Asian Australas. J. Biosci. Biotechnol. 2016, 1 (2), 199-205

Asian-Australasian Journal of Bioscience and Biotechnology
ISSN 2414-1283 (Print) 2414-6293 (Online)
www.ebupress.com/journal/aajbb

Article

Keeping quality of Red Chittagong Cows' milk obtained under different hygienic conditions

Md. Ashraful Islam[1*], Md. Nurul Islam[2] and M. A. Samad Khan[2]

[1]Department of Dairy Science, Patuakhali Science and Technology University, Babugonj, Barisal-8210, Bangladesh
[2]Department of Dairy Science, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh

*Corresponding author: Md. Ashraful Islam, Department of Dairy Science, Faculty of Animal Science and Veterinary Medicine, Patuakhali Science and Technology University, Babugonj, Barisal-8210, Bangladesh.
E-mail: sohel238@pstu.ac.bd

Received: 04 August 2016/Accepted: 16 August 2016/ Published: 31 August 2016

Abstract: To determine the effect of milking hygiene on keeping quality of milk at room temperature, 15 "Red Chittagong" milking cows were randomly and equally distributed under three different milking hygiene conditions (just previous to milking) such as T_1=Washing and disinfection of udder and teats, milkers' hands, and a sanitary rinse of milking pails with home-made calcium hypochlorite solution (200ppm Cl); T_2=Washing of udder and teats, milkers' hands, and milking pails with normal farm water; and T_3=dry milking without any preparation of cows' udder and teats , milkers' hands and milking pails. Microbial study revealed an average Standard Plate Count (million cfu/ml) respectively for T_1, T_2, and T_3 of 0.057±0.014, 0.708±0.8 and 0.161±0.037 at 0 hr; 0.263±0.080, 2.548±0.780 and 0.746±0.170 at 4 hr; 1.820±0.370, 18.711±5.050 and 5.578±1.840 at 8 hr; and 20.478±4.050, 25.578 ±2.200 and 22.367±3.400 at COB+ve. Average coliform count (thousand cfu/ml) were 0.038±0.024, 0.359±0.092 and 0.123±0.022 at 0 hr; 0.166 ± 0.100, 1.754±0.450 and 0.510±0.110 at 4 hr; 0.990±0.500, 12.810±4.010 and 3.350±0.850 at 8 hr; and 20.456±8.610, 25±5.440 and 24.440±5.460 at COB+ve respectively for T_1, T_2, and T_3. Microbial counts up to 8hr were significantly ($p<0.01$) different among treatments. The methylene Blue Reduction Time were significantly ($p<0.01$) different at different holding and a reduction time of 5.5 hr or more (>330 min.) was retained for at least 6 hr by milk of T_1, for only 2 hr by T_2 milk and for 4 hr by T_3 milk. There were non-significant differences in MBRT at COB+ve. Average acidity percentage were 0.1584, 0.1622 and 0.1611 at 0 hr; 0.1667, 0.1778 and 0.1683 at 4 hr; 0.1800, 0.2103, and 0.1846 at 8 hr; and 0.240, 0.258 and 0.240 at COB+ve respectively for T_1, T_2 and T_3. There were no significant differences in acidity percentage initially and at COB+ve. Average COB+ve time were 12.39, 8.83 and 11.94 hr, and normal smell of milk retained on an average up to10.83 , 6.22 and 8.83 hr, respectively for T_1, T_2 and T_3. Both the COB+ve time and normal smell hr of T_1 milk were significantly higher ($p<0.01$). Keeping quality time calculated by averaging the results of COB+ve time and evaluation of milk smell (hr) were on an average 11.67hr, 8hr and 10.08 hr respectively for T_1, T_2 and T_3. Milk produced with better milking hygiene showed longer keeping quality time.

