

Chronic Mastitis in Small Dairy Cattle Herds in Muang Khon Kaen

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Abstract

Monthly records of bulk tank milk somatic cell count (BMSCC) of all 194 dairy herds in Muang, Khon Kaen were used as an indicator of chronic mastitis problems; i.e. BMSCC > 500,000 cells/ml at least 3 consecutive records. Twelve chronic mastitis herds in Muang, Khon Kaen were data collected on 1) general herd information, 2) milking equipment, 3) milking procedure and 4) awareness and management of farmers to the mastitis problem. All possible mastitis pathogens were investigated in individual milk samples from 138 lactating cows of the herds. At least 40% of dairy herds in Muang, Khon Kaen had mastitis at each record and 23% (44 herds) had the long-term problem. Farmers of the 12 studied herds had poor managements on mastitis control, such as improper milking machine maintenance, no use of pre-dipping; foremilk had not been checked for abnormality, mastitis cows were milked with fresh cows. At the cow level, the prevalence of contagious bacteria, coagulase negative staphylococci (CNS) and opportunistic bacteria was 39%, 64% and 22%, respectively. Milking practice was identified as a risk factor of cow being infected intramammary by contagious bacteria and CNS.

Keywords: Coagulase negative staphylococci, dairy cattle, Khon Kaen, mastitis, *Staphylococcus aureus*, *Streptococcus agalactiae*

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บทคัดย่อ

เต้านมอักเสบเรื้อรังในฟาร์มโคนมขนาดเล็กในเขตเมืองขอนแก่น

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ทำการบันทึกรายเดือนค่าเซลล์โซมาติกในถังนมรวมของฟาร์มโคนมจำนวน 194 ฟาร์มของสหกรณ์โคนมแห่งหนึ่งใน อ. เมืองขอนแก่น นำมาวิเคราะห์ปัญหาเต้านมอักเสบเรื้อรังซึ่งมีค่าเซลล์โซมาติกในถังนมรวมสูงกว่า 500,000 เซลล์ต่อน้ำนมดิบหนึ่ง มล. ติดต่อกันมากกว่า 3 เดือน ฟาร์มในกลุ่มนี้จำนวน 12 ฟาร์มได้ถูกเลือกเพื่อศึกษาและเก็บข้อมูลของฟาร์มซึ่งได้แก่ 1) ข้อมูลทั่วไป 2) การดูแลอุปกรณ์รีดนม 3) กระบวนการรีดนม และ 4) ความตระหนักและการจัดการปัญหาเต้านมอักเสบของเกษตรกร เชื้อแบคทีเรียก่อโรคเต้านมอักเสบถูกจำแนกจากแม่โครีดินมจำนวน 138 ตัวของทั้ง 12 ฟาร์ม ผลการศึกษาพบว่ามากกว่าร้อยละ 40 ของฟาร์มโคนมมีปัญหาเต้านมอักเสบและร้อยละ 23 (44 ฟาร์ม) มีปัญหาเต้านมอักเสบระยะยาว เกษตรกรของทั้ง 12 ฟาร์มมีปัญหาในการควบคุมเต้านมอักเสบ เช่น ดูแลรักษาระบบและเครื่องรีดนมไม่เหมาะสม ไม่ใช้ยาจุ่มก่อนรีด ไม่ตรวจน้ำนมก่อนรีด รีดโคที่เป็นเต้านมอักเสบร่วมกับโคปกติ ผลการเพาะแยกเชื้อพบความชุกของเชื้อเต้านมอักเสบติดต่อ โคแอกกูเลสเนกาทีฟ สตาฟไฟโลคอคโคและ เชื้อในสภาพแวดล้อม ที่ร้อยละ 39, 64 และ 22 ตามลำดับ กระบวนการรีดนมเป็นปัจจัยเสี่ยงต่อการติดเชื้อเต้านมอักเสบติดต่อและโคแอกกูเลสเนกาทีฟ สตาฟไฟโลคอคโค

คำสำคัญ: โคแอกกูเลสเนกาทีฟ สตาฟไฟโลคอคโคโคนม ขอนแก่น เต้านมอักเสบ สตาฟไฟโลคอคคัส ออเรียส สเตรปโตคอคคัส อกาแลคติอี

