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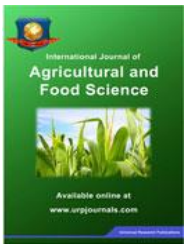

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Original Article

Microbiological Quality and the Impact of Hygienic Practices on the Raw Milk Obtained from the Small-scale Dairy Farmers in Sabah, Malaysia

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Abstract

The aim of this study was to investigate the raw milk hygiene and quality among the dairy farmers within Sabah area. A total of 150 raw milk samples were obtained from different dairy farmers as well as at the milk collecting center (MCC) of the Sebrang Station, Keningau. The results revealed that the bacteriology quality of raw milk was poor as the total plate count was more than 10^7 CFU/ml. Both the coliform (2.96 – 4.03 log CFU/ml) and *Staphylococcus* counts (2.73 – 3.55 log CFU/ml) were high in all tested samples. The microbial load of the raw milk increased ($p<0.05$) upon reaching MCC. A total of 47 samples were tested positive for the presence of *E.coli* while the *Staphylococcus aureus* was the second prevalence pathogenic bacteria (8.3-41.6%) found in this study. Only 17 raw milks were found positive with the presence of *Salmonella* spp, but none of the pathogenic species of *E.coli* O157:H7 were detected in this study. Stepwise tracer study revealed that personal hygiene of farmers and unhygienic utensils used during milking process contributed most to the bacteriology quality of raw milk. A significant reduction on microbial count was observed after the hygienic practices were introduced to the dairy farmers.

Key words: raw milk quality, hygiene, microbial count, dairy farmers, hygienic practices

Introduction

Milk is a food of good nutritional value which ensures benefits from its consumption. For families in Sabah especially the rural areas where milk represent a good source of protein, calcium and vitamin D that stimulate the growth and body functions [1]. Hence, milk will inevitably become an important part of people diet. The total milk consumption in Malaysia adds up to about 1000m liters per annum. In order to satisfy this demand, Malaysia produces about 56m liters of milk, which is equivalent to a self-sufficiency index of about 5%. However, the rest of the milk is imported, notably from Australia. The growth in demand for milk is driven by the changes in dietary preferences, convenience and also influenced by the growth of dairy value-added products such as cheeses and yoghurt.

Raw milk serves a good medium for microbial grow such as *Lactobacillus, Leuconostoc, Lactococcus, Streptococcus, Enterococcus*, yeast, moulds and *Enterobacteria*. In addition of that, the detection on foodborne pathogens such as *Listeria monocytogenes*, *Salmonella*, *Campylobacter*, *Staphylococcus aureus*, *Bacillus cereus*, *Clostridium botulinum* and *Shigella* have been reported by several authors in raw milk samples [2, 3]. Meanwhile, the detection on coliform bacteria or pathogens in milk can used as an indicator for udder infection (mastitis), contamination in milking utensils or water supply [4]. The high bacterial count as well as the present of pathogenic bacteria in milk not only degrades the milk quality and shelf-life of milk or milk-related products, but it also poses serious health threat to consumers. Hence, many dairy programmes have been carried out to improve the production as well as quality of raw milk [5]. This includes: keeping and storing the milk in clean containers at refrigerated temperature (4°C) immediately after milking process, control and modification on milking process or environment to eliminate cross contamination on raw milk harvested from milk utter.

Sabah, one of the important agriculture states of Malaysia is well recognized for its palm oil plantation and

also the livestock industry. The milk production in Sabah is more towards self-sufficiency level as both the Keningau and Tenom are the major milk production district with an annual production that exceeding 7m liters. Nevertheless, most of the small-scale dairy farmers still performing poor hygienic practices in milking process together with ineffective farm management, which in turn degrades the milk quality and causes great economic loses to the industry. Therefore, this study was done to investigate the raw milk hygiene and microbiological quality among the dairy farmers to as well as MCC and to strategize possible dairy hygiene practices that can be implemented to improve and upgrade the milk quality in Sabah, more specifically in Keningau and Tenom district.

Materials and Methods

Sample Collection

An approximately 100-300ml of raw milk samples were collected from individual dairy farmers located in Keningau (10 farmers) and Tenom (1 farmer). In addition, milk samples were also taken from the respective collector churns at Milk Collecting Center (MCC), Sebrang station, Keningau and transported back to Microbiology Research Laboratory of School of Food Science and Nutrition, Universiti Malaysia Sabah (UMS) at temperature 4-5°C. The microbiological analysis was performed immediately upon arrival to the laboratory.