Keywords: keeping quality; milking hygiene; Red Chittagong Cattle; milk

1. Introduction

Milk is considered as nearly complete food among the naturally occurring single article of diets. Being a nutritious food for humans, it also provides an ideal environment for microbial growth. Microorganisms which may gain entry into milk can multiply and bring about spoilage of milk and milk products and render them unsafe due to potential health hazards (Sharma-Varsha *et al*., 2005). Whenever we think about milk as a nutritious food for human being, we should assure legal standards for nutritional constituents, microbiological

quality and keeping quality Thus, production of "clean milk" which contains only small amount of hazardous bacteria and which is capable of remaining fresh for a considerable period is of extreme importance.

Although spoilage type of microorganisms present in milk are not considered harmful to human health and even some microorganisms have recognized therapeutic effects, these are harmful to milk itself, as they adversely affects the keeping quality (self life) of milk by bringing about a physical (curdling, roppiness, colour production etc.) and chemical (production of enzymes, pigments, toxins, decomposition products of fats, carbohydrates and proteins, etc.) changes in milk. From a technical point of view contamination of milk with undesired microorganisms may cause difficulty in the manufacture of desired dairy products also.

Since the numbers and types of organisms in milk usually get increased either by contamination or by growth of organisms already present. It is therefore generally recognized that necessary steps should be followed in milk production to prevent the entrance and subsequent growth of microorganisms as far as possible. In modern systems, milk is drawn nearly aseptically by machine and contains significantly lower initial microbial load, the subsequent growth of microbes are minimized by prompt cooling. But both the facilities are not available and practically applicable in Bangladesh's context. Milk production in our country is characterized by low yield non-descript cows (major portion) and buffaloes (small amount), small producers with little or no holding, use of crop residues and natural herbage with or without costly concentrates as feed supplement, scarce land for pastures and forage production. Milk is produced in small lots in remote rural areas and interval between production and consumption of milk is about 4 – 5 hrs or more. The rural producers and a few small or medium dairies produce milk without maintaining proper hygiene measure, which results in milk with higher initial microbial load, and the time interval between production and consumption favors a great increase in the microbial number and make the milk bacteriologically of poor quality.

In addition to the above conditions, due to the non enforcement of milk inspection act and standards relating to the hygienic quality of milk, the consumers of this country are deprived of getting quality milk and this valuable diet is unable to retain its keeping, organoleptic and safety qualities.

Rresults obtained by different researchers has revealed that proper milking hygiene could significantly reduce the microbial load in milk (Islam *et al*., 2009; Petrovic, *et al.*, 2006; Sandu and Man, 2006) and that lower microbial load is associated with better keeping quality (Lakhani and Singh, 1998; Koshy and Padmanaban, 1990).

Red Chittangong is the only recognized type of cattle in Bangladesh**.** Attempts have been taken to improve this type and to establish it as a potential dairy animal in our country contexts. In the present study, we collected milk from *Red Chittagong* cows subjected three different hygienic conditions before milking to compare the effect of different milking hygiene conditions on overall keeping quality of milk.

2. Materials and Methods

This study was conducted at Dairy Farm and Dairy Microbiology Laboratory of the Department of Dairy Science, Bangladesh Agricultural University, Mymensingh. Three different hygienic measures before milking were studied on *Red Chittangong* milking cows and milk samples from each treatment group were analyzed to assess the keeping qualities at different holding times.

2.1. Selection of animals

Fifteen (15) milking cows were selected from the elite herd of *Red Chittangong Cattle* kept at Bangladesh Agricultura University Dairy Farm. Cows were of similar yield characteristics and randomly and equally divided into three treatment groups containing five (5) cows in each.

2.2. Treatment application

All the milking cows were regularly groomed and washed to keep them clean, and similar floor hygiene and managemental conditions were provided. Udder and teats of 5 cows were washed with 200 ppm chlorine solution (previously made) , milking pails and milkers' hands were also sanitized with 200ppm chlorine solution just previous to milking and considered as treatment group-I (T_1), in the treatment group-II (T_2) udder and teats of the cows, milkers' hands and milking pails were washed with normal clean water, and in the group-III (T_3) cows were milked dry without any preparation of cows' udder and teats , milkers' hands and milking pails.

2.3. Preparation of calcium hypochlorite solution

According to the procedure described by Islam *et al*., 2009.

