ORIGINAL ARTICLE

ASSESSMENT OF THE BACTERIOLOGICAL QUALITY OF MILK AT DAIRY FARMS AND INDIVIDUAL BREEDERS IN JIMMA TOWN, SOUTH WEST ETHIOPIA

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ABSTRACT

BACKGROUND: ***Food-borne diseases are major public health concern worldwide. Many people around the world acquire food poisoning due to consumption of raw, contaminated milk which are spread either from infected cows, by handling or during milk processing. The aim of this study was to assess the sanitary standards of dairy farm as well as individual breeding areas in Jimma town, evaluate the hygienic practices of milk handlers and to determine the bacteriological quality of milk at dairy farms and individual breeders.***

METHODS: ***A cross-sectional study was conducted from January to July2001. In the study, survey using interview methods using structured questionnaire and close assessment concerning cleanliness and design of the barn, status of animal health, hygienic practices and health status of the milk handlers was made. In addition, bacteriological investigation on the quality of milk, sterility of the milk utensils and containers was made. Standard laboratory procedures including gram staining, culture and biochemical tests were employed to identify potential bacterial pathogens.***

RESULTS: ***Of all the milk samples tested for quality, only 52/60 (86.7%) of the milk samples tested were found to be free of any pathogenic microorganisms and acceptable for consumption while 9/13 (69.2%) of milk containers were bacteriologically unacceptable for utilization.*** **Staphylococcus aureus** ***was the commonest bacterial pathogen isolated from freshly drawn milk (in 13.9%) of cases while the 3 klebsiella species (K. pneumoniae, K. ozanae & K. oxytoca) were isolated from collection cans (bulk) and milking utensils (in 7 and 5 cases respectively). Some milk samples contained multiple pathogens. Polymicrobial isolation was observed in 2 cases of each in freshly drawn milk, milking utensils and milk from bulk cans. In addition, on environmental assessment, cows were not regularly checked for animal diseases.***

CONCLUSION: ***Consumption of raw milk from market is potentially dangerous for health. Thus, proper boiling of the milk is recommended. The farm managers and individual breeders must take corrective measures so as to produce milk for human consumption that meets the existing milk standard, which is free of harmful bacterial pathogens. The cows should be regularly checked at animal clinics***.

KEY WORDS*:* **Milk, milk handlers, bulk, bacteria, barn**.

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INTRODUCTION

Milk is a well balanced diet, nutritious and easily digested by the human body (1). It contains proteins, fats, lactose and inorganic substances.

Milk serve for bacterial growth. Food-borne diseases are major public health concern worldwide. Millions of people consume milk daily either raw or pasteurized (2). As a result many of them acquire different zoonotic diseases, which are spread by infected milk either from infected cows or during milk processing. Such diseases include tuberculosis, typhoid fever and salmonella food poisoning, dysentery, diphtheria, staphylococcal intoxications, gastroenteritis, brucellosis and Q-fever (6). Milk is usually contaminated by microorganisms at the site of production or during processing. Microorganisms in milk come from the cow, air, milk containers and the milk handlers. It should be protected against direct or indirect contact with any source of external contamination during all the steps of milking, collection and transport. The bacteria causing contamination may originate from the udder of the cow, milking equipment or after milking handling procedures. The actual magnitude and impact of food-borne diseases on health are not exactly known. Only a small proportion of cases are reported. According to WHO, it is believed that in developed countries only about 5-10% of cases are reported (5). In many developing countries including Ethiopia, reliable quantitative data are also lacking. The problem in Jimma dairy farm is not an exception since it is a rural setting without adequate introduction of modern facilities. The handling of milk is traditional which needs investigation.

The number of bacteria per milliliter of milk added from the various sources depends upon care taken to avoid contamination. Standard milk should not contain any pathogenic bacteria, including coliform bacilli in 0.001 ml (6).