Microbiological Analysis of Milk Samples

The respective milk samples were tested for total plate (TP), total coliform (TC), total faecal coliform (TFC) and total Staphylococcus (TS) count. A total of 25 ml samples were taken and homogenized with 225ml of sterile saline water prior 10-fold serial dilution was performed. The diluents (0.1ml) was transferred aseptically to specific culturing mediums, namely plate count agar (PCA) for TP count, violet RB agar for TC and TF count, Baird Pard agar (BPA) for TS count. The TP, TC and TS count was incubated at 35°C for 24-48hrs, whereas the TFC count was incubated at 42°C for 24-48hrs.

The milk samples were also detected for the presence of pathogenic organisms, namely *Listeria monocytogenes* [6], *E. coli* H7:O157 [7] and *Salmonella typhimurium* [8].

Application of Proper Hygienic Practices along Milking Process

Proper hygienic practices were recommended by the State Veterinary Department to the dairy farmers in order to see the effectiveness in improving the raw milk quality. These includes establishment of routine cleaning schedule on farms, the use of soap or sanitizer to clean the milker hands, providing the milker proper hair glove and coat during milking and also the use of clean wet towel to rub cow's udder and teats after every milking session. Hence, a few sampling sessions on the raw milk was performed after the hygienic practices were introduced to the dairy farmers.

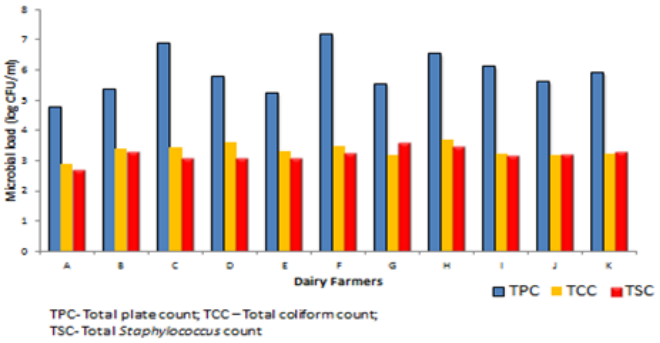

Figure 1a: Bacteriological quality of raw milk from different dairy farmers (n=150)

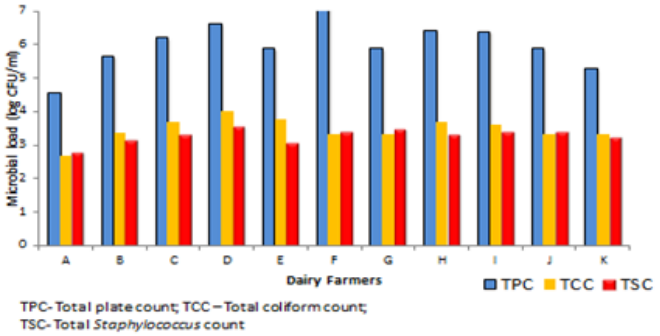

Figure 1b: Microbiological quality of raw milk collected at MCC level (n=150)

Statistical Analysis

The data were collected triplicately and the result was showed in the mean values and standard deviation form. Significant difference between the mean values was determined by analysis of variance (ANOVA) which applied a Duncan multiple tests. The statistical analysis was performed using SPSS version 17.0 (SPSS Inc. Chicago).

Results and Discussion

Microbial quality of raw milks obtained from farms and MCC level

A total of 150 raw milk samples were collected from individual dairy farmers as well as at MCC level in this study. The bacteriological quality for most raw milk samples from individual farmers was poor, with a total plate count exceeding 10^6 CFU/ml (Figure 1a). The microbial load in this study was even higher than those found in the study of Gran et al, [9] who mentioned that improper hygienic practices during milking process affect the microbiological quality of raw milk. Nevertheless, the highest TPC was recorded by farm F (7.33 log CFU/ml) due to poor hygiene farm practices during milking process. In addition, ineffective sanitizing routine on post milking process that leaving herds' manure in contact with cows' udder also contributed to the high bacterial level in raw milk. This eventually brings losses to farmers as the cow herds may be infected by mastitis disease. Meanwhile, it was observed that both the coliform (2.96- 4.03 log CFU/ml) and *Staphylococcus* counts (2.73 – 3.55 log