2.4. Milking procedure and sample collection

Milking was done by hand and in open pails. Milks from each treatment group were collected in three different buckets. Mixed milk samples of 1 liter from each group was taken into three sterile containers and kept at normal temperature. Milk samples were brought to laboratory for analysis immediately after milking. Sample bottles were marked accordingly T_1, T_2, and T_3 before taking milk samples of respective group.

2.5. Parameters studied

The milk samples were analyzed for keeping quality with the help of Standard Plate Count (SPC), Coliform Count (CC), Methylene Blue Reduction Time (MBRT) test, Acidity test and Clot on Boiling (COB) test at the beginning (0 hour) and subsequently at different holding periods. SPC and CC were performed at 4^{th} and 8^{th} hour of holding and at the time of COB+ve. MBRT test and Acidity test were performed at interval of 2 hours till production of COB+ve. COB test was performed at interval of 30 minutes until the milk samples have shown positive results. The smell of milks obtained under different hygienic conditions was observed at 30 minutes interval.

2.6. Statistical analysis

The statistical analyses were done by plotting data in Completely Randomized Design (CRD). Statistical package MSTAT-C was used for analytical purpose. LSD test was carried out to find out the significant difference among different treatment means.

3. Results and Discussion

To work out overall keeping quality of milk Standard Plate Count and Coliform Count were performed at 4 hr interval up to COB+ve; MBRT-test and acidity test were performed at 2 hr interval; and Smell test and COB test were performed at 30 minutes interval.

3.1. Standard Plate Count (SPC)

Standard Plate Count (SPC) million/ml as obtained from different hygienic groups (T_1, T_2 and T_3) at the beginning (0 hr) and subsequently at interval of 4 hr till production of COB+ve are presented in Table 1. Average SPC (million/ml) were 0.057±0.014, 0.708±0.070 and 0.161±0.037 at 0 hr; 0.263±0.08, 2.548±0.78 and 0.746± 0.17 at 4 hr; 1.842±0.37, 18.711±5.05 and 5.578±1.84 at 8 hr; and 20.478±4.05 (15.2-27.3), 25.578±2.20 (21.0-28.8) and 22.367±3.4 at COB+ve, respectively for T_1, T_2 and T_3. The mean SPC values at 0 hr, 4 hr and 8 hr were significantly different ($p<0.01$) and the differences between mean values at COB+ve were non-significant at 1% level but were significant at 5% level. Similar results have been reported from Lakhani and Singh (1998).

Table 1. Standard Plate Counts (SPC) as obtained from milk of different hygienic groups at the beginning and subsequently at intervals of 4 hr till production of COB+ve.

Observation at	Treatment Group	SPC(Mean±SD) $\times 10^6$cfu/ml	Degrees of freedom	Level of Significance
0 hr	T_1	$0.057^c \pm 0.014$	26	$p < 0.01$
	T_2	$0.708^a \pm 0.070$		
	T_3	$0.161^b \pm 0.037$		
4 hr	T_1	$0.263^c \pm 0.080$	26	$p < 0.01$
	T_2	$2.548^a \pm 0.780$		
	T_3	$0.746^b \pm 0.170$		
8 hr	T_1	$1.820^c \pm 0.370$	26	$p < 0.01$
	T_2	$18.711^a \pm 5.050$		
	T_3	$5.578^b \pm 1.840$		
COB+ve	T_1	$20.478^b \pm 4.050$	26	$p < 0.05$
	T_2	$25.578^a \pm 2.200$		
	T_3	$22.367^b \pm 3.400$		

$^{a, b,c}$ = values in the same column at each holding with different superscripts differ significantly

Milk of T_2 and T_3 become poor at 8 hr while T_1 milk remains fair and if the growth continues in the same trend then the T_1 milk will remain fair for at least 9 hr as per IS 1479 (Part III), 1969.