In Ethiopia, raw milk, ice cream, cheese and other milk products are frequently consumed in the homes and cafeterias. Due to high probabilities of contamination by different pathogenic bacteria, many people suffer from milk-borne diseases. Morbidities due to diarrhoeal diseases are common, especially in children. The aim of this study was therefore to assess the sanitary standards of dairy farm as well as individual breeding areas in Jimma town, evaluate the hygienic practices of milk handlers and to determine the bacteriological quality of milk at dairy farms and individual breeders.

MATERIALS AND METHODS

This cross-sectional study was carried out in Jimma town from January to July 2001. The aim of the study was to assess the general sanitary conditions of milking areas in dairy farms as well as individual breeders and examine bacteriological quality of milk. Essential information on sanitary status of the barn, cows and individuals responsible for milking were collected using a questionnaire. In addition, the health status of cows and barn was assessed. For data collection, the selected study area was classified in 3 study sites, which represented the whole town where cattle were raised. Accordingly, data and milk samples were collected from the Ethiopian Dairy farm of Jimma branch, dairy farm of Jimma College of Agriculture and individual breeders living in Jimma town. First, a pilot survey was made in order to gather information on the locations of dairy farms and individual breeders who owned lactating cows. Then, breeders who owned more than 2 lactating cows at the study time were selected and included in the study. This was ideal for randomly

selecting of cows and to sample from bulk and distribution centers for commercial purposes. One hundred twenty one lactating cows were identified in the 3 study sites. A total of sixty cows, 20 from each of the 3 zones, were randomly selected by lottery system for the study.

Survey of environmental sanitation was conducted. In this respect, all workers in the dairy farm and individuals involved in milking as well as handling of the milk were interviewed by pre-tested questionnaire. The major variables included conditions of animal health, hygienic practices and the health status of milk handlers. Checklist was employed to study the behavioral aspect of the workers and the other indicators such as personal hygiene and the sanitary condition of the environment (4,9).

In addition to the epidemiological survey, bacteriological analyses of milk were conducted. Milk samples were aseptically collected directly from the teats of lactating cows, from bulk store, and from distribution site (immediately before distribution) using sterile sample bottle of 250 ml. Swab samples were also collected from milking can, storage can and other utensils that have intimate contact. The samples were transported to the microbiology laboratory using an icebox. Samples were cultured within 1 hour of collection in order to avoid bacterial multiplication. The collected milk samples were diluted in 1:10, 1:100, and 1:1000 in sterile bottles and mixed 25 times. These were inoculated on to yeast extract milk agar as recommended by Senior *et al* (6). A portion of the milk samples was directly inoculated onto the solid media of MacConkey, Shigella-Salmonella (SSA), mannitol salt agar (MSA) and Blood agar to isolate the possible pathogens. The plates were incubated aerobically at 37^0C for 24-72^0C hours. Similarly, 1ml of every milk sample was inoculated onto MacConkey broth and incubated at 37 ^{0}C for 72 hours to determine the growth of coliform bacteria. Biochemical tests including carbohydrate or sugar fermentation tests as well as coagulase tests were performed where necessary in the identification of the bacterial species.

Verbal consent was obtained from all responsible authorities of the farms and individual breeders prior to data collection.

Data were cleaned and percentages computed using simple descriptive statistical methods.

RESULTS

Of the 120 cows in the two dairy farms and individual breeders' houses in the town, 60 milk samples were collected and analyzed. Animal clinic was found in only 1dairy farm while 1 dairy farm and the individual breeders do not regularly make check up for the cows. In addition, all the 20 milk personnel did not make any regular medical check up (Table 1).