Table 1: Prevalence of pathogenic bacteria in raw milk obtained from different farmers

Farmers	Number of samples	Num. of positive sample (%)			
		E-coli	*Staphylococcus aureus*	*Salmonella* spp.	*Listeria* spp.
A	18	2(11.1)*	2(11.1)	2(11.1)	1(5.6)
B	12	3(25.0)	1(8.3)	1(8.3)	1(8.3)
C	12	4(33.3)	2(16.7)	2(16.7)	1(8.3)
D	12	5(41.6)	2(16.7)	3(25.0)	2(16.7)
E	12	4(33.3)	4(33.3)	1(8.3)	1(8.3)
F	12	5(41.6)	4(33.3)	1(8.3)	1(8.3)
G	12	4(33.3)	5(41.6)	1(8.3)	-
H	12	4(33.3)	2(16.7)	1(8.3)	-
I	12	2(16.7)	5(41.6)	1(8.3)	2(16.7)
J	12	3(25.0)	5(41.6)	2(16.7)	2(16.7)
K	12	6(50.0)	4(33.3)	1(8.3)	2(16.7)
L	12	5(41.6)	4(33.3)	1(8.3)	-
Total	**150**	47(31.3)	40(26.7)	17(11.3)	13(8.7)

*The figures in the bracket represent the percentage of samples tested positive for pathogenic bacteria
- notdetected

Table 2: Effect of hygienic practices on the microbiological quality of raw milk

Sample	After hygienic practices		
	TPC	**TCC**	**TSC**
A	3.88±0.21 (18.49%)*	2.37±0.10 (18.27%)	1.98±0.08 (26.67%)
B	4.75±0.11 (11.38%)	2.23±0.08 (34.02%)	2.27±0.15 (31.41%)
C	6.47±0.05 (6.09%)	2.56±0.17 (25.79%)	2.08±0.05 (33.97%)
D	5.28±0.13 (8.65%)	2.50±0.05 (30.75%)	2.16±0.12 (30.32%)
E	4.83±0.25 (7.47%)	2.54±0.12 (23.49%)	2.02±0.07 (35.26%)
F	5.72±0.07 (20.11%)	2.62±0.15 (25.14%)	2.15±0.13 (33.85%)
G	4.77±0.15 (13.74%)	2.28±0.02 (28.30%)	2.06±0.19 (42.78%)
H	5.59±0.28 (14.26%)	2.12±0.18 (42.55%)	2.07±0.16 (40.0%)
I	5.27±0.14 (13.61%)	2.26±0.21 (30.03%)	2.04±0.21 (35.65%)
J	4.75±0.17 (15.18%)	2.13±0.14 (33.02%)	2.09±0.13 (34.89%)
K	5.08±0.19 (13.89%)	2.19±0.22 (32.19%)	2.01±0.18 (33.67%)

TPC – total plat count; TCC – total coliform count; TSC – total Staphylococcus count
*Figures in the bracket indicate the percentage of reduction after hygienic practices were conducted

4.03 log CFU/ml) and *Staphylococcus* counts (2.73 – 3.55 log CFU/ml) were high in all tested samples. The high number of *Staphylococcus* and coliform counts indirectly revealed the farms were not properly manage due to poor personal hygiene practices throughout the milking process. Furthermore, the contaminated utensils especially the milking churns, teats cup or any utensils that intact with raw milk might influence the microbial quality of raw milk along the chain.

As a common practice in dairy management, the milk will be send to MCC immediately after milking processto maintain its quality. Hence, it would be of our interest to investigate the microbiological quality of raw milk during transportation to MCC. Majority of the samples (farm D, E, F, G, I and J) exhibited higher TPC count at MCC level as compared to the TPC count at the farm level (Figure 1a and 1b). For instance, the TPC count obtained from sample G was 5.08 ± 0.05, while the count increased to 5.96 ± 0.12 log CFU/ml ($p<0.05$) at the MCC level. The current result clearly indicated that the delayed in sending the raw milk to MCC together with the inefficiency in bringing down the raw milk temperature (4-5°C) could be detrimental to the raw milk quality, which allow the speedy proliferation of bacterial load within short period of time. Furthermore, improper storing of raw milk using unhygienic containers or utensils at farm level prior send to MCC also contributed to this situation. The current result was in accordance to the finding of Millogo et al, [10] that the milk is easily contaminated after milking and during transportation to the milk collecting center or markets due to external contamination or temperature abuse was taken place. In addition, International Dairy Federation (IDF, 1990) mentioned that the violation of any temperature will result in

the increase of microorganisms in the milk [11].