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Introduction

Mastitis, an inflammation of mammary glands, is the most frequent and costly disease of dairy cattle worldwide. In most of the cases, mastitis is caused by infection of microorganism. The causative microorganisms could be classified by its means of transmission into contagious, environmental and opportunistic pathogens. The contagious mastitis pathogens comprise of *Streptococcus agalactiae* (*Str. agalactiae*), *Staphylococcus aureus* (*Sta. aureus*) and mycoplasmas (Jayarao and Wolfgang, 2003). The environment group consists of other streptococci and gram negative bacteria. Opportunistic infection of the mammary gland usually resulted from infection with coagulase-negative staphylococci (CNS). All pathogens cause an inflammation of the mammary glands, which then increase number of somatic cell in the produced milk, which subsequently increase the bulk milk somatic cell count (BMSCC). Thus BMSCC is used as a mastitis indicator in dairy herd (Jayarao and Wolfgang, 2003; Jayarao et al., 2004).

In Muang district of Khon Kaen province, most of the dairy herds were small to medium size, i.e. 3-20 lactating cows/herds that produce milk on average 12 kg/cow/day. Farmer usually send milk for sell at the nearby local co-operative milk collection centre (MC) twice a day. At the MC the quality of

milk from each farm will be tested by monthly BTM collect. The milk samples will be examined, i.e. BMSCC and total bacterial count, once a month at the Veterinary Research and Development centre, Khon Kaen. The result of milk quality testing is used to set price of buying. In 1999, the Department of Livestock Development of Thailand announced a dairy herd standard which stated that the BMSCC should not excesses 500,000 cells/ml of raw milk. Many encourage has been used to push Thai farmers aware of losses due to the intramammary infection (IM) but more than 40% of the Thai dairy herds still had BMSCC greater than the standard (Bureau of Quality Control of Livestock Products, 2010), and mastitis stills the most concerned problem of the farmers. Many studies had been done on identification of the pathogenic organisms and evaluate risk-factor of mastitis in Thai dairy herds; however, in Muang Khon Kaen there was only one study done in the last 10 years (Aiumlamai et al., 2000). The aim of this study was to gain a current status of bovine mastitis in the area and also to determine on-farm risk factors that could be associated with prevalence of each mastitis pathogen groups in long-term high BMSCC dairy cattle herds in Muang Khon Kaen.

Materials and Methods

Prevalence of Mastitis in Muang, Khon Kaen and herd selection: Throughout the study period, 194

dairy herds sent their milk to a milk collection centre where their bulk tank milk (BTM) was monthly collected for milk quality and BMSCC evaluation. BMSCC records from January-April 2009 were used to identify a chronic mastitis problem i.e. BMSCC greater than 500,000 cells/ml for at least 3 consecutive tests. Twelve dairy herds that had the indication were selected for herds visiting, data collection, and individual milk sampling for bacterial identification.

Milk samples and on-farm data collection: Herd visit and individual milk sampling was carried out in April-May 2009. The collected herd's data comprise of 4 aspects; 1) general herd information, 2) milking equipment, 3) milking procedure and 4) awareness and management of farmers to the mastitis problem. Quarter milk samples were taken aseptically from all lactating cows directly after the milking. The samples were transported chilled at 4°C to the Faculty of Veterinary Medicine, Khon Kaen University.

Bacterial identification: The bacteriological analyses were done within 3 hours after sampling. One-hundred microlitre of milk was inoculated onto 5% bovine blood agar plate and MacConkey agar plate. After 18 hours of incubation, specific bacterial colonies were examined. Species identifications were done by conventional biochemical tests according to National Mastitis Council (1987) and Quinn et al. (1994).

Identification of *M. bovis* was done on filtrated, cultivated milk samples. Firstly, 0.5 ml of milk were mixed with 2 ml of modified Heyflick's broth (Difco™ PPLO broth plus 30% of Difco™ PPLO Supplement) before were filtrated through 0.45 micron What Man® filter. The filtrated milk dilution was then incubated at 37°C for 8 days. A high Pure PCR Template Preparation Kit™ (Roche, Germany) was used to extract bacterial DNA from the cultured broth; all the procedures were followed the manufacturer's

instruction. A commercially available nested polymerase chain reaction (nPCR) kits for detection of *M. bovis* (Genekam™, Germany) was used according to the manufactured suggestions; positive and negative controls which provided with the kits, were analyzed in every steps of the nPCR (Kampa et al., 2009).

