample collected was assessed visually; noting that normal camel milk is white in colour. Any colour change observed from the normal white was noted and recorded. Colour change mostly noticed was yellowish.

Odours/smell assessment: The milk samples collected were assessed for any bad smell/odours.The sample with good normal smell was recorded as negative for bad odours/smell and that with bad smell/odour [sour smell] was recorded as positive for bad odour.

DETERMINATION OF SPECIFIC GRAVITY OF CAMEL MILK

A 250ml glass cylinder was first half-filled with camel milk at 20^0 C. Lactodensimeter[lactometer] was then inserted; and more milk was added to fill the glass cylinder to the brim. Reading of the specific gravity was taken directly from the lactometer, while kept at temperature of 20^0 C

The density of cattle milk ranges between 1.026 g/litre and 1.034 g/litre [Giangiacomo, 2001]. The mean specific gravity of camel milk is 1.0305 g/litre, with an average butter fat content of 3.678% [7,8]. Specific gravity was determined by means of lactodensimeter[lactometer] at 15 - 20^0C. When cattle milk is adulterated with water, specific gravity will be less than 1.026 g/litre while in cases of adulteration with solids like sugars, specific gravity will be higher than 1.034 g/litre [6].

Determination of pH of camel milk

pH of camel milk was measured with a pH indicator paper [Universal-Indikatorpapier – Germany] which was dipped in the milk sample and the resulting colour assessed against the standard values provided on the pack. The pH value was read as it matched the respective colour on the standard chart.

This gave the rough estimate of the acidity of milk. The normal values for milk are 6.6 – 6.8. Lower values generally mean acidification process due to development/growth of bacteria. Higher values mean presence of mastitis [6].

Alcohol test

Alcohol test was used to determine acidic, mastitic and colostrum milk, which was unsuitable for further processing. This test used 68% alcohol according to the standard method [6]. Five [5] ml camel milk was mixed with 5 ml of 68% alcohol in a clean test tube. Formation of flakes indicated unsuitability. A positive alcohol test was denoted by formation of flakes, indicating unsuitability while the negative one formed no flakes.

Assessment of bacteriological quality

Bacteriological quality of milk was measured using "Total Viable Bacterial Count [TVBC]", "Total Coliform Count [TCC]" and "Resazurin test".

TOTAL VIABLE BACTERIAL COUNT

Total Viable Bacterial Count [TVBC] was determined using standard Plate Count Agar [PCA] media [9]. Serial dilutions of milk samples were carried out. Briefly, one millilitre [ml] of 10-fold serially diluted milk sample [1ml milk sample in 9ml potassium hydrogen sulphate buffer or normal saline] was placed on a Petri dish, followed by pouring of 20ml molten Plate Count Agar [PCA] cooled to 45^0 C onto the dish [9]. The sample and the agar were then mixed and left to solidify, after which the plates were incubated at 37^0 C for 24 -48 hours. Bacterial colonies [colony forming units **[cfu]** between 30 and 300] were counted using a manual colony counter and multiplied by the dilution

factor to get TVBC value in colony forming units per ml **[cfu/ml]** of milk [6,10,11]. Four plates were inoculated with each dilution and an average number calculated.

Total coliform count

Total Coliform Count [TCC] was determined using Violet Red Bile Agar [VRBA] medium, which is selective for coliforms, according to United States [US] standard method [12]. The TCC served as an indicator of faecal contamination, and therefore poor hygiene and public health risk if numbers present exceeded the Kenya Bureau of Standard [KEBS] set limits [5]. One [1] millilitre of 10-fold serially diluted milk sample [1 ml of milk sample in 9 ml Potassium hydrogen sulphate buffer or normal saline] was placed in a Petri dish, followed by pouring of 20 ml molten Violet Red Bile Agar [cooled to 450C]. The sample and the agar [VRBA] were then mixed well and left to solidify, after which the plate was incubated at 370 C for 24 - 48 hours. Bacterial colonies [colony forming units [cfu] between 30 and 300] were counted using a manual colony counter [four plates were inoculated with each dilution and an average number calculated]. The average number of cfus was then multiplied by the dilution factor to get TCC value, expressed in colony forming units per ml [cfu/ml] of milk. .

Resazurin test

Resazurin test was used to determine the microbial load in milk [5.6]. Briefly, Resazurin solution was prepared by dissolving one Resazurin tablet in 200 mls of hot distilled water. One milliliter of the dye solution was placed in a sterile 15 ml test tube and 10 mls of the milk sample added. The tube was then stoppered, placed in an incubator at 36^0 C for one hour, examined and classified according to the resultant colour [13,14] at the end of the one hour. The result of the test was read according to the reference table provided which gave a relationship of colour and the quality of milk after incubation for a specified time [one hour].

Culture and identification of theisolated bacteria: Bacteriological examination was carried out following standard methods [15,16,17.], using Blood agar, McConkey agar, Eosin Methylene Blue [EMB]medium, Mannitol salt agar. Inoculated plates were incubated aerobically at 37^0 C for 24 – 48 hours. Presumptive identification of bacterial isolates on primary culture were made based on colony morphology, haemolytic characteristics on blood agar and production of greenish metallic sheen on EMB medium. Respective biochemical tests, including coagulase test for *Staphylococcus* and CAMP test for *Streptococcus*, were carried out.

Primary bacterial isolation was done in the field laboratory [Garrissa District Veterinary Investigation Laboratory]. Bacterial colonies from the two primary isolations [7 % Sheep Blood Agar and MacConkey Agar] were inoculated into Nutrient Agar slants [Transport media], incubated at 37 ^{0} C for 12 hours, and then stored at 4^0C in Garissa VIL. These colonies were later transported in a cool box to the University of Nairobi, Bacteriology laboratory, for secondary bacterial culture and further biochemical testing/ characterization, using the same type of media.

