# Reduction in Numbers of Bacteria after Pre-milking Teat Dipping in Milking Dairy Cows

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# ABSTRACT

This study was conducted to evaluate the amount of bacteria at the teat tips after pre-milking teat dipping, compared to cleaning with chlorine solution disinfectant only. Twenty-four clinically-healthy milking cows from a smallholder dairy farm in a Mae-on cooperative, Chiang Mai, Thailand, were used. After washing, udders were soaked in clean water containing 100 ppm of chlorine and dried with a single-used cloth. Then, samples from the teat tip area were collected after the teats became dry to form a control group (CONTROL). After the same preparation, a teat disinfectant was applied, as for pre-milking teat dipping, to form a treatment group (TREATMENT). Numbers of the colonyforming unit (CFU) of bacteria were counted on a standard agar plate, with those of coliform bacteria counted from a specific culture. Mean log of the CFU was compared between CONTROL and TREATMENT by the Student's T-test. A P-value <0.05 was considered as significant. Based on values of the geometric mean of both groups, total bacterial count of CONTROL was about 3.5 times higher than that of TREATMENT. A result from statistical analysis showed that total bacterial counts in CONTROL were significantly higher than those in TREATMENT. In conclusion, pre-milking teat dipping is advantageous for smallholder dairy farms in which cows are intensively cleaned with chlorine solution disinfectant.

**Key words:** Bacterial count, Chlorine solution disinfectant, Dairy cow, Pre-milking teat dipping

# **INTRODUCTION**

Of farms in Thailand, smallholder dairy farms are in the majority where most dairy farmers have been trained and advised on dairy farm management, including the mastitis control program, by the Dairy Promotion Organization, Thailand. Bovine mastitis is the single most common disease syndrome in adult dairy cows, accounting for about 38% of morbidity (Smith, 1996). Mastitis is also associated with the number of zoonotic diseases in which milk acts as a vehicle of infection (Jenkins, 1982). To prevent mastitis, farmers must provide optimal milking procedures applied with the most hygienic method to minimize pathogens invading udders.

More than 95% of smallholder dairy farms in Thailand have less than 25 milking cows each, and so farmers can spend more time with their cows while milking. Approximately one hour before milking, all milking cows are washed to remove dirt from the udders, and they subsequently stand with an overhead lock until milking. Clean water with chlorine is used for teat disinfection and the udders are dried with a single-used cloth or paper towel before attaching a milking unit. In addition, post-milking dipping is accepted as a general protocol for many dairy farmers in Thailand. However, mastitis prevalence in this area remains high, especially mastitis from environmental pathogens (Boonyayatra et al., 2007). In Chiang Mai, Thailand, Boonyayatra and Chaisri (2004) studied on smallholder dairy farms and found that the prevalence of monthly subclinical mastitis ranged from 36.4% to 83.3%.

Pre-milking teat dipping, followed by post-milking teat dipping, has been established as a more effective procedure against major mastitis pathogens than post-milking teat dipping alone (Oliver et al., 1993). It remains doubtful, however, whether pre-milking teat dipping is advantageous for smallholder dairy farms in which cows are intensively cleaned with chlorine solution disinfectant. Therefore, this study evaluated the amount of bacteria at the teat tips after pre-milking teat dipping, and compared it to cleaning with only chlorine solution disinfectant.

# MATERIALS AND METHODS

### Animals and study design

Twenty-four clinically-healthy milking cows from a smallholder dairy farm in Mae-on cooperative, Chiang Mai, Thailand, were used. All milking cows (n = 24) were fed post-harvest corn stem and rice straw *ad libitum*, and concentrates according to their milk production. A majority of the cows were crossbred Holstein-Friesian, and their average overall dry matter intake was 14.5 kg/cow.

Systematic random sampling was used to assign the cows into either a control (n=12) or treatment (n=12) group. After washing, their udders were soaked in clean water with 100 ppm of chlorine and dried with a single-used cloth. Samples from the teat tip area were collected for the control group (CONTROL) once the teats became dry. After the same preparation, a teat disinfectant containing fatty and lactic acid as main ingredients (Lauricare, 3M, USA) was applied to the

treatment group (TREATMENT), as in pre-milking teat dipping. At least three-fourths of each teat was covered with pre-dip solution. Pre-dip remained in contact with the teat for 30 seconds before drying, and then swab samples were collected from the right-rear quarter of each cow. Sterile cotton buds were used for swabbing in a 1x1 cm<sup>2</sup> area of teat tips and collected in transport media. Samples were immediately transported to the laboratory of the Faculty of Veterinary Medicine, Chiang Mai University.