Statistical analyses: Data from on-farm visit were coded and analyzed. Prevalence of each microbial group were analyzed both at quarter- and cow-level. Due to prevalence of infection and management relied on herd's practice that affect individual cow rather than individual udder, thus predictors were measured at the cow-level. Unconditional association between outcome of the interest (group of pathogen) and each of the predictors was examined by using simple logistic regression. Only predictors showing association with outcome of the interest at $p \leq 0.15$ were considered for the subsequent multivariable logistic regression. Non-significant variables were removed sequentially using backward elimination at $p \leq 0.05$. Evaluation of interaction among predictors was done by using the Hosmer-Lemeshow test (Dohoo et al., 2003). The analysis was performed using statistic software, Stata version 8.2 (Stata Corporation, College Station, Texas US).

Results

Prevalence of mastitis herds in Muang Khon Kaen: By BMSCC records, a problem of mastitis in Muang Khon Kaen was analyzed. In all records of January-April 2009, the average monthly BMSCCs of the 194 herds were greater than 500,000 cells/ml (Table 1) and prevalence of herds with mastitis problem were 47%, 40%, 47% and 48%, respectively. In addition, 44 herds (23%) had BMSCC above 500,000 cells/ml. at least 3 consecutive tests which indicated the chronic mastitis problem. Among the chronic mastitis group, 12 herds were selected.

Table 1. Bulk milk somatic cell count of 194 dairy herds in Muang Khon Kaen from January-April 2009.

Month	Geometric Mean (x10 ³ cells/ml)	Range (x10 ³ cells/ml)	SD (x10 ³ cells/ml)	Median (x10 ³ cells/ml)
January	629	61-3943	544	468
February	611	39-3746	595	422
March	590	47-2996	464	463
April	741	71-5766	737	474

Herd's data

General herd information: An average size of the studied dairy herds was 11 milking cows, with mixed Holstein-Friesian and Sahival breeds, and with a daily milk production of approximately 11.7 kg/cow (range 7.5-16 kg/cow). The BMSCC from January to April of each herd shows in Figure 1. Last year cows in herd No. 6, 7 and 8 were culled because of failure in reproduction while mastitis was not a main reason for culling. Exception for herd No. 1 and 9, the other passed the official standard for Thai dairy herd. No

animals were permitted to stand in farm ponds or deep mud. Vaccination against mastitis was not practiced in that area.

Milking equipment: Only herd No. 1 and 10 ever used a commercial brand milking machine while the others used local produced-milking machine. There was no routine efficacy checking report within two years. Immediately cleaning of milking equipment after milking was done only in herd No.7.

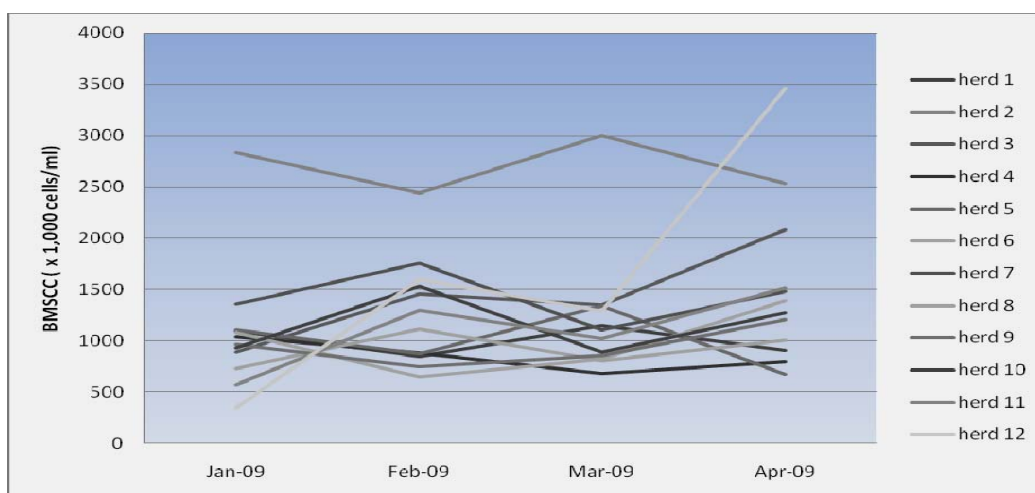


Figure 1. Monthly bulk tank milk somatic cell counts of 12 dairy herds in Muang Khon Kaen during January-April 2009.