All bacterial isolates were preserved in glycerol-Nutrient broth at 0^oC until time to work on them.

Search for *E. coli* serotype O157:H7: From the *E. coli* bacteria isolated above, an attempt was made to isolate and identify serotype O157:H7, which is capable of producing shiga-like toxin. This was done using the methods described by [18] and [19] using Sorbitol MacConkey [SMAC]. The medium was incubated aerobically at 37o C overnight. Non-sorbitol-fermenting, colourless colonies on this medium were taken as suspect organisms. Due to limitation of antiserum, only 16 samples were serotyped for O157:H7. These were randomly selected. The serotyping was done at Kenya Medical Research Institute [KEMRI], Centre for Microbiology, using standard procedure.

RESULTS

Assessment of the physical characteristics of camel milk samples

Results of gross dirt, colour, bad odour, pH and alcohol test reactions, with respect to the milk samples, for the 2 study counties, separately and combined, are given on Table 1 and Figures 1 and 2a, b and c**.** Both Garissa and Wajir milk samples showed similar patterns of high gross dirt content [over 70%; slightly more in Wajir than in Garissa], most of the milk being white [over 80%; slightly more in Garissa than in Wajir], while yellowish milk was at less than 10% [slightly more in Wajir than in Garissa], bad odour at about 20% [more in Garissa than in Wajir], alcohol test reaction at about 33% [both areas giving almost the same percentage], more milk samples in Wajir [40.9%] than in Garissa [24,4%] had pH of 6, more milk samples in Garissa[57.8%] than in Wajir [45.5%] had pH of 7, and more milk samples in Garissa[17.8%] than in Wajir [13.6%] had pH of 8. Although most of the milk samples were at neutral pH, the acid and alkaline pHs denote possibility of bacterial effect or other chemical change.

Table 2 gives specific gravity figures/percentages for the milk samples, for the 2 study counties, separately and combined. Respective frequency comparisons are given in Figure 3. More milk samples in Wajir [44.8%] than in specific gravity ranging between 1.030-1.032. Overall, most milk samples [45.4%] had specific gravity ranging between 1.025-1.029; a slightly lower percentage [40.5%] had specific gravity ranging between 1.019-1.024.

Results on total viable bacterial count, total coliform count, Resazurin test, with respect to the 2 districts, separately and combined

These are given on Tables 3a and b and Figures 4 and 5**.** The total coliform count had more samples giving high counts, highest number being within the 110-190x10^5cfu/ml bracket [47.4% for Garissa and 42.9% for Wajir]. The pattern, with respect to concentrations, was similar for the 2 places, although Garissa recorded slightly higher figures than Wajir.

The reverse was the case for viable bacterial counts – most of the samples fell within the 110-190x10^5 bracket [73.5% for Garissa; 63.6% for Wajir]. The pattern was also similar for both places, being higher for Garissa samples.

Reading of the Resazurin test was by colour change and

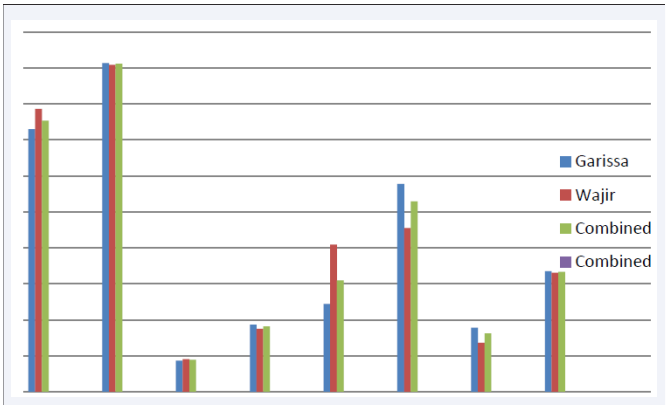

Figure 1 Comparison of frequencies, in percentages, of milk sample physical characteristics, presented per characteristic.

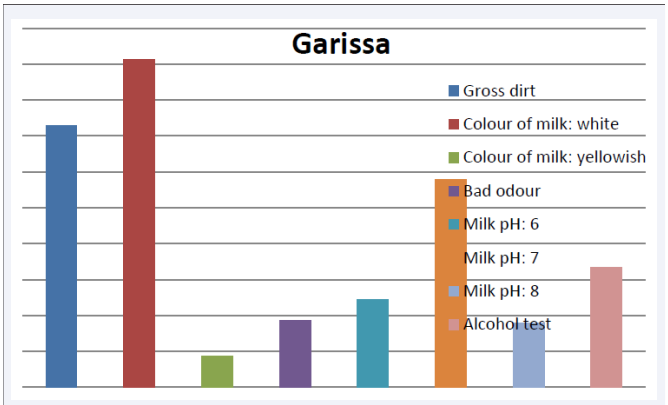

Figure 2a Frequencies, in percentage, of milk sample physical characteristics, for Garissa County.

