MINATION OF BACTERIOLOGICAL AND PHYSICAL QUALITY OF RAW MILK AND LOCALLY TLED MILK IN CAFETERIAS OF JIMMA TOWN, ETHIOPIA

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RACT

**GROUND:** Food-borne diseases are major public health concern worldwide. People around the world acquire food poisoning due to consumption of raw, unheated milk which spread either from infected cows, by handling, or during milking. This study aims to examine the bacteriological quality and adulteration of milk and locally bottled milk in Cafeterias of Jimma Town.

**ODS:** Laboratory-based cross-sectional study was conducted in February 2004. Six Cafeterias were selected randomly. Hygienic practice and sanitary conditions of the cafeterias were assessed using observational checklist. Raw milk samples were collected from cafeterias' bulk storage after mixing with sterile plunger, using sterile cupped container with 250 ml capacity. One bottle of unopened locally bottled milk was selected randomly from the shelf and transported using an icebox to the mental health laboratory, Jimma University. Nutrient agar, Columbia agar and water were used to grow total microbes. Loury's sulphate tryptose broth with bent pad and membrane filter were used to determine coliforms. Lactometer and refractometer were used to measure the specific gravity and temperature of milk.

**RESULTS:** All the milk handlers have no health check up and certificate. Among the handlers 53.8% wash their hands before and after break and after visiting toilet, and only 30% practice hand washing after coughing and sneezing. 80.8% of raw milk were found in poor or grade C quality, while 19.2% were grade B. Among bottled milk samples 9% were grade B while 91% were grade C. Among various sources, majority of them are obtained from individual breeders (84.6%). Microbial count of raw milk was found that 73.1% were grade B - and 23.1% grade C. Of locally bottled milk were grade C. 65.4% of raw milk samples were adulterated water, most of them were collected from individual breeders. Adulteration was significantly associated with microbial count (\(\chi^2 = 5.787, P < 0.025\)) this indicates concentrated milk was found with high microbial count.

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CONCLUSION: Consumption of locally bottled milk is not advisable. Thus cafeteria owners should avoid preparation of locally bottled milk, unless proper boiling and bottling is practiced with a license of professional. Intensive health education should be given a good hygienic practice to the workers and owners. The concerned authorities should strengthen technical supervision and design strong rules for health check-up and certification of workers.

KEY WORDS: bacteriological quality, bottled milk, adulteration, and cafeterias.

INTRODUCTION

Milk is an ideal medium for growth of microorganisms. It is highly perishable food because it contains nutrients required for microbial growth, such as, protein (3.5%), fat (3.7%), carbohydrate (4.9%), minerals (0.75%) and water (87.2%). It is also an excellent culture media for many kinds of microorganisms, being high in moisture content, nearly neutral pH (6.5 – 6.6) and rich in microbial food; which can easily become a source for the spread of pathogens or a good medium for bacteria and easily adulterated with any color change (4).

For a number of decades, WHO has recognized the wide spread nature of food borne diseases and their impact on communities in both the developing and developed world. The annual incidence of some 1.5 billion episodes of diarrhea in children under 5 years of age and more than 3 million resultant deaths are an indication of the magnitude of the problem, as a significant proportion of the diarrheal disease cases are food borne in origin (5).

In recent years, a number of industrialized countries have even experienced a significant increase of food borne diseases. In several countries surveys have pointed out to an annual incidence of 5 – 10% of the population involved (5, 6). Even the United States has the safest supply of all nations, however, it is estimated 25 million food borne illness cause and 16,000 deaths occur each year. From this approximately 66% of all food borne patients are caused by bacterial pathogen (7) while in developing countries like Ethiopia, the reported food borne cases may be as low as 1%, because individual cases or small out breaks of food borne disease generally remain unnoticed or may not be reported (5, 6).