Table 1. Environmental survey, personal hygiene milk handlers and animal health, Jimma town, January - July 2001

Characteristics	Respondents	
	Number	Percent
Frequency of cleaning of the barn (n=20)		
Once a day	6	30.0
Twice a day	10	50.0
Every other day	4	20.0
Source of water for cleaning of barn & materials		
Pipe	19	95.0
Unprotected spring	1	5.0
Adequacy of water for washing		
Adequate	20	100.0
Not adequate	-	-
Animal Health		
Get animal health service	1	5.0
Do not get health service	19	95.0
Milk handlers (n = 20)		
Health problem in the last 6 month		
Yes	4	20.0
No	16	80.0

A total of 86 samples, 60 (69.8%) of freshly drawn cows' milk, 13 (15.1%) of swabs from milk utensils, 13 (15.1%) of milk collection cans/bulk from distribution centers, were collected and bacteriologically analyzed. The majority of the samples 64/86 (74.4%) were obtained from milk and containers from individual breeders while the remaining 22/86 (25.6%) were collected from 2 dairy farms in the town (Table 2).

Table 2. Sources and quantities of milk and swab bacteriologically analyzed, Jimma town, January - July 2001.

Source of sample	Milk from cows' teats No (%)	Milk from Bulk No (%)	Swab from Utensils No (%)	Total samples No (%)
JCA Dairy farm	8 (9.3)	1 (1.16)	1 (1.16)	10 (11.6)
Jimma Dairy Farm	8 (9.3)	2 (2.4)	2 (2.4)	12 (14.0)
Indiv Breeders	44 (51.2)	10 (11.6)	10 (11.6)	64 (74.4)
Total	60 (69.8)	13 (15.1)	13 (15.1)	86 (100.0)

From all the milk samples analyzed bacteriologically, pathogenic bacterial were isolated in 8/60 (13.3%) in pure pathogen isolation. Similarly, 9/13(69.2%) of the swabs 10/13 (76.9%) of the bulk harbored pathogenic bacteria (Table 3). Of these 13 different species isolated, *S. aureus* was the most frequently isolated pathogen in 6 of 13 (46.2%) followed by *E. coli* in 2 of 13 (15.4%).

Table 3. Evaluation of milk and milk collection vessels, in view of their acceptability, Jimma town, January - July 2001

Source of sample anlayzed	Acceptable No (%)	Unacceptable* No (%)	Total No (%)
Teat milk	52 (86.7)	8 (13.3)	60 (100.0)
Bulk	3 (23.1)	10 (76.9)	13 (100.0)
Milk utensil	4 (30.8)	9 (69.2)	13 (100.0)
Total	59 (68.6)	27 (31.4)	86 (100.0)

*Unfit (unacceptable) quality: If pathogenic bacteria isolated from milk or milk collection vessels or cans.

Thirteen different potential pathogenic bacterial species were isolated from the different samples tested. This is after exclusion of commensal bacteria (data not shown). These pathogenic bacterial species were isolated from the following specimens: directly from milk, milk containers and collection tank (bulk). Up to 4 different species were isolated from a single specimen. The over all positivity for bacteria isolation with microbiologically unacceptable qualities of milk and utensils was 31.4% (27/86). Bacterial species were isolated in mixed form with other pathogens in 12/86 (14.0%) of cases. Of these isolates, *S. aureus* was the commonest bacterial pathogen isolated from direct milk (in 6 cases) while the 3 Klebsiella species (*K. pneumoniae, K. ozanae & K. oxytoca*) were frequently isolated from collection cans (bulk) and milking utensils in 7 and 5 cases respectively (Table 4).

Some milk samples were contaminated with more than one pathogen. In freshly drawn teat milk, 2 samples were contaminated with more than one pathogen. In the same manner, mixed bacterial contaminations in milking utensils and bulk cans were identified in 3 and 6 occasions respectively (Table 5). In addition to the bacterial species indicated in table 5, other contaminants and resident bacteria of the skin which are some times considered as opportunistic pathogens like Coagulase negative staphylococcus (*Staphylococcus epidermidis* and *Staphylococcus hemolyticus*) were isolated both from milk and milk containers.