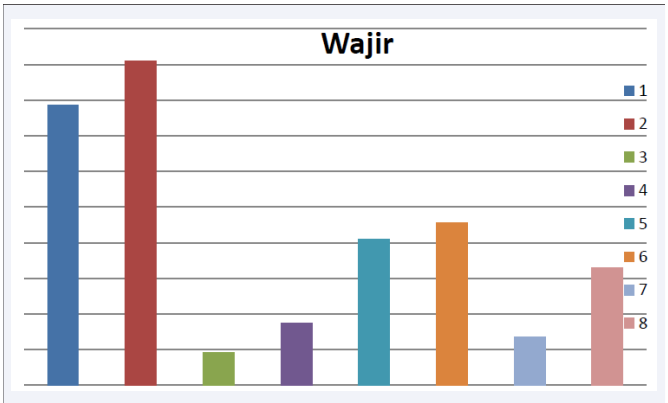

Figure 2b Frequencies, in percentage, of milk sample physical characteristics, for Wajir County.

referred to bacterial quality of the milk sample; blue colour denoting "Excellent", deep mauve colour denoting "Good", deep pink colour denoting "Fair", pinkish-white colour denoting "Poor" and white denoting "Bad". In this study, the bacterial count brackets 120-190x10^4, 110-190x10^5 and 110-120x10^{6}cfu/ml gave Resazurin readings of "Excellent", "Good" and "Fair", respectively. Count brackets 130-160x10^6 and above were rated as "Poor" and "Bad" [the actual counts were not ascertained].

The Resazurin test picked other milk qualities which were not covered in the viable cell scaling. These are given in Table 4 and Figure 6. Sixteen point five percent [16.5%] of the samples in Garissa and 15.6% of the samples in Wajir were rated "Fair" by Resazurin testing, while 9.6% of samples in Garissa and 14.3% of samples in Wajir were rated "Bad".

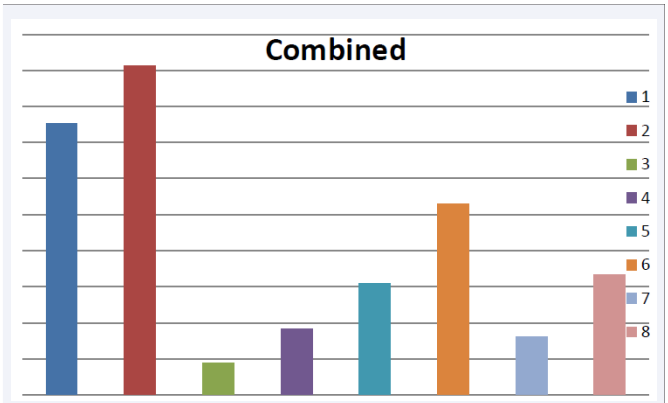

Figure 2c Frequencies, in percentage, of milk sample physical characteristics, for combined Garissa and Wajir counties.

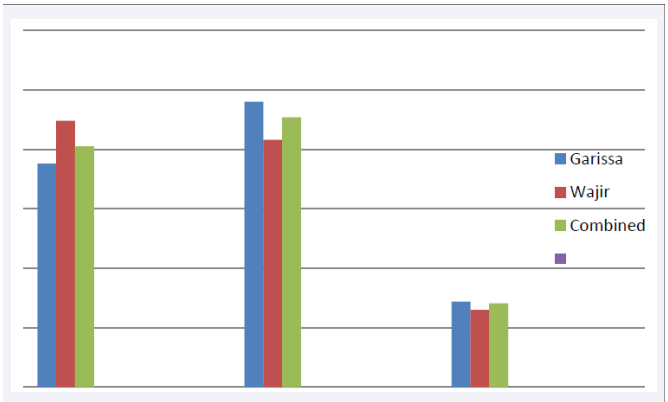

Figure 3 Frequencies, in percentage, of milk sample specific gravity ranges, for the 2 study areas, separately and combined.

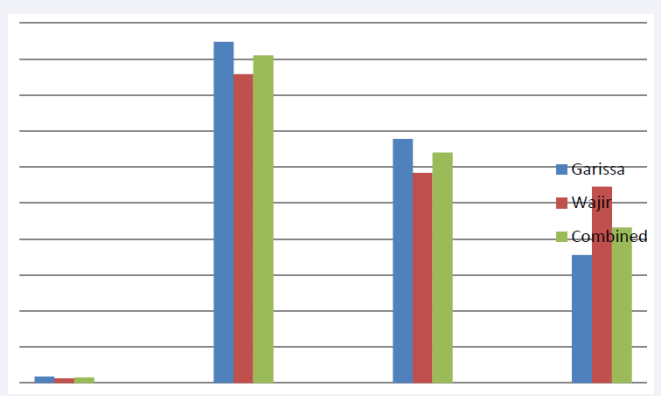

Figure 4 Frequencies, in percentage, of coliform isolation brackets [presented as colony forming units [cfu]/ml], for the 2 study areas, separately and combined.

Table 1: Results on gross dirt, colour, bad odour, pH and alcohol test reactions, with respect to the milk samples, for the 2 study counties, separately and combined.

		Garissa n=230		Wajir n=154		Combined n=384	
		Number +	%	Number +	%	Number +	%
Gross dirt		168	73.0	121	78.6	289	75.3
Colour of the milk sample	White	210	91.3	140	90.9	350	91.2
	Yellow	20	8.7	14	9.1	34	8.9
Bad odour/smell		43	18.7	27	17.5	70	18.2
pH reading	pH 6	56	24.4	63	40.9	119	31.0
	pH 7	133	57.8	70	45.5	203	52.9
	pH 8	41	17.8	21	13.6	62	16.2
Alcohol test reactions		77	33.5	51	33.1	128	33.3

Footnote:
- Gross dirt includes grass, leaves, particles of sand/soil/charcoal
- Alcohol test was detected through flakes formation

Table 2: Specific gravity figures [in grammes per litre] for the milk samples, for the study counties, separately and combined.

Specific gravity ranges	Garissa: n=229		Wajir: n=154		Combined: n=383	
	Number Within range	%	Number Within range	%	Number Within range	%
1.019-1.024	86	37.6	69	44.8	155	40.5
1.025-1.029	40	48.0	64	41.6	174	45.4
11.030-1.032	33	14.4	21	13.0	54	14.1

Note: The specific gravity of one sample [from Garrissa District] was not determined since the sample quantity was too small [30 ml] to be determined by a lactometer.