The problem in Ethiopian is not an exception. There was a study done on bacteriological quality of raw cows milk obtained from dairy farms and a milk collection center in and around Addis Ababa by Gedefay and Molla (8). This study showed that milk samples from collecting buckets in the dairy farms had an average of 1 x 10^4 colony forming units (CFU)/ml, those from storage container had an average of 1 x 10^5 CFU/ml and the count reached 1 x 10^6 cfu/ml up on arrival at the processing plant. The mean coliform counts ranged from 1.3 x 10^4 CFU/ml (storage container before cooling) to 7.1 x 10^5 CFU/ml can arrival at the processing plants. The hygienic quality of raw milk from the collection center was poor with a mean total bacterial count of 1.3 x 10^7 cfu/ml. Lack of knowledge about clean milk production, use of unclean milk handling equipment and lack of potable water for cleaning purposes was some of the factors which contributed to the poor hygiene quality of raw milk in the study farms (8).

An early study conducted by Asefani and Mogesie (9) on college of dairy farm in Awassa compared the microbial load of milk directly obtained from the under milk and fresh milk from utensils. This study revealed that aerobic mesophiles counts for under milk ranged between 103 – 104 cfu/ml, whereas, raw milk from collecting utensils had counts as high as 106 cfu/ml. Coliform counts in under milk was about 10 cfu/ml, while this count in fresh milk (raw) collected from utensils was as high as 105 cfu/ml indicating that major contamination occurred during the milk collection process.

In Ethiopia, raw milk, and milk products are frequently consumed in different establishments and residential home. But the hygienic status or quality of milk and the prevalence of milk related related breakages was not well assessed. Different studies were conducted on bacteriological quality of milk on dairy farms of Addis Ababa, Awassa and in Jimma (8-10). However no study was carried out at food and drinking establishments on bacteriological quality of foods that are consumed by the peoples for immediate consumption or stayed for longer time.

This study assessed the bacteriological quality of raw milk and locally bottled milk and adulteration of these products. It helps to give clue for the quality of milk served in the cafeterias and for other researchers to study the bacteriological quality of any food items in different food serving areas. In addition, it may initiate the local government as well as the country at large to give priority attention to modernize or alter the existing hygienic condition of food and drinking establishments, which are the potential sources of food borne diseases.

METHODS AND MATERIALS

Study Design: Laboratory based cross-sectional study was carried out to examine bacteriological quality and adulteration of raw milk and locally bottled milk in 26 cafeterias in February 2004. It was conducted in Oromiya regional state Jimma special zone, Jimma town, which is found 335 km from Addis Ababa, South west of Ethiopia. The study variables include; total colony count, coliform count, specific gravity of milk, source of milk, ownership of cafeterias, type of container used, educational level of workers, and hygienic practice of milk handlers.

Sampling Technique: 26 cafeterias were selected by applying the population proportionate selection formula from municipality documentation and stratified based on ownership status; institutional and private cafeterias, then study samples were selected using simple random sampling technique.

Milk sample: Milk sample was collected from the selected cafeterias.

1. Locally bottled milk – 1 bottle of unopened sample was selected randomly from the stack or shelf.
2. Raw milk – sample was taken from the bulk or tank in sufficient amount.

Raw milk samples from the bulk were collected after thoroughly mixed with a sterile plunger. Sample was collected below the surface with a sterile dipper and aseptically poured into a screw – capped container of a capacity 250 ml.

The samples were transported using icebox with a suitable temperature ranges (0 – 4°C) to environmental health laboratory, Jimma University and examined within 24 hours (11).

Data were collected using observational checklist. Data regarding laboratory results was collected on a daily basis at the end of each laboratory examination of milk.

Colony Count (plate count): Colony count is used for the determination of the quality of raw and pasteurized milk, it assesses the number of viable bacteria in milk.

A sample of 1ml milk was diluted with sterile distilled water in 1:10, 1:10^2, 1:10^3 and 1:10^6 ranges and these were inoculated to Nutrient agar and Colombia agar and then incubated at 35°C to 37°C for 48 hours or 72 hours.
Determination of milk quality was made based on standard maximum acceptable level of colony count and E. coli count based on most probable number result. 

**Specific Gravity:** Specific gravity of milk was measured using lactometers to determine adulteration of milk with water. Specific gravity is usually taken at 15.5°C of milk temperature. If possible correct to this temperature (13). This temperature of milk was maintained using refrigerator and evaporative food cooler. All data collected by observational checklist were checked for the completeness and fulfillment daily. The data were processed manually using scientific calculator and analysis was done using descriptive and analytical methods.