Table 4. Bacteriological analysis of sampled milk and utensils count and the frequency of isolation of bacterial species from specimens, Jimma town, January - July 2001

Bacterial species isolated	Milk from Teat N= 60	Milk from Bulk N=13	Swab from milking utensils N=13	Total Samples N= 86 No (%)
Staph. aureus	6	3	3	12 (13.9)
E. coli	2	1	-	3 (3.5)
Pseudomonas spp	1	-	2	3 (3.5)
Kleb. pneumoniae	-	2	2	4 (4.7)
Kleb. ozanae	-	1	1	2 (2.3)
Kleb. oxytoca	-	-	1	1 (1.2)
Ente. aerogenes	-	4	2	6 (7.0)
Citrobacter spp	-	1	-	1 (1.2)
Yersinia spp.	-	1	-	1 (1.2)
Serratia spp.	-	1	1	2 (2.3)
Proteus vulgaris	1	-	-	1 (1.2)
Streptococcus spp	-	-	1	1 (1.2)
Shigella spp	1	-	-	1 (1.2)
Total	11	14	13	38

Table 5. Polymicrobial isolation from milk and different milk containers, Jimma town, January - July 2001

Source and bacteria species isolated	Frequency of isolation
Teats milk	
S. aureus /E. coli	2
S.aureus/ Shigella spp	1
Swab milking utensils	
S. aureus / Enterobacter Spp	1
Klebsiella spp/ Strept spp	1
Klebsiella/ Pseudomonas spp	1
Bulk cans	
S. aureus/ Enter. aerogenes	2
Enterobacter/ Citrobacter spp	1
S. aureus /E. coli /Klebsiella ozanae/ Yersinia spp	1

DISCUSSION

Cow's milk may be contaminated from different sources and at different processes. Milk that is contaminated by animals, air-borne dust or droplets at the site of production and during processing presents a health hazard. It may be contaminated from the cow itself, from air/ dust, unclean milk containers and the milk handlers. Milk can be contaminated by microorganisms directly from the milk handlers who have direct or indirect contact with the milk especially if these persons are in the process of shedding pathogenic organisms. Pathogens and other organisms can gain access to milk as a result of the milk handlers activities such as coughing, sneezing, scratching and from body surfaces in contact with milk, particularly the fingers (7). Personal cleanliness of the milk handler is necessary particularly during milking process and distribution. In the present study, the majority of cows are kept in unclean barns. Milk handlers as well as the cows lack regular medical check ups (Table 2). The provisions of adequate facilities for the cleansing, disinfection and storage of utensils and milking equipment and the refrigeration of milk to a temperature of 38^0F are basic essentials (7,9). The milking areas must be clean and should be free from harmful microorganisms and chemicals (10).

The environmental factors such as the design and cleanliness of buildings and installations, the adequacy of the water supply, the manner in which wastes are disposed and the amount of dust in the immediate surroundings are important in contributing to the microbial contamination of surfaces (3). To keep the barn in good sanitary standard, the area should be well ventilated and there should be no crowding. Efficient artificial illumination must be provided for milking hours. To reduce the hazards of dust borne contamination cattle should not be fed and the barn should not be cleaned during or within l hours prior to milking (3). Fly breeding are associated with improper storage, handling and disposal of animal wastes. The primary control measures should be cleanliness and prevention of pollution at the source (9).

Milk containers, receptacles and equipment which have direct physical contact with milk are most likely to be a source of microbial growth and multiplication if they are not properly designed and maintained for this reason. Milk utensils and equipment are specially designed and made in order to mitigate conditions that are favorable for microbial growth and multiplication and to facilitate easy cleaning and sanitizing process.

The water used for cleaning purpose was obtained from tap water and unprotected spring. It must be provided in adequate quantities and must be physically clean, free from harmful microorganisms and of a suitable chemical quality and properly handled. According to WHO recommendation, the working figure desired for water is about 50-75 liters per head per day (3).