Table 3a: Results on total viable bacterial count [presented as cfu/ml] and Resazurin test, with respect to the 2 counties, separately and combined.

		Total viable bacterial count			
		$120\text{-}190x10^4$	$110\text{-}190x10^5$	$110\text{-}120x10^6$	$130\text{-}160x10^6$
Garissa N=230	Number	46	169	10	5
	%	20.0	73.5	4.4	2.2
Wajir N=154	Number	35	98	13	8
	%	22.7	63.6	8.4	5.2
Combined N=384	Number	81	267	23	13
	%	21.1	69.5	6.0	3.4
Resazurin rating per bacterial count		Excellent [Blue colour]	Good [Deep mauve colour]	Fair [Deep pink colour]	Poor [whitish pink]

Table 3b: Results on total coliform count [presented as cfu/ml] and Resazurin test, with respect to the 2 counties, separately and combined.

		Total coliform count [TCC]			
Garissa N=230	Number	2	109	78	41
	%	0.9	47.4	33.9	17.8
Wajir N=154	Number	1	66	45	42
	%	0.7	42.9	29.2	27.3
Combined N=384	Number	3	175	123	83
	%	0.8	45.5	32.0	21.6
Resazurin rating per bacterial count		Excellent [Blue colour]	Good [Deep mauve colou]	Fair [Deep pink colou]	Poor [whitish pink]

Key for tables 3a and b: cfu means colony forming units

Bacteria isolated from the camel milk samples

Table 5 shows bacteria [and their respective prevalences] isolated from the camel milk samples from Garissa and Wajir, respectively, and as combined data, while Figure 7 gives comparison of occurrences, with respect to the various bacteria isolated; Figures 8 and 9 give breakdowns of isolated *Staphylococcus* and *Streptococcus*, with respect to coagulase production and CAMP reaction, respectively.From the 384 samples processed, using various media, 230 [59.9%] of the isolates were *E. coli* while 368 [95.8%] were *Klebsiella/ Enterobacter.* This means some samples yielded more than one

type of microorganism.

The 2 areas had similar patterns of bacterial occurrences; the highest across board was *Klebsiella/Enterobacter* group, isolated at 96%, followed, frequency-wise, by *Staphylocccus* [94% in Wajir and 88% in Garissa]; *Streptococcus,* at 85% in both areas; *E. coli* [Garissa 59.9% and Wajir 40.3%]; and lastly *Bacillus* [Garissa 45.8% and Wajir 50.4%]. Thus, Garissa yielded more of *Bacillus* and *E. coli* organisms than Wajir, while Wajiryieded more *Staphylococcus* than Garissa. *Streptococcus* and *Klebsiella/ Enterobacter* were isolated at more-or-less the same rate in both areas [Table 5; Figure 7].

Garissa yielded more [30%] coagulase positive *Staphylococcus* than Wajir[14.3%], despite more *Staphylococcus* having been isolated from Wajir[93.5%] as compared to Garissa [87.8%] [Figures 7,8]. Garissa also yielded more [31.3%] CAMP positive *Streptococcus*than Wajir[19.5%]; *Streptococcus* was isolated at same prevalence [85%] for the two areas [Table 5, Figure 7].

E. coli O157:H7

Of the 230 *E. coli* isolates streaked onto Sorbitol MacConkey, Table 6 gives a breakdown of suspect and non-suspect strains isolated, per county. When 16 of the suspect samples were serotyped for O157:H7, one sample gave positive reaction; the others were negative. Thus the percent positive cases translated to a minimum of 6.25%.

DISCUSSION

A total of 384 camel milk samples were collected from Garissa [230 samples] and Wajir [154 samples] counties and were used to determine milk quality, through assessment of physical characteristics and bacteriological carriage. Parameters used to assess the physical characteristics included organoleptic tests [assessment of gross dirt, colour and smell/odour], measuring of specific gravity, determination of pH and alcohol test. Bacteriological parameters included: Total Coliform Count [TCC; done on Violet Red Bile Agar [VRBA]], Total Viable Bacterial Count [TVBC; done on Plate Count Agar [PCA]] and Resazurin test [which gauges the level of microbial load in the milk].

Assessment of physical characteristics of the camel milk samples

A combined assessment of physical characteristics of the 384 camel milk samples from the two districts [Garissa and Wajir], showed that 289 samples [75.26%] had gross dirt/particulate matter including grass/leaves, sand/soil particles and/or black charcoal particles. This could be attributed to the low level of hygiene in cleaning of the milk containers and lack of milk filters after milking; before packing the milk in containers. The black charcoal particles were attributed to the tradition of smoking milk containers especially traditional gourds as had been observed in another study carried-out by the researchers and by other researchers [20]. Camel milk is traditionally produced by way of hand milking, handled and transported under low hygienic conditions and the common practice of smoking traditional milk containers and milking buckets [made from gourds, natural fibres] contributes to the introduction of gross dirt, especially charcoal particles, in the milk [20].

Thirty four [34] samples [8.85%] had an abnormal yellowish colour. This is a deviation from the normal white opaque colour of camel milk. Such milk is unsuitable for consumption; hence unsuitable for further processing. Indeed, there was a strong positive correlation [26 out of the 34 samples; 76.47%] between the yellowish colour of milk and flake formation in the Alcohol test, which is used to determine acidic, mastitic and colostrum milk. Apart from being white opaque in colour, normal camel milk has a faintly sweetish odour and a sweet but sharp taste. It is thinner than cow or buffalo milk [21,22]. Camel milk has a much slower natural creaming rate than cow milk, both in its raw and heat treated states [23 Farah &Ruegg, 1991; 24 Farah, 1993]. Seventy [70] samples [18.23%] had offensive/bad odour/smell [sour or foul smell]; the smell was that of fermenting or souring milk. There was a positive correlation [26 out of the 70 samples; 37.14%] between the bad smell and the acid pH of "6" found in these milk samples.