Milking procedure: Before milking, cows were teat washing by using of either ground water or well water. The water was treated with chlorine before used only in herd No. 7, 8 and 10. Cows in herd No.1 and 6 were teat wiped with individual cloth rag, whilst teats of cows in the other herds were wiped with common cloth rag. Pre-dipping was not practiced in all 12 herds. Milk and udders were not examined for any abnormality (color, texture, nodules) before attaching teat cups. California mastitis test (CMT) or other tests for detecting of mastitic milk had not been used during the last 2 years in all 12 herds.

Most of cows were not milk out within 3-5 minutes after cleaning. Milking was not done in order neither by production, lactation nor mastitis status. Cow could go to their desired position and was fed concentrate food at the milking times. During the milking time, teat cups were not dipped aseptically between cows. After milking, only cows in herd 12 were teat disinfected with a commercial post-dipping solution.

At the sampling time, observers found

abnormal teats in cows of herds 2-5 and 8-12. And by udder massage and milk examination, except herd No. 12, we found mastitis cow for at least 1 of each herd.

Awareness and management of farmers to the mastitis: By the questionnaire, farmers knew how to discriminate clinical and subclinical mastitis, how to detect mastitis by using CMT, how to prepare the chlorinated water, how to clean their milking apparatus properly and mastitis cows should be milked in the last order. In case of clinical mastitis, farmer treated their cows without suggestion from veterinarian.

Bacterial identification: Milk samples of 138 cows were collected. Some cows had blind teat thus 544 quarter milk were sampled in total. At the quarter level (Figure 2), the prevalence of contagious, CNS and environmental bacteria was 22%, 30% and 7%, respectively ($p < 0.05$). Prevalence of *Staphylococcus aureus* was 3% while the prevalence of *Str. agalactiae* was 19%. *Mycoplasma bovis* was not found in any milk samples. In-herd prevalence of each identified bacteria, except *Str. dysgalactiae* and *Str. uberis*, were significantly difference ($p < 0.05$).

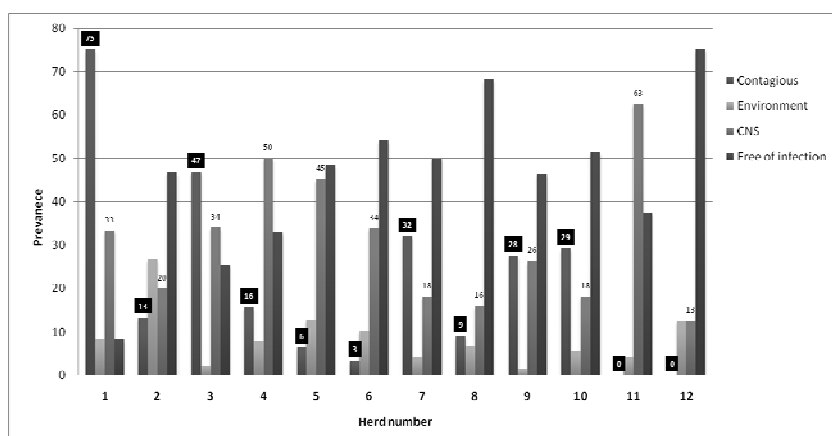


Figure 2. Prevalence of microorganisms in 544 quarter milk samples from 138 lactating cows 12 dairy cattle herds in Muang Khon Kaen. Prevalence of contagious pathogen and coagulase negative staphylococci (CNS) are labeled.

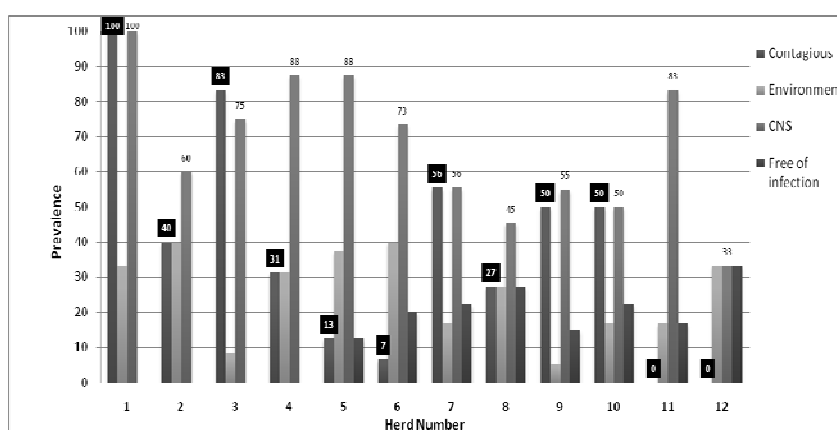


Figure 3. Prevalence of microorganisms in milk samples from 138 lactating cows of 12 dairy cattle herds in Muang Khon Kaen. Prevalence of contagious pathogen and coagulase negative staphylococci (CNS) are labeled.