3.2. Coliform Count (CC)

Table 2 represents the results of coliform count (CC) thousand per ml for milk of different hygienic groups (T_1, T_2 and T_3) at the beginning and subsequently at intervals of 4 hr till production of COB+ve. Average CC ($x10^3$/ml) were 0.038±0.024, 0.359±0.092 and 0.123±0.022 at 0 hr; 0.166±0.100, 1.754±0.45 and 0.51±0.11 at 4 hr; 0.99±0.5, 12.81±4.01 and 3.35± 0.85 at 8 hr; and 20.456±8.61, 25±5.44 and 24.44 ±5.46 at COB+ve, respectively for T_1, T_2 and T_3.

Table 2. Coliform Counts (CC) as obtained from milk of different hygienic groups at the beginning and subsequently at intervals of 4 hr till production of COB+ve.

Observation at	Treatment Group	CC(Mean±SD) $\times 10^3$ cfu/ml	Degrees of freedom	Level of Significance
0 hr	T_1	$0.038^c \pm 0.024$	26	$p < 0.01$
	T_2	$0.359^a \pm 0.092$		
	T_3	$0.123^b \pm 0.022$		
4 hr	T_1	$0.166^c \pm 0.100$	26	$p < 0.01$
	T_2	$1.754^a \pm 0.450$		
	T_3	$0.510^b \pm 0.110$		
8 hr	T_1	$0.990^c \pm 0.500$	26	$p < 0.01$
	T_2	$12.810^a \pm 4.010$		
	T_3	$3.350^b \pm 0.850$		
COB+ve	T_1	20.456 ± 8.610	26	Non significant
	T_2	25.000 ± 5.440		
	T_3	24.440 ± 5.460		

$^{a, b,c}$ = values in the same column at each holding with different superscripts differ significantly

Although initial coliform count(0 hr) and coliform count at 4 hr and 8 hr were different but there were no significant differences in the coliform count under T_1, T_2 and T_3 groups at the time of COB+ve. The mean CC shows that a standard of 1000 cfu/ml could be maintained for T_1 up to at least 8 hr, 4 hr for T_2 and < 8 hr for T_3. An overall slower growth was observed between 0 - 4 hr periods, which may be due to initial germicidal action of organisms. But growth of organisms in T_2 was higher than T_1 and T_3 at 0-4hr interval and continued up to 8hr, which indicates that contaminating organisms in T_2 group showed less lag phase but active growth during the period. This observation suggests that higher coliform contamination can overcome the germicidal action of milk faster and show an active growth from the beginning. Walstra *et al.* (1999) stated that contaminating organisms originating from milking equipment have no lag phase.

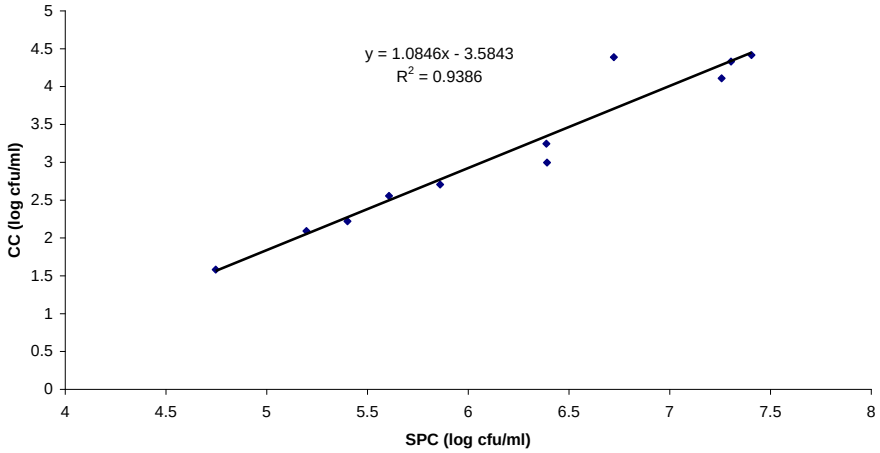

Figure 1. Correlation between log SPC and CC.

The overall relationship between SPC and CC is presented in Figure 1. Relationship between average log SPC and CC at the beginning up to COB+ve was highly significant (r=0.96), indicating that higher SPC correlates higher CC at any level of holding milk samples.