The range of specific gravity of the samples tested was between 1.019 g/litre to 1.032 g/litre with 155 samples [40.47%] being between 1.019 – 1.024 g/litre range, 174 samples [45.43%] being between 1.025 – 1.029 g/litre and 54 samples [14.10%] being between 1.030 – 1.032 g/litre range. The mean specific gravity of normal camel milk is 1.0305 gms/litre, with an average butter fat content of 3.678% [7,8]. When camel milk is adulterated with water, specific gravity will be less than 1.026 g/ litre while in cases of adulteration with solids like sugars, specific gravity will be higher than 1.034 g/litre [6]. As many as 155 samples [40.47%] had a specific gravity between 1.019 – 1.024 g/litre, an indication of adulteration of marketed camel milk; thus casting great concern on the quality of camel milk supplied to the consumers. Addition of up to 15% water to marketed camel milk has been reported from southern Somalia [20]; the quality of the added water presenting an additional hygienic risk. The specific gravity of camel milk tested in three large commercial herds in Kenya over a two months' period varied between 1.026 g/litre and 1.029 g/litre, indicating a difference to the specific gravity of the camel's milk [20].

Results of pH determination indicated that 119 samples [30.99%] had a pH of "6", 203 samples [52.86%] had a pH of "7" and 62 samples [16.15%] had a pH of "8". This gave a rough estimate of the acidity of milk. The normal values for milk are 6.6 – 6.8. Lower values generally mean acidification process due to development of bacteria. Higher values mean presence of mastitis [6].

Formation of flakes in the Alcohol test was recorded in 128 samples [33.33%] indicating they were either acidic, mastitic or colostrum milk. Formation of flakes indicates unsuitability of the milk for consumption; this also means the milk is unsuitable for further processing

Comparison of Garissa and Wajir counties, with respect to physical properties

Samples from Wajir County had a higher incidence of having gross dirt [by 5.53%]than those from Garissa County. The incidence of adulteration of milk was thus higher in Wajir district [by 7.26%] than in Garissa district. Wajir district had a higher incidence of acidic milk [by 16.56%] than Garissa district.

This could be attributed to the long distance travelled by the pastoralist from the grazing fields to the watering wells where the market is available.

Analysis for milk hygiene, with respect to total coliform count and Total Viable bacterial count

Total coliform counts [TCC], given as colony forming units per millilitre of milk [cfu/ml], was assessed against the various parameters which included gross dirt in milk, colour of the milk sample, presence of bad odour, Alcohol test, Resazurin test, isolation of *Escherichia coli, Klebsiella* species and *Enterobacter* species. Milk samples with gross dirt had a higher mean [89.11× 10^6] of TCC than the negative samples [75.64× 10^6]. Milk samples having yellowish colour had a higher mean [94.45× 10^6] of TCC than the samples with the white colour [84.93× 10^6]. Milk samples with bad odour had a higher mean [92.96× 10^6] of TCC than the negative samples [84.35× 10^6]. Milk samples with a positive alcohol test had a higher mean [94.38× 10^6] of TCC than the negative samples [81.53× 10^6]. Milk samples with an excellent Resazurin test had the lowest mean [56.63× 10^6] of TCC compared with samples with poor Resazurin test [106.30× 10^6]. Milk samples with no *Escherichia coli* isolation had a lower mean [79.78× 10^6] of TCC compared to the ones where sorbitol and non-sorbitol fermenting *Escherichia coli* were isolated [80.89× 10^6 and 93.74 ×10^6 respectively]. Milk samples with *Klebsiella/Enterobacter* species isolation had a higher mean [87.12× 10^6] of TCC compared to the samples where no *Klebsiella/Enterobacter* species were isolated [85.68× 10^6]. The above results show that there was a strong positive correlation between the parameters used here [i.e. positive gross dirt, positive abnormal colour, positive bad odour, positive alcohol test, poor Resazurin test and isolation of *Escherichia coli, Klebsiella*species, and *Enterobacter*

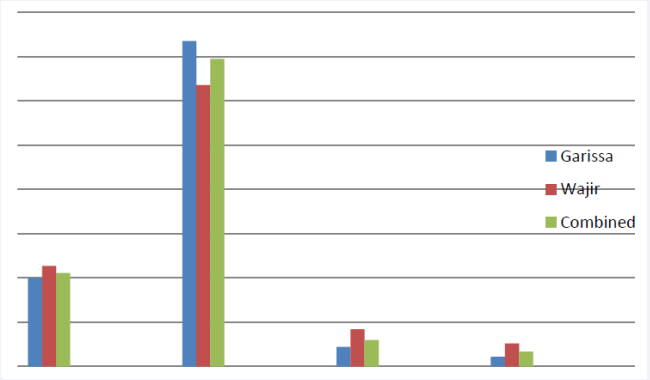

Figure 5 Frequencies, in percentage, of viable bacterial isolation brackets [presented as colony forming units [cfu]/ml], for the 2 study areas, separately and combined.