At the cow level (Figure 3), the prevalence of contagious, opportunistic and environmental bacteria was 39%, 64% and 22%, respectively ($p < 0.05$). In the contagious group, prevalence of *Str. agalactiae* and *Sta. aureus* was 36% and 7%, respectively. In-herd prevalence of most identified bacteria, except *Str. dysgalactiae*, *S. pyogenes* and *Str. uberis*, were significantly difference ($p < 0.05$). Every cow in herd number 1-4 had intramammary infection. More than 30% of cows in herd 1-4, 7, 9 and 10 had intramammary infected with contagious pathogen. Environmental streptococci and enteric bacteria were found in a high prevalence ($> 30\%$) in herd 1, 2, 4-6 and 12. Coagulase-negative streptococci were isolated in all herds with herd-prevalence between 33-100%.

On-farm risk factor to intramammary infection with each group of mastitic pathogens: Table 2 shows predictors that were unconditionally associated ($p \leq 0.15$) with prevalence of each group of mastitis pathogens. Cows that were teat cleaned with non-chlorinated water before milking had a significant higher chance to have intramammary infection (Odd Ratio 2.02, $p < 0.05$). Risk of being IMI with contagious pathogens was significantly greater ($p < 0.05$) with milk production, age of the milking machine, improper maintenance of the machine and milking system, use of common cloth rag and poor hygiene in using intramammary drugs (OR 2.64, 0.06, 1.25, 0.71 and 0.64, respectively). Risk of being IMI with environmental pathogen was significantly greater with improper maintenance of the milking system and poor hygiene in using intramammary drugs (OR 1.30 and 3.11, respectively). Opportunistic pathogen had a significantly higher chance ($p < 0.05$) to be found in cows that were cleaned only at the teat rather than the whole body before milking and using of common cloth rag in teat cleaning (OR 1.83 and 0.34, respectively).

Multivariable statistical analyses produced a model with 2 significant ($p < 0.05$) predictors to contagious pathogens. There was no evidence of lack of fit of the model as indicated by the Hosmer-Lemeshow goodness of fit test for final model of contagious and environment microorganisms ($p = 0.9684$ and $p = 1.000$). By the final model (Table 3), the risk of IMI with contagious pathogen was higher

when using the common cloth rag between cows and when teat was not been cleaned properly before using of an intramammary antimicrobial drug. Additionally, the later factor also increases risk of being intramammary infected by environmental microorganisms.

Discussion

Analysis of BMSCC records revealed a failure in mastitis control of dairy farmers in Muang Khon Kaen. The problem could be controlled by a good hygienic milking practice but almost a half of the dairy population had a high BMSCC. Furthermore, 23% of the dairy herds had high BTMSCC longer than 3 months period, indicated a chronic mastitis problem. Based on monthly BMSCC reports of Dairy Milk Quality Control in Thailand, in every record from June 2008 to January 2010, more than 50% of dairy herds had mastitis (Bureau of Quality Control of Livestock Products, 2010). Thus, mastitis was also found in the other cattle population as well and some of mastitis herds probably had the chronic problem. With high proportion of mastitis herds, both in Muang Khon Kaen and all over Thailand pointed to a necessary to use a pragmatic procedure for improving the situation.

In the 12 chronic mastitis herds in Muang Khon Kaen, 117 out of 138 lactating cows were intramammary infected. When compare to study by Aiumlamai et al. (2000) that had done in the same population, at the quarter level, prevalence of *Sta. aureus* in the present study was lower (17% and 3%, respectively) but *Str. agalactiae* was higher (7% and 19%, respectively). However, the prevalence of total contagious bacteria and CNS in this study was close to the previous 10-years report. This situation could be explained by no improvement of milking hygiene in the studied herds, i.e. inappropriate maintainance of milking system, clean teats with non-chlorinated water before milk, using of common cloth rag, no foremilk checking of mastitis, incorrect use of intramammary drug. Nevertheless, because of mastitis treatment was relied on farmer's decision only, mutation of the infective bacteria and drug resistant may disturbed the efficiency of antibiotic that farmer used. Therefore a continuing study of the

mutation and drug sensitivity test should be done.