3.3. Methylene blue reduction time (MBRT)

To get an idea about the overall change over time in microbial population of milk obtained under T_1, T_2 and T_3; MBRT tests were performed at intervals of 2hr. The results of MBRT tests are presented in Table 3. Results show that there was gradual reduction in MBR time with the progress of storage time. It is evident from the table that a reduction time of 5.5 hr or more (>330 min.) can be retained for at least 6hr by milk of T_1, for only 2hr by T_2 milk and for 4hr by T_3 milk. Thus, milk of T_1 group kept its quality good for longer period than T_2 and T_3 according to MBR time. An overall lower MBR time was recorded for T_2 and an increasing rate of decrease were found for T_1 and T_2 respectively after 8hr and 6 hr.

For generalized discussion and comparing MBRT values with microbial counts, results at 4 hr interval were analyzed. Average MBR time (minute) respectively for T_1, T_2 and T_3 were 693±23, 513±85 and 650±45 at 0hr; 527±97, 300±85 and 447±98 at 4 hr; 300±79, 71±38 and 173±42 at 8 hr; 21±05, 17±04 and 17±05 at COB+ve. The average MBR time were significantly different ($p<0.01$) and were in the order of $T_1=T_3>T_2$ and $T_1>T_3>T_2$ respectively at 4 hr and 8 hr. MBR time at COB+ve showed non-significant difference, although there were significant differences at the beginning and at subsequent holdings.

Table 3. Methylene blue reduction time (minutes) at 2hr interval up to COB+ve.

Treatment	Observed MBRT in minutes (Mean±SD) at							
	0 hr	**2 hr**	**4 hr**	**6 hr**	**8 hr**	**10 hr**	**12 hr**	**COB+ve**
T_1	693^{a}±23	647±34	527^{a}±97	432±78	300^{a}±79	143±68	62±39	21±05
T_2	513^{b}±85	410±69	300^{c}±85	180±65	71^{c}±38	-	-	17±04
T_3	650^{a}±45	590±62	447^{b}±98	297±80	173^{b}±42	83±25	-	17±05
Level of Significance	$p < 0.01$	-	$p < 0.01$	-	$p < 0.01$	-	-	Non-significant

$^{a, b,c}$ = values in the same column with different superscripts differ significantly

3.4. Acidity test

To observe the overall pattern of increase in acidity samples were taken at 2 hr interval and per cent titrable acidity were recorded. Results obtained at 2 hr interval under different hygienic groups (T_1, T_2 and T_3) are presented in Table 4. Very little increases in acidity were found in the milks obtained from T_1, and T_3 during first 2 hr. In contrast to this, milk obtained under T_2 showed a bit higher increase from the very beginning. Results of 2 hr interval show that acidity level below 0.2 per cent could be maintained up to 10 hr for T_1 and T_3 but it could only be 6 hr for T_2.

Average values of acidity from 0-8 hr at an interval of 2 hr and the subsequent values at COB+ve were used to show the trend of increase in acidity (Figure 2). The trend lines of the three treatment groups namely T_1, T_2 and T_3 in the early hours are so closer that it is looking like the same line, then the lines differentiate keeping T_2 at the top, T_3 in the middle and T_1 at the bottom. At the COB+ve hr the trend line of all the treatments showed a tendency to meet a definite point.

For generalized discussion in relation to the values of SPC and CC, acidity values at the beginning and subsequently at 4 hr interval were analyzed. The mean values of per cent acidity for T_1, T_2 and T_3 respectively were 0.1584±0.00695, 0.1622±0.00833 and 0.1611±0.00928 at 0 hr; 0.1667±0.00866, 0.1778±0.00972 and 0.168±0.00791 at 4 hr; 0.1800±0.00935, 0.2103±0.02685 and 0.1846±0.00543 at 8 hr; 0.2420±0.1764, 0.258±0.018 and 0.2409±0.01843 at COB+ve, respectively for T_1, T_2 and T_3. Mean values of acidity per cent at 0 hr and at COB+ve showed no significant difference among T_1, T_2 and T_3, thus indicating no effect of hygiene on initial acidity and acidity at COB+ve of milk. Mean Acidity values at 4 hr and 8 hr were in the order of $T_2>T_3=T_1$ ($p<0.05$). Thus there were no significant differences between the acidity percent under T_1 and T_3 from beginning up to COB+ve.