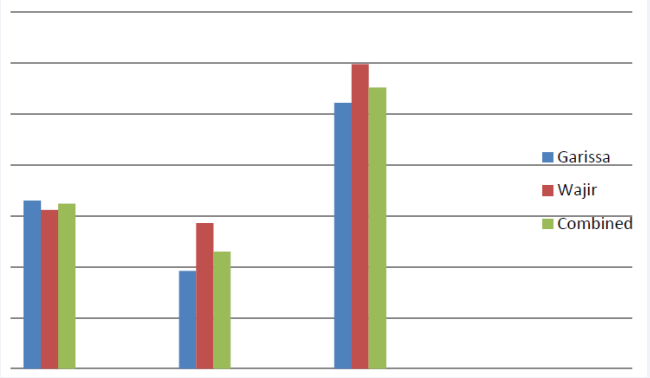

Figure 6 Frequencies, in percentage, of Resazurin-based milk quality gradings, for the 2 study areas, separately and combined.

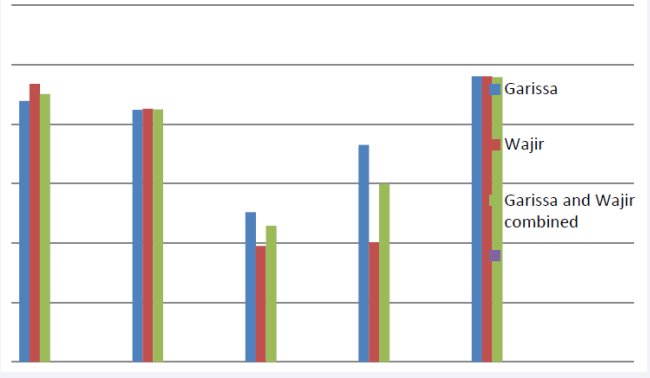

Figure 7 Comparison of percentage occurrences per bacterial organism.

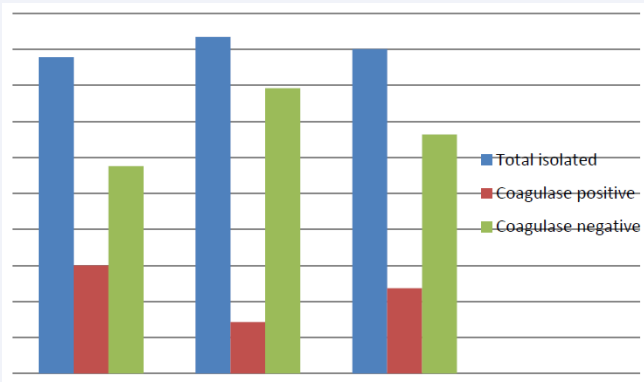

Figure 8 *Staphylococcus* prevalences [%]: Total, Coagulase positive, Coagulase negative.

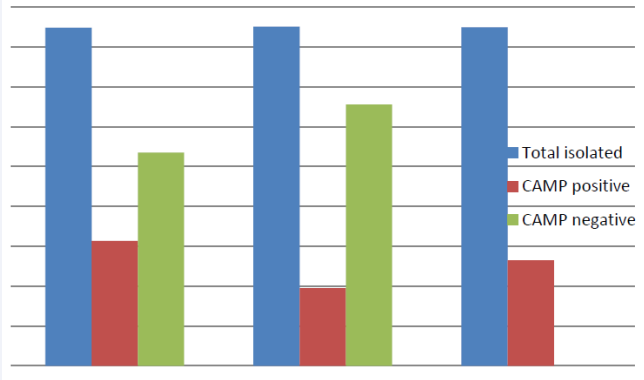

Figure 9 *Streptococcus prevalences [%]: Total, CAMP positive, CAMP negative.*

Table 4: Resazurin results for milk samples from the 2 counties, separately and combined.

	Poor [pinkish-white		Bad [white]		Total poor/bad quality samples	
	Number	%	Number	%	Number	%
Garissa n=230	38	16.5	22	9.6	60	26.1
Wajir n=154	24	15.6	22	14.3	46	29.9
Composite n=384	62	16.2	44	11.5	106	27.6

Table 5: Bacteria isolated from the camel milk samples from Garissa and Wajir counties, separately and collectively.

		Garissa n = 230		Wajir n = 154		Garissa and Wajir Combined n = 384	
		No.	%	No.	%	No.	%
Staphylo coccus	Total isolated	202	87.8	144	93.5	346	90.1
	Coagulase positive	69	30.0	22	14.3	91	23.7
	Coagulase negative	133	57.6	122	79.2	255	66.4
Strepto coccuc	Total isolated	195	84.8	131	85.1	326	84.9
	CAMP positive	72	31.3	30	19.5	102	26.5
	CAMP negative	123	53.5	101	65.6	224	58.3
Bacillus spp.		116	50.4	60	39.0	176	45.8
E. coli		168	73.0	62	40.3	230	59.9
Klebsiella/ Enterobacter		221	96.1	148	96.1	368	95.8

Table 6: Breakdown of samples yielding suspect and non-suspect *E. coli O157:H7* colonies for the 2 counties, separately and combined.

		Garissa n=168		Wajir n=62		Combined n=230	
		Number	%	Number	%	Number	%
Suspect *E.coli* O157:H7	+ve	67	39.9	5	8.1	72	31.3
	-ve	101	60.1	57	91.9	158	68.7
Total number of *E. coli* screened		168	100	62	100	230	100

species] and the poor hygiene of camel milk. This phenomenon was largely attributed to poor handling [poor sanitary practices] of camel milk during milking and subsequent transportation/ handling to the markets.