Detection of pathogenic organisms

Fresh milk normally contains low level of bacterial load as well as presence of certain pathogenic bacteria. Nevertheless, the presence of some indicator organisms such as *E. coli* and coliform bacteria indicates contamination occurs during the milk supply chain. Most importantly, the presence of foodborne pathogens also indicates an emerging threat to the public if raw milk is consumed without further treatment. A total of 47 samples (out of 150) was tested positive for the presence of *E.coli* among the milk samples (Table 1). Samples with highest prevalence of *E. coli* were obtained from farmer K (50.0%), while samples from dairy farmer A recorded the lowest of *E. coli* prevalence (11.1%). Their presence in milk normally associated with faecal contamination of water sources or poor hygiene practices during milking process. However, none of the samples were detected for *E. Coli* O157:H7.

Another pathogen that frequently degrades the raw milk quality is the *Staphylococcus aureus*. It was observed that the *Staphylococcus aureus* was the second prevalence pathogenic bacteria that detected in this study. They were found in the range of 8.3-41.6% among the tested samples and this phenomenon might due to improper milking practices among the dairy farmers. The *Staphylococcus aureus* was originated from infected cattle with mastitis problem or improper handling practices. According to Rysanek et al, [12] mastitis was usually caused by contagious pathogens namely *Staphylococcus aureus* and *Streptococcus agalactiae* than mastitis caused by environmental pathogens such as *Streptococcus uberis* and *Escherichia coli*. Hence, an effective post-milking practices including proper disinfection on cattle's teat or workers' hand are imperative to curb this problem.

Only 17 (of 150 samples) raw milks were found positive to the presence of *Salmonella* spp. Nevertheless, samples from dairy farmer D seem to have highest rate of detection among tested samples. Till date, the occurrence of *Salmonellosis* in local raw milk product was still low, however they still pose a health threatening diseases especially typhoid fever to the consumers who consumed any milk product that might contains *Salmonella* spp. The present of *Salmonella* spp in raw milk was mostly due to the post-processing contamination starting from the milking system to the supply chain. Most of the processors use heat treatment such as pasteurization to eliminate the heat sensitive *Salmonella* spp that may found in raw milk.

Impact of Hygiene Practices on Raw Milk Quality

Our stepwise tracer study revealed that the sources of contamination along the milking chain mostly due to unhygienic milking environment, contaminated milking equipment (milking machine, teat cups, filter cloth, milking churns), personnel hygiene during milking process and the abuses of temperature on raw milk at farm site prior send them to MCC. Hence, some basic hygiene practices were introduced in order to improve the raw milk quality among the dairy farmers. It was observed that the microbial load of the raw milk was reduced significantly after the hygienic practices were implemented among the dairy farmers (Table 2). The hygienic practices had most impact against farm F, as the TPC of the raw milk from farm F was log 5.72 ± 0.07 CFU/ml, which equivalent to a total of 20.11% of reduction on microbial count compared to its previous result (before the implementation of hygienic practices). Meanwhile, the reduction for the TCC (18.27 – 42.55%) and TSC (26.67-42.78%) among all tested samples was noticeable after the hygienic practices were implemented. The current result was supported by Mohamed and Fatima [13] that the application of hygienic practices on farm site managed to reduce the bacterial load of raw milk along the milking chain.

Conclusion

The current results showed that the microbiological quality of raw milk obtained from local dairy farmers was very low. High bacterial loads, the presence of several pathogenic bacteria in several samples not only affect the raw milk quality but definitely pose a safety issue to consumer. The introduction of proper training and hygiene practices during milking or post-milking process to the dairy farmers was found to be efficient in reducing the bacterial load or contamination of the raw milk. Hence, the finding or the recommendation on proper hygienic practices from this study will become a useful parameter and model for other dairy farmers within the state to improve and upgrade their dairy production.

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