Table 4. Acidity percentage at 2 hr interval up to COB+ve.

Treatment	Observed acidity percent (Mean±SD) at							
	0 hr	**2 hr**	**4 hr**	**6 hr**	**8 hr**	**10 hr**	**12 hr**	**COB+ve**
T_1	0.158 ± 0.007	0.159 ± 0.006	0.164^{b} ± 0.007	0.171 ± 0.008	0.180^{b} ± 0.009	0.191 ± 0.009	0.213 ± 0.021	0.240 ± 0.013
T_2	0.161 ± 0.007	0.165 ± 0.007	0.174^{a} ± 0.008	0.185 ± 0.009	0.210^{a} ± 0.026	-	-	0.258 ± 0.018
T_3	0.160 ± 0.008	0.162 ± 0.007	0.167^{b} ± 0.007	0.174 ± 0.005	0.184^{b} ± 0.005	0.197 ± 0.006	-	0.240 ± 0.018
Level of Significance	Non-significant	-	p <0.05	-	p<0.06	-	-	Non-significant

$^{a, b}$ = values in the same column with different superscripts differ significantly

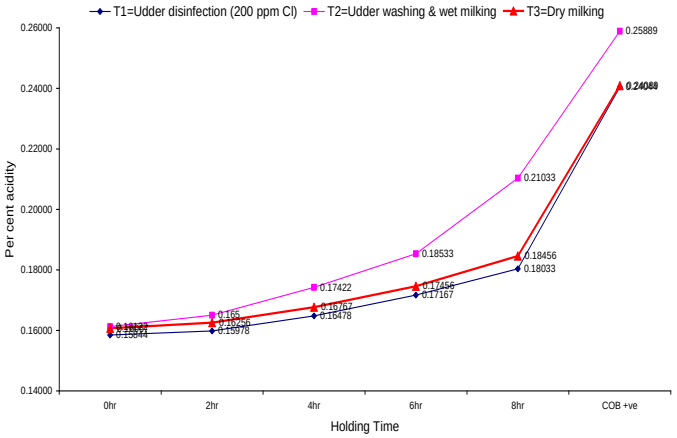

Figure 2. Trend of increase in acidity per cent from the initial period up to COB+ve.

Lakhani and singh (1998) in their keeping quality study with milk obtained by machine versus hand milking, showed that there were no significant differences in the acidity percent initially under both systems of milking. With the increase of storage time there was increase in acidity percent because of more acid production due to increase in bacterial count.

3.5. Clot-on-Boiling (COB) test

COB tests were performed at an interval of 30 minutes. The results are summarized in Table 5. The mean values of COB+ve time (hr) were 12.389±0.33, 8.833±0.56 and 10.944±0.39 respectively for T_1, T_2 and T_3. The COB+ve times were significantly different among treatments ($p<0.01$) and were in the order of $T_1>T_3>T_2$ (lsd=0.4263).

Thus COB+ve time indicates that milk obtained under T_2 get spoiled on average 3.556 hr and 2.111 hr earlier than T_1 and T_2 respectively. Thus milking hygiene can improve the keeping quality of milk by maintaining a lower initial microbial contamination. Lakhani and Singh (1998) showed that average COB+ve time for milk obtained by machine and hand milking were 10.6 hr and 9.3 hr. Sharma and Lavania (1988) reported COB+ve time of only 8 hr for raw milk from small dairies. Results of the present experiment fairly agree with the previous findings and show the importance of hygienic practices during milking.

3.6. Organoleptic evaluation of milk smell

Results obtained by organoleptic evaluation of milk smell under each treatment group are presented in Table 5. On an average normal milk smell were retained for 10.83 hr, 6.22 hr and 8.83 hr under T_1, T_2 and T_3 respectively. It is evident from Table 5 that normal milk smell deteriorates a considerable time earlier than COB+ve time.

Hardling (1995) stated that if the bacterial count of milk were allowed to increase significantly, (e.g. to over 3 million/ml) this could lead to significant degradation of the fat, protein or lactose causing off-flavour. Present study fairly agrees with previous findings.