**RESULTS**

**Hygienic Practice:** The study revealed that there was no regular health checkup to milk handlers and no workers found with health certificate. Twenty two (84.6%) of the milk handlers wear protective cloth, 20 (76.9%) use outer garments, 2 (7.7%) use hair cover, and the rest 4 (15.4%) never use protective cloth.

Among the milk handlers 18 (69.2%) wear protective cloth always, 4 (15.4%) wear sometimes and another 4 (15.4%) do not use at all.

Regarding hand washing habit 10 (38.5%) wash their hands before and after work, 14 (53.8%) wash their hands before and after break and after visiting toilet, the remaining 2 (7.7%) practice hand washing after sneezing and coughing.

Twenty one (80.7%) of milk handlers keep their fingers clean and trimmed and 5 (19.3%) do not. All the studied cafeterias have access to pipe water, which is from the public distribution system of the town. Equipment-washing activities of cafetaria are done found that 4 (15.4%) use warm water, 20 (76.9%) use soap and water, while the remaining 2 (7.7%) use only cold water. Among the studied cafeterias, 23 (88.5%) use sink and the rest 3 (11.5%) use bowl to wash equipments.

The current condition of milk storage materials found that 15 (57.7%) clean, 9 (34.6%) stained and the rest 2 (7.7%) rusted.

**Milk:** From a total of 26 cafeterias studied, 22 (84.6%) rented milk from individual breeders, 3 (11.5%) use their own cattle and 1 (3.8%) obtains from market. The study findings indicated that all cafeterias sale raw milk. Among the cafeterias studied 11 (42.3%) were selling locally bottled milk, while 5 (19.2%) cafeterias were selling Yogurt and 1 (3.8%) cafeteria was selling powdered milk.

These cafeterias use different kinds of storages or raw milk collected from sources. Among the coffee containers used bucket were 12 (46.15%), Plastic were 10 (38.46%) while Tin cans, which used least, were 4 (15.38%). Methods of drawing milk from the bulk is by pouring (50%) and dipping (50%). Availability of functional refrigerators in cafeterias is shown in Figure 1.

**Laboratory Analysis:** Examination of microbial quality of raw milk and locally bottled milk were done on 26 raw milk and 11 locally bottled milk samples from 26 cafeterias of Jimma town.

Among the studied samples of raw milk colony count, no one was found in grade A standard, 5 (19.2%) were found with fair quality (grade B) and the rest of the majority 21 (80.8%) were in poor quality (grade C), based on American Public Health Service Standard (6).

Out of 11 locally bottled milk samples only 9% were found in grade B and the rest 10 (91%) were poor quality (grade C) (Table 1).

In addition all samples were analyzed for coliforms, of which 3.8% were found in good quality, which is less than 100 c.f.u/ml of milk, 19 (73.3%) were found in fair quality and the rest 6 (23.1%) were poor quality (Table 2).

Coliform test of locally bottled milk revealed that 2 (18.18%) of samples have good quality and the rest 9 (81.81%) have poor quality of milk, which have a colony count of greater than 10 c.f.u/ml of milk (Table 2).

**Table 1.** Microbial growth in standard plate count of milk sample in cafeterias of Jimma town, February 2004.

<table>
<thead>
<tr>
<th>Type of milk sample</th>
<th>Colony count (Cfu/ml of milk)</th>
<th>Number of cafeterias</th>
<th>Percent (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw milk</td>
<td>≤2x10</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>2x10²-1x10⁵</td>
<td>5</td>
<td>19.2</td>
</tr>
<tr>
<td></td>
<td>≥1x10⁶</td>
<td>21</td>
<td>80.8</td>
</tr>
<tr>
<td>Total</td>
<td>26</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Locally bottled Milk</td>
<td>≤3x10⁴</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td></td>
<td>3x10⁴-5x10⁶</td>
<td>1</td>
<td>9</td>
</tr>
<tr>
<td></td>
<td>≥5x10⁴</td>
<td>10</td>
<td>91</td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>100</td>
<td></td>
</tr>
</tbody>
</table>