Table 2 Unconditional association ($p \leq 0.15$) between the risk factor and preserve of mastitis pathogens in dairy herds in Muang, Khon Kaen.

Group of pathogen	Variable	Odds Ratio	p-value
Overall intramammary infection (Yes or No)	<i>Milking procedure:</i> Before milking, the cow was cleaned only at teats and udders rather than the whole body	2.02	0.0511
	The teats were cleaned by non-chlorinated water before milking	2.64	0.0421
Contagious pathogens (Yes or No)	<i>General herd information:</i> Cow produce milk > 12/kg/day	0.0620	0.000
	<i>Milking equipment:</i> Age of the milking machine	1.245	0.0008
	Milking machine was not washed immediately after use	0.3645	0.0224
	Pipe line had been washed more than 1 time/month	0.7092	0.0220
	<i>Milking procedure:</i> Use of common cloth rag	0.6421	0.0011
	<i>Awareness and management of farmers to the mastitis problem:</i> The teats were improperly cleaned and non-sanitized before administering treatment	0.4227	0.0273
	The intramammary infusion administered was not using the 'partial insertion' technique	0.500	0.0760
	<i>General herd information:</i> Cow produce milk > 12/kg/day	2.25	0.0754
	<i>Milking equipment:</i> Pipe line had been washed more than 1 time/month	1.3007	0.0297
Environmental pathogens (Yes or No)	<i>Milking procedure:</i> The chlorinated water was prepared improperly	0.6929	.0779
	Use of common cloth rag	0.4439	0.0748
	<i>Awareness and management of farmers to the mastitis problem:</i> The teats were improperly cleaned and non-sanitized before administering treatment	3.1127	0.0306
	Mastitic cow was treated with other methods rather than using of antibiotic intramammary/ systemically	0.9814	0.1319
	<i>Milking procedure:</i> Before milking, the cow was cleaned only at teats and udders rather than the whole body	1.8275	0.0163
	The mammary glands were wiped by non-chlorinated water before milking	1.9630	0.0597
	Use of common cloth rag	0.3381	0.0189
Opportunistic pathogen	<i>Awareness and management of farmers to the mastitis problem:</i> Mastitic cow was treated with other methods rather than using of antibiotic intramammary and systemically	0.9815	0.0970

Table 3. Final multivariable logistic regression model for on-farm significant risk factors for finding of mastitic pathogens in dairy herds in Khon Kaen province.

Groups of pathogen	Risk factors	Odd ratio	p-value	95% Confidence interval
Contagious	The teats were improperly cleaned and non-sanitized before administering treatment	0.0161	0.039	0.0003-0.8106
	Use of common cloth rag	7.5716	0.013	1.5387-37.2576
Environment	The teats were improperly cleaned and non-sanitized before administering treatment	5.2703	0.037	1.1024-25.1968

There was no evidence of *M. bovis* mastitis from this study, even a high sensitivity method as a nested PCR was used and the organism was identified in a BTM of dairy herd in studied area (Kampa et al., 2009). During this study period, the previously found *M. bovis* herd had a low BMSCC, thus was not included into studied group. In general prevalence of *M. bovis* is very low, 0.5-2%, (Gonzalez and Wilson, 2003) and there were a small number of visiting herds thus difficult to identified in this study.

Most of the cows (64%) were intramammary infected with CNS. In the past, only *Sta. aureus* is only

one important mastitis staphylococci because of its ability to cause clinical and subclinical mastitis. Taponen and Pyörälä (2009) reviewed an important of CNS in its persistence in mammary gland, increasing somatic cells in milk and resistance to various antibiotics. An important of IMI with CNS showed in herd number 11 and 12 where prevalence of contagious pathogen were 0 and prevalence of environmental pathogen were low (17%). In April, both herd's BMSCC were greater than 1,500,000 cells/ml. An increasing prevalence of coagulase-negative staphylococci in many cattle populations,

give rise in its role as another important mastitic pathogen. For example, in the Netherlands, the prevalence of CNS among bacterial isolated in both subclinical and clinical mastitis, 16.2% in 1999 to 42.2% in 2004 and from 7.3% to 14.1%, respectively (Sampimon et al., 2007). In other countries i.e. USA, Norway, Germany and England and Wales, CNS is among the most isolated microorganisms from subclinical mastitis (Rajala-Schultz et al., 2004; Østerås et al., 2006; Tenhagen et al., 2006).