### Laboratory procedure and statistical analysis

Numbers of the colony-forming unit (CFU) of bacteria were counted on a standard agar plate, with those of coliform bacteria counted from a specific culture. All samples were tested in a single microbiology lab by using identical bacterial growth media provided by the same manufacturer. A microbiologist made final reports. Data were entered and analyzed in Statistix 8.0. Since data were not distributed normally, a logarithm was used to transform CFU data for normal distribution. The mean log of the CFU was compared between CONTROL and TREATMENT by the Student's T-test. A P-value <0.05 was considered as significant.

#### RESULTS

From the total of 12 samples from either CONTROL or TREATMENT, 11 samples (91.7%) of both groups were free from coliform bacteria which were, therefore, not used for a statistical tests for a difference of means. Data on total bacterial counts in both groups are shown in Table 1. Medians of bacterial counts, based on a  $1x1 \text{ cm}^2$  area of teat tips, were 1,489 cfu and 499.5 cfu in CONTROL and TREATMENT, respectively. Based on values of the geometric mean of both groups, total bacterial count of CONTROL was about 3.5 times higher than that of TREATMENT. A result from statistical analysis showed that total bacterial counts in CONTROL were higher than those in TREATMENT (P<0.05).

**Table 1.** Comparison of total bacterial counts between CONTROL (conventional<br/>preparation of teats with 100 ppm chorine in smallholder dairy farms<br/>in Thailand) and TREATMENT (conventional preparation, as in CON-<br/>TROL, followed by pre-milking teat dipping for 30 seconds and dried<br/>with a single-used cloth)

	Ν	Min.	Median	Max.	Geo.Mean	Lower than 95% CI	Above 95% CI
Control	12	225	1,489	28,800	1,846	836	3,587
Treatment	12	90	499.5	3,330	518.3	321.5	949.8

T statistic = 2.65, Degree of freedom = 22, P = 0.01

# DISCUSSION

In this study, intensive cleaning was carried out in a smallholder dairy farm by washing, soaking with chorine solution and drying to minimize the number of bacteria and coliform count at the teat tip area. Most coliform-free teats observed in this study were similar to total-coliform teats found after wetting only, followed by manual drying (Fairchild et al., 1982). Geomean numbers of bacteria in CON-TROL were at the same levels as those in milk after teat preparation by either using wet towel, sanitizer and drying or disinfectant dipping and drying (Galton et al., 1984). However, the most pathogens causing mastitis around the world including Thailand were gram positive bacteria, such as environmental Streptococci and *Staphylococcus* spp. counting for more than 90% of total(Boonyayatra et al., 2007; Suriyasathaporn, 2010; Radostits et al., 2007). Therefore, the use of total bacteria count was a better indicator for measurement of teat dipping efficacy.

The cow-prevalence of subclinical mastitis in tropical countries ranges from 40% to 90%, including approximately 45% in India (Roman et al., 2000; Joshi and Gokhale, 2006), 38.2% in Ethiopia (Workineh et al., 2002) and from 75.9% (Karimuribo et al., 2006) to 90.3% (Kivaria et al., 2004) in Tanzania. In northern Thailand, environmental mastitis has the highest prevalence in the country, even on farms with intensive milking preparation (Boonvavatra et al., 2007). Most cows are fixed with an overhead lock but free movement of the hind quarter allows contamination with feces in the bedding area. The optimal temperature and humidity in Thailand, as a tropical country, exacerbates the numbers of environmental bacteria on the floor and also various cow surface areas, especially the udder. In this study, we showed that bacteria were effectively reduced by approximately 3.5 times after adding pre-milking teat dipping, and drying after washing and soaking with a sanitizer (Table 1). This reduction level would help lactating cows decrease intramammary infection. In western countries, it has been established that pre-dipping teats with a germicidal teat dip reduces new cases of environmental mastitis during lactation (Hogan and Smith, 1987).

The result from this study showed that the pre-dipping teat procedure was advantageous for smallholder dairy farms in which the udder was intensively washed and soaked with chorine solution disinfectant. The use of paper or cotton towels to wipe out pre-milking teat disinfectant from the skin was necessary in the case of cleaning with iodine (Galton et al., 1984; Rasmussen et al., 1991). The suggestion that iodine residues in milk originate from contaminated teat surface rather than absorption through the skin is rather valid, because iodine is a residue in milk.

In conclusion, pre-milking teat dipping is advantageous for smallholder dairy farms in which cows are intensively cleaned with chlorine solution disinfectant. The use of pre-milking teat dipping would help smallholder dairy farms reduce the prevalence of intramammary infection and mastitis.

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