**Table 2.** Coliform count in milk samples of cafeterias in Jimma town, February 2004.

<table>
<thead>
<tr>
<th>Type of milk Sample</th>
<th>colony count per/ml of milk</th>
<th>Number of cafeterias</th>
<th>Percent (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw milk</td>
<td>&lt; 100</td>
<td>1</td>
<td>3.8</td>
</tr>
<tr>
<td></td>
<td>101-2000</td>
<td>19</td>
<td>73.1</td>
</tr>
<tr>
<td></td>
<td>≥ 2000</td>
<td>6</td>
<td>23.1</td>
</tr>
<tr>
<td>Total</td>
<td>26</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Locally bottled Milk</td>
<td>&lt; 10</td>
<td>2</td>
<td>18.2</td>
</tr>
<tr>
<td></td>
<td>≥ 10</td>
<td>9</td>
<td>81.8</td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>100</td>
<td></td>
</tr>
</tbody>
</table>

**Score:**

- Colony count ≤ 2 x 10⁵ c.f.u/ml is Grade A, Good quality
- 2 x 10⁵ - 1 x 10⁶ c.f.u/ml is Grade B, Fair quality
- ≥ 1 x 10⁶ c.f.u/ml is Grade C, Poor quality

The same grading system is used for figures in coliform count table i.e.

- < 100 Grade A, good quality
- 101 - 2000 Grade B, fair quality
- ≥ 2000 Grade C, poor quality
The American public health service standard for milk and milk products (5, 6, 12).

**Specific gravity:** A total of 37 raw milk and locally bottled milk samples were collected from 26 cafeterias. Specific gravity of milk was measured using a lactometer to determine whether the milk is adulterated with water or not. Out of 26 raw milk samples, 17 (65.38%) were found adulterated and the rest 9 (34.6%) were found in the normal range of milk specific gravity (Table 3).

Out of 11 locally bottled samples, 5 (45.45%) were found adulterated and 6 (54.54%) were in the normal range.

Specific gravity less than 1.027 is an adulterated-normal range of milk specific gravity is between 1.027 and 1.035 (5, 6, 12).

As shown in Table 4, there is a statistically significant association between adulteration and colony count.

**Table 3.** Specific gravity of milk samples in cafeterias of Jimma town February, 2004

<table>
<thead>
<tr>
<th>Sample Milk</th>
<th>Specific gravity</th>
<th>Number of cafeterias</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw milk</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt; 1.027</td>
<td>17</td>
<td>65.4</td>
<td></td>
</tr>
<tr>
<td>1.027-1.035</td>
<td>9</td>
<td>34.6</td>
<td></td>
</tr>
<tr>
<td>&gt; 1.035</td>
<td></td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>26</td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Locally bottled Milk</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&lt; 1.027</td>
<td>5</td>
<td>45.5</td>
<td></td>
</tr>
<tr>
<td>1.027-1.035</td>
<td>6</td>
<td>54.5</td>
<td></td>
</tr>
<tr>
<td>&gt; 1.035</td>
<td></td>
<td>100</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>11</td>
<td>100</td>
<td></td>
</tr>
</tbody>
</table>

**DISCUSSION**

This study was made in cafeterias of Jimma town, and the quality of raw and locally bottled milk is unsatisfactory. According to this study, the workers who wear protective cloth were 84.62% and the rest 15.38% do not have protective cloth. This indicates a better awareness of food and drink establishment owners to provide protective cloth to their workers. Among the milk-handlers only 7.7% wash their hands after coughing and sneezing. This may affect the bacteriological quality of milk, because the quality of milk was found unsatisfactory. The study identified that there was no regular health check up to milk handlers, and no continuous follow up towards the quality of milk by the responsible authorities. Probably, this is the main contributing factor for milk quality deterioration. Most of the time milk is contaminated with external source at different processes by microorganisms directly from the milk handlers, who have direct or indirect contact with milk, especially if the milk handlers are in the process of shedding pathogenic organisms, during sneezing, coughing, scratching and from body surface in contact with milk (7). Personal cleanliness is necessary particularly during milking, processing and distribution.

According to Ehlers (13), when milk leaves the cow's udder it contains some bacteria, ordinarily, harmless, but if the animal is diseased some may be pathogens. The other bacteria enter the milk from the air, dust, the milkier hands and surface of milk storage container (3).