Total viable bacterial counts [TVBC], given as colony forming units per millilitre of milk [cfu/ml], was also assessed against the various parameters which included gross dirt in milk, colour of the milk sample, presence of bad odour, alcohol test, Resazurin test, isolation of *Escherichia coli, Klebsiella* species and *Enterobacter* species. Milk samples with gross dirt had a higher mean [25.24× 10^6] of TVBC than the negative samples [18.00× 10^6]. Milk samples having yellowish colour had a higher mean [31.70× 10^6] of TVBC than the samples with the white colour [22.65× 10^6]. Milk samples with bad odour had a higher mean [25.01× 10^6] of TVBC than the negative samples [23.11× 10^6]. Milk samples with a positive alcohol test had a higher mean [27.23× 10^6] of TVBC than the negative samples [21.57× 10^6]. Milk samples with an excellent quality in Resazurin test had the lowest mean [12.11× 10^6] of TVBC compared with samples with poor quality in Resazurin test [35.52× 10^6]. Milk samples with no *Escherichia coli* isolation had a lower mean [16.35× 10^6] of TVBC compared to the ones where sorbitol and non-sorbitol fermenting *Escherichia coli* were isolated [23.54× 10^6 and 26.24 × 10^6 respectively]. Milk samples with *Klebsiella/ Enterobacter* species isolation had a higher mean [23.63× 10^6] of TVBC compared to the samples where no *Klebsiella/Enterobacter* species were isolated [18.84× 10^6]

The above results show that there was a strong positive correlation between the parameters used here [i.e. positive gross dirt, positive abnormal colour, positive bad odour, positive

alcohol test, poor Resazurin test and isolation of *Escherichia coli, Klebsiella* species, and *Enterobacter* species] and the poor hygiene of camel milk. This phenomenon was also largely attributed to poor handling [poor sanitary practices] of camel milk during milking and subsequent transportation/handling to the markets.

Summary and assessment of camel milk hygiene/ bacteriological quality

Assessment of camel milk quality by bacteriological tests from the two counties[Garissa and Wajir; n = 384 samples] showed that Total Coliform Count [TCC] ranged between 130 × 10^4 - 190 × 10^6cfu/ml and Total Viable Bacterial Count [TVBC] ranged between 120 × 10^4 - 160 × 10^6cfu/ml. The total range of TCC and TVBC were divided into four categories each, for ease of interpretation and discussion.

The findings of TCCcompare well with those of analysis for Total bacterial count [TBC] done earlier in camel milk in Kenya which indicated a TBC of 10^3 – 10^5cfu/ml from transport containers, immediately after the end of milking [20]. The same study indicated a TBC of 10^2 – 10^4cfu/ml for camel milk from udders milked directly into clean containers. The latter results show that good quality raw camel milk is initially produced but it deteriorates rapidly as it enters the informal marketing chain. Pooling of different raw milk batches and usage of unhygienic plastic containers accelerate spoilage, with non-refrigerated bulk milk reaching a TBC of 10^8cfu/ml [20 Younan *et al.* 2002]. This milk turns sour in less than 24 hours when kept at 25ºC. Under hot pastoral conditions [35ºC], this can happen in less than 12 hours

Coliforms isolated from the collected milk samples included *Escherichia coli* [59.90% = 230 samples] and *Klebsiella/Enterobacter* species [95.83% = 368 samples], while *Enterobacteriaceae* were detected in all the 384 samples [100%]. The occurrence of total coliforms, in this study, was equivalent to that reported for Ethiopian raw camel milk [100%] by 25]. The existence of coliform bacteria may not necessarily indicate a direct faecal contamination of milk, but serves as an indicator for poor sanitary practices during milking and further handling processes. However, the presence of faecal coliforms, i.e. *Escherichia coli,* implies a risk that other enteric pathogens may be present in the sample. *Escherichia coli* O157:H7 was identified from one of the samples that were serotyped with *Escherichia coli* antisera O157 and H7. Having one sample yielding *E. coli* O157:H7 is significant especially considering the fact that one organism multiplies very fast. There is also a possibility that there may have been more samples carrying this organism, since serological testing could be rendered negative [false negative] by presence of some non-specified K antigens [26]. A sure way of determining if the organism has the ability to produce the vero-toxin is through usage of polymerase chain reaction [PCR], where a specific primer for the specific polypeptide chain/gene/plasmid/phage that codes for the toxin is used [27,28]. When infected with respective phage, *E.coli* serotype 0157:H7 and O157:non-motile produce one or more verocytotoxins[Shiga-toxins] and are the most frequently identified diarrheagenic *E.coli* serotypes in North America and Europe [29]. Shiga toxin-producing *Escherichia coli* [STEC], also known as enterohaemorrhagic *E. coli,* is one of the four categories of diarrheagenic *Escherichia coli* [30].Shiga toxin-producing *Escherichia coli* O157:H7 and other STEC serotypes cause human illness that can present as mild non-bloody diarrhoea, severe bloody diarrhoea [haemorrhagic colitis], and haemolytic-uremic syndrome [HUS] [31].Additional symptoms of *E. coli* O157:H7 infections include: abdominal cramps and lack of high fever. The organism O157 STEC readily colonizes dairy and beef cattle; thus, not surprisingly, ground beef has caused more O157 STEC outbreaks than any other vehicle of transmission [31]. Other known vehicles of transmission include raw milk, sausage, roast beef, unchlorinated municipal water, apple cider, raw vegetables and sprouts [alfalfa and radish] [31,32]. Isolation of the zoonotic serotype in camel milk means that camels could also be a source of infection for humans.