Table 5. Keeping Quality Time (KQT) based on COB test and milk Smell.

Treatment	Observed parameter (Mean±SD)		
	COB+ve (hr)	Normal smell (hr)	KQT (hr)
T_1	12.39a ± 0.33	10.83a ± 0.61	11.67a ± 0.39
T_1	8.83c ± 0.56	6.22c ± 0.47	8.00c ± 0.69
T_3	10.94b ± 0.39	8.83b ± 0.25	10.08b ± 0.46
Level of Significance	$p < 0.01$	$p < 0.01$	$p < 0.01$

$^{a, b, c}$ = values in the same column with different superscripts differ significantly

3.7. Keeping quality time (KQT)

The term keeping quality is generally used to denote the length of time that milk will remain sweet before it commences to sour and become unfit for use, and this period represents the useful life of the liquid. Keeping quality of sample of milk is the period in hours elapses from its production until it is considered unsuitable for consumption, either because it curdles on boiling or develops an undesirable odour or flavour. Time required to develop detectable abnormal smell and to give COB+ve were averaged to work out the limit of overall acceptable keeping quality time (KQT) in Table 5. There were significant differences ($p< 0.01$) among the average keeping quality times (KQT) of milks obtained under different hygienic conditions. Milk obtained from T_1 showed 3.67 and 1.59 hr extended keeping quality time compared with that of T_2 and T_3, respectively and indicating that milking hygiene practices with 200 ppm chlorine effectively increases the keeping quality of milk.

4. Conclusions

The overall study revealed that milking clean cows in dry condition gives better microbiological and keeping quality of milk compared to washing of udder, milker hands and milking pails with normal water just previous to milking. Washing and disinfection of udder and milker hands, and sanitary rinse of milking pails just previous to milking significantly improved microbial and keeping quality of milk compared to above mentioned two milking hygiene conditions. It is also evident that calcium hypochlorite solution (200ppm Cl) can provide satisfactory reduction in microbial number and can improve keeping quality of milk for a considerable period if used for washing and disinfection of udder and teats, milkers' hands, and rinsing of milking pails just previous to milking.

Conflict of interest

None to declare.

References

Hardling F, 1995. Milk Quality. Aspen Publishers, Inc. Gaithersburg, Maryland, pp.40-45.

Islam MA, MN Islam, MAS Khan, MH Rashid, and SM Obaidullah, 2009. Effect of different hygienic condition during milking on bacterial count of cows' milk. Bang. J. Anim. Sci., 38: 108-114.

IS 1479 (Part III), 1962. Bacteriological analysis of milk. Indian Standards Institution, Manak Bhavan, New Delhi.

Koshy CP and VD Padmanaban, 1990. Studies on P H acidity, thermoduric load and thermophilic bacterial load with reference to keeping quality of market milk. Indian Veterinary Journal, 67: 448-452.

Lakhani GP and VP Singh, 1998. Keeping quality of raw milk by machine milking versus hand milking in buffaloes. Journal of Dairying Foods and Home Sciences, 17: 75-77.

Petrovic M, Z Pavicic, A Tomaskovic and M Cergolj, 2006. Stocarstvo. 60: 403-411.

Sandu M and C Man, 2006. Studied regarding the milk quality from dairy farms and backyard farm in an industrial processing unit. Buletinul-Universitali-de-stinte-Agricole-Si-medicina_Veterinara-Cluj-napoca,-seria-zootehnie-Si-biotehnologii. 62: 107-110

Sharma JM and GS Lavania, 1988. Keeping quality of milk at room temperature. Dairy guide, 10: 21-24.

Sharma-Varsha, KP Singh, RK Sharma and SP Nema, 2005. Microbial qulity of buffalo milk and its public health significance. Buffalo-Bulletin. 24: 78-83.

Walstra P, TJ Geurts, A Noomen, A Jellema and MAJS Van Boekel, 1999. Dairy Technology - Principles of Milk Properties and Processes. Marcel Dekker, Inc. New York.