Similarly, from milk storage containers 42.3% were not clean. Probably the unhygienic condition may be the cause of poor quality of milk in cafeterias.

The bacteriological standards of dairy products established by United States public health service in 1978, indicated that, the plate count of grade A raw milk should be less than $2 \times 10^5$ c.f.u./ml, grade B is between $2 \times 10^4$ c.f.u./ml to $1 \times 10^5$ c.f.u./ml and grade C is greater or equal to $1 \times 10^5$ c.f.u./ml of milk (3, 14). Based on this standard, all samples of raw milk were not found in grade A, while 19.2% were grade B and almost all in grade C were found (18). Another study done by Godefay and Molla in and around Addis Ababa on storage containers was found 4 $\times 10^5$ c.f.u./ml (18). This indicates that milk handlers as well as milk storage materials, which have direct physical contacts, are most likely to be a source of contamination. So milk utensils and equipments should be designed and cleaned properly to avoid the favorable conditions for microbial growth.

Regarding the quality of locally bottled milk found that, a considerable number (i.e. 91%) was poor with a colony count of greater than or equal to $5 \times 10^4$ c.f.u./ml. This may be due to improper boiling and bottling procedures during preparation. Coliform test for dairy products are not intended to indicate fecal contamination, but to reflect over all dairy farm and distribution place sanitation. The analysis of the study showed that 3.8% of samples were in good quality (i.e. $< 100$ c.f.u./ml), 73.1% were fair and the rest 21.1% were poor quality. The study is almost similar to the finding of Godefay and Molla in and around Addis Ababa. Over all, it indicates that, coliform counts are much greater than the standard set by APHS (American Public Health Service Standard) (6). This may be due to poor hygienic practice of milk handlers, absence of health check up and usage of stained and unclean storage materials. In addition, milk source of most cafeterias are individual breeders, because 90.9% of milk
were found high in microbial count and 65.4% were adulterated. Whereas, milk collected from their own cattle were in good quality and normal range of specific gravity.

Almost all, 81.8% of locally bottled milk samples were poor quality i.e. > 10 c.f.u/ml, coliform. The presence of coliform indicates that, either improper boiling and bottling or contamination after processing by human or other warm-blooded animal or both has occurred. Probably, this contamination may be from milk handlers during preparation and storage of the milk after boiling.

From the study samples of raw milk 65.4% was adulterated (i.e specific gravity < 1.027) and the rest 34.6% was found in the normal range of specific gravity (i.e. between 1.027 and 1.035). Adulteration with water may be done intentionally for profit or the cafeteria owners may not be aware about the quality of milk rented from individual breeders. Because, 22 (84.6%) of the milk collected from individual breeders were found adulterated (65.4%).

Accordingly, the study indicated that there was a statistical significant association between adulteration and microbial count (x² – 5.787, P < 0.025). The high increment of colony count is due to adulteration with water. Whether the water is clean or not, and contaminated by milk handlers during adulteration may affect the quality. Whereas milk collected from their own cattle was found that all in normal range of specific gravity and grade B. microbial quality.

CONCLUSION AND RECOMMENDATION

Based on the American Public Health Service Standard accepted in many countries of the world, the microbial quality of milk in Jimma is unsatisfactory. Consumption of raw milk and locally bottled milk from cafeterias is not advisable because both are found, high in microbial count, coliform count and adulterated with water. Especially milk rented from individual breeders is more adulterated and poor in terms of microbial quality. Locally bottled milk is not guaranteed in its quality as a bottled and bottled product. Based on the findings of the study the following recommendations are forwarded.

- The concerned authorities, municipality and local health authorities need to strengthen technical supervision on cafeteria service and frequent inspection should be practiced.
- Cafeteria owners should to avoid preparation of locally bottled milk, unless properly boiled and bottled with a license and approval of qualified personal.
- Intensive health education should be given both to the owners and the workers about good hygienic practice and the effect of unhygienic condition.
- Ministry of health must establish strong rules for any public eating and drinking establishment workers to have periodical health check up.

ACKNOWLEDGEMENTS

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REFERENCES