In the study, different species of bacteria were isolated from the milk as well as the milk collection utensils. *E. coli, Staphylococcus aureus, Klebsiella,* and *Yersinia species* were common organisms isolated from milk or milk containers (Table 4). Some of these bacterial pathogens were isolated singly or in combination with other pathogens (Table 5). The mixed bacterial isolation was higher in milk containers and bulk cans than in the freshly drawn milk. This shows that, besides contamination of milk by milk handlers, milk containers lack proper washing after use. In one study conducted in Kenya, about 7% of milk from farmers' cows was of poor quality compared with 11% of milk sampled from societies that processed farmers' milk (10).

This shows that quality even deteriorates in the hands of distributors to consumers. This may be attributed to longer distances from collection points to processing, poor ambient storage temperatures, poor cleaning techniques of milk tanks in the dairies etc.

The presence of coliform bacteria (*E. coli*, *Enterobacter spp*, *Citrobacter spp*, and others) in milk confirms the contamination of the source by human or other warm-blooded animals or both since *E. coli* is mostly abundant in human and animal feces (12). *Staphylococcus aureus* is another pathogen contaminating milk. The organism may come from the hands of the milk personnel. In the present study, *Staphylococcus aureus* was isolated in 13.9% of cases (Table 4), which is comparable to the study conducted in Kenya by Ombui *et al* (10). These investigators found that 7-10% of raw milk and some pasteurized milk products from Nairobi were contaminated with multi-drug resistant *Staphylococcus aureus.* In addition, coliforms such as *Enterobacter aerogenes, E. coli* and other bacteria such as different Klebsiella species and *Pseudomonas aeruginosa* were identified in considerable rate in the raw milk in this study. In this study no salmonella species were detected.

Colony counts and species identifications showed that non-pathogenic contaminant bacteria - which most likely come from the skin of the milk handler, were also isolated from the different samples. This shows that no care was taken to avoid contamination to the milk. Hands should be washed, rinsed, with effective bactericidal solution and dried immediately before milking, and should there be any interruption where by the hands may have become contaminated, such as handling a handkerchief for touching the floor or any unclean object, the disinfectant should again be used. The worker should never indulge in sneezing, coughing, spitting, or the use of tobacco while handling of milk. Since colds and other respiratory irritations are sometimes the first symptoms of serious diseases, it is advisable to exclude persons so affected from milking or other close proximity to the milk (4). The health of personnel should be checked regularly, personnel must be properly instructed and supervised at all times with respect to sanitary practice (3). In addition, the food handler must take all the precautions necessary to protect food and drink from the risk of contamination (7,9).

In developing countries such as Ethiopia, modern preservative methods such as ultra-high temperature (UHT), pasteurization, canning and aseptic packaging that are advanced processes which necessitate skillful operation and capital investment are only limited to large cities like Addis Ababa (3). The provisions of adequate facilities for the cleansing, dis-infection and storage of utensils and milking equipment and the refrigeration of milk are basic essentials (7).

In conclusion, the farm managers as well as individual breeders should take maximum precaution to produce and supply safe milk for human consumption that meets the existing milk standard, which is free of harmful bacterial pathogens. If care is not taken during milk processing so as to destroy all vegetative bacteria in the milk, resistant species like *S. aureus* may cause serious diseases, which may be difficult to treat. In addition, since farm animals are the main sources of drug resistant enteric bacteria, inspection of the animals is also important (13). Particular care should be taken to avoid the direct physical contact of milk with unclean surfaces such as milking utensils, udders, teats and the hands of milking man (3). In addition, the dairy farm or barn must be kept clean from dust and dirt during milking hours (4).

With respect to animal health, the dairy inspector should look for abnormal conditions of teats and udders, particularly inflammation and lumps and should recommend the services of a veterinarian. Preventive measures applied to the cows such as periodic examinations by veterinarian, segregation of infected animals, sanitation and good milking practices and good herd management are recommended. Consumers of milk are also advised to pasteurize or heat the milk prior to consumption. Products of milk such as ice cream should also be prepared from pasteurized milk.

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