Other bacterial microorganisms isolated from the milk samples alongside the coliforms included: *Staphylococcus* species [90.10% = 346 samples], *Streptococcus* species [84.90% = 326 samples] and *Bacillus* species [45.83% = 176 samples]. Of the 346 *Staphylococcus* species isolated, 91 [23.70%] were coagulae-positive. Thus, the existence rate of *Staphylococcus aureus*, in the present study, was relatively high. However, the organism has been detected in all tested samples [n = 12] in Morrocan camel milk [33] Benkerroum *et al.,* 2003 [25] reported that *Staphylococcus aureus* isolates represent 15% of the total bacteria isolated from composite camel udder milk. The reported incidence of mastitis in camel herds [19.5%] and the high frequency of *Staphylococcus aureus* [31.5%] as the causative agent may explain these results [34]. According to the European Commission [EC] standards for raw cow's milk intended for direct consumption [European commission, 2001], 51% [n = 17] of the samples were found to have *Staphylococcusaureus* counts higher than the fixed acceptable limits [≤ 10^5cfu/ml]. An overview of the annual reports of food-borne diseases from seven countries indicated that milk and milk products were implicated in 1 to 5 % of the total bacterial outbreaks. *Staphylococcus aureus* was by far the most frequent pathogen associated with these outbreaks [85.5%], followed by *Salmonella* [10%] [35]; 45.83% [n = 384] of the collected camel milk samples were contaminated by psychrotrophic *Bacillus* species [*cereus*]. The results of psychrotrophs are comparable with the average reported for raw cow milk by [36] and [37]. No documentation on the content of psychrotrophs in camel milk was found in the literature. Psychrophilic bacteria are responsible for an increased production of proteinases and lipases, which can survive heat treatments [i.e. pasteurization] thus affecting the shelf life and quality of milk [38].

Resazurin test to determine the microbial load/quality of camel milk showed that a total of 278 samples [72.39%] were of good quality [124 samples [32.29%] were of excellent quality, 135 samples [35.15%] were of good quality, 19 samples [4.95%] were of fair quality], while a total of 106 samples [27.61%] were of poor quality [62 samples [16.15%] were of poor quality and 44 samples [11.46%] were of bad quality]. It was observed from another study, carried-out by the researchers, that pastoralists of Garissa and Wajir counties occasionally washed and smoked their milking vessels, but the personal hygiene of the milker was poor; this being due to lack of good hygiene awareness/practice, inaccessibility of soap/disinfectant, and insufficient clean water supply. This resulted in high contamination of milk after milking.

Camel milk possesses superior keeping quality compared to cow milk; a property that makes raw camel milk a marketable commodity even under conditions of high temperatures and very basic hygiene [38]. This is due to its high content of proteins that have inhibitory properties against bacteria.

In Somalia and Kenya, camel milk production areas are often located far from markets as observed by [39]. Distances to provincial markets range from 20 km to 90 km and may be up to 400 km for distant urban markets. During periods of milk surplus [rainy season] transport on dirt roads is unreliable resulting in breakdowns and delays in milk delivery. Storage in unhygienic containers [plastics and traditional gourds], mixing of evening and morning milk, pooling of milk from different suppliers, prolonged transport times, high environmental/ambient temperatures and road-side selling out in open containers, all increase contamination and spoilage of milk. This explains the high values of more than 10^7 cfu/ml of TCC and TVBC observed in most of the samples [>80%] in the present study. However spoilage does not always equal wastage. Sour milk is part of the traditional diet [Somali "Susa", Arabic "Al-Garss"] and sour milk of acceptable quality is sold and consumed comfortably by the pastoralist communities [39,40]. However growth of contaminants in raw camel milk poses a threat to consumer health when milk of poor hygiene is sold. Spoilage reduces the market value of the milk causing income losses to producers and vendors. Souring or sour camel milk is also unsuitable for heat treatment in dairy plants.

The common practice of smoking traditional milk containers and milking buckets [made from gourds] with natural fibres achieves high temperatures and appears to have a beneficial effect on the keeping quality of milk, although this has not yet been studied in detail.

However, the obvious advantage of plastic containers [cheap, light weight, durable, large volume per container better suited for transport in vehicles] coupled with the limited availability, high costs and small volumes of traditional containers leads to the increasing use of these plastic containers in the camel milk trade. Plastic jerricans of cheap quality [e.g. re-cycled cooking oil containers] have a fast corroding surface and are very difficult to clean in pastoral areas because of the lack of clean water. The non-availability of safe clean water also implies that the introduction of common hygiene recommendations will be difficult and adapting hygiene practices and guidelines to the pastoral situation remains a challenge [20].

Comparison of Garissa and Wajir counties, with respect to milk hygiene

Wajir County had more milk samples [27.27%] with a TCC of between 160 × 10^6 - 190 × 10^6cfu/ml than Garrissa County [17.83%]. This could be attributed to the long distance [and hence increased time] pastoralists in Wajir had to travel under hot pastoral conditions [35ºC] from the pastures to the watering wells where the milk is sold. This favoured the multiplication of microorganisms. The incidence of isolating *Escherichia coli* from the milk samples was higher [by 32.78%] in Garrissa County than in Wajir County. This could be attributed to increased use of unhygienic plastic containers, pooling of different milk batches, poor personal hygiene of milkers and handling of milk in unclean environment.

Garissa yielded more [30%] coagulase positive staphylococci than Wajir [14.3%], despite more staphylococci having been isolated from Wajir [93.5%] as compared to Garissa [87.8%]. Garissa also yielded more [31.3%] CAMP positive streptococci than Wajir [19.5%]; Streptococcus was isolated at same prevalence [85%] for the two areas. CAMP-positive streptococci are common causes of mastitis in cows [40] Buxton and Fraser, 1977].

CONCLUSION FOR THE STUDY

This study has shown that there is substantial contamination of milk produced in the two districts of North—Eastern province, Kenya. The contaminants include both physical substances and bacteria. Inadequate availability of water and ignorance on good/hygienic milking practices are the main causes of the contamination, making the milk dangerous to human health. There is, therefore, need for the Government to avail ample clean water to the areas and to undergo training sessions on good/hygienic milking, packaging and transporting practices.

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