Because the relatively small number of cows and herds include in the study, only variables that were strongly associated with identified pathogens could be evaluated in the final model. The absence of the variables, especially which was significant in unconditional association test, from the final model may be the limited sample size. Additionally, some variables that obviously suggested for mastitis could not be evaluated because it appeared in most of the studied herd. Moreover, this study was done only on herds that had long-lasting high BMSCCs thus no comparison to herd that had low BMSCCs which their practice would differ.

To correct the problem and increase a good quality milk production, a pragmatic preventive program should be seriously pushed to farmers. Because when a good preventive program practiced, significant improvements in udder health have been reported (Green et al., 2007). However the success of the preventive program depends on farmer motivation and behavior, especially the socio-economic factors (Lam et al., 2007). Studies in Dutch dairy farmer also show inspiration influence the farmer's effort is to get high net return but to keep the farm management simple (Jansen et al., 2004; Kuiper et al., 2005). Identification of pathogen and source/cause of the infection will help to reduce the opportunity of microbial spreading. This can be done through segregation and treatment of the infected cows. Farmers of all 12 herds did not detected mastitis in their cow neither by CMT or any physical tests. Many cows had nodules in udder which our investigators could found them by udder palpation. Thus, none of the herd had segregate mastitis cows from other cows in their herd, gave rise of spreading through milking procedure. Hence, fresh cows were milked with cows that have mastitis. An on-site test as CMT has been show its efficiency and cost-effective and user-friendly but had never been used in the problematic herds. Strong encourage farmers to realize the mastitis problem and motivate them to solve it is a core-component to reduce the prevalence of mastitis and improve raw milk quality of the Thai dairy farms.

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References

- Aiumlamai, S., Chanlun, A., Sukolapong, V., van Oostveen, F., Jarassang, C., Krajangpo, C. and Pattanawong, J. 2000. Prevalence of organisms causing subclinical mastitis in Amphur Muang, Khon Kaen. Proceeding of the 26th Veterinary Annual Conference, Bangkok, Thailand, 15-17 November 2000, pp. 53-63.
- Bureau of Quality Control of Livestock Products, Department of Livestock Development, Thailand. 2010. "Results of Milk Quality Control" [Online]. Available: http://www.dld.go.th/qcontrol/index.php?option=com_content&view=article&id=162:2010-01-11-06-15-16&catid=54:2009-11-09-07-23-09&Itemid=61
- Dohoo, I.R., Martin, W. and Stryhn, H. 2003. Veterinary Epidemiologic Research. AVC Inc.
- Green, M.J., Leach, K.A., Breen, J.E., Green, L.E. and Bradley, A.J. 2007. National intervention study of mastitis control in dairy herds in England and Wales. *Vet. Rec.* 160(9): 287-293.
- Jansen, J., Kuiper, D., Renes, R.J. and Leeuwis, C. 2004. Report Baseline Survey Mastitis: knowledge, attitude, behavior. Wageningen University, Wageningen, the Netherlands.
- Jayarao, B.M., Pillai, S.R., Sawant, A.A., Wolfgang, D.R. and Hegde, N.V. 2004. Guidelines for monitoring bulk tank milk somatic cell and bacterial counts. *J. Dairy Sci.* 87(10): 3561-3573.
- Jayarao, B.M. and Wolfgang, D.R. 2003. Bulk-tank milk analysis: A useful tool for improving milk quality and herd udder health. *Vet. Clin. North Am. Food Anim. Pract.* 19(1): 75-92, vi.
- Kampa, J., Sukolapong, V., Buttasri, A. and Charoenchai, A. 2009. Prevalence of *Mycoplasma bovis* and other contagious bovine mastitis pathogens in bulk tank milk of dairy cattle herds in Khon Kaen province, Thailand. *Thai J. Vet. Med.* 39(3): 275-280.
- Kuiper, D., Jansen, J., Renes, R.J., Leeuwis, C. and van der Zwaag, H.G. 2005. Social factors related to mastitis control practices: The role of dairy farmer's knowledge, attitude, values, behavior and network. Proceeding of the 4th IDF Int. Mastitis Conference, Maastricht, the Netherlands, pp. 576-582.
- Lam, T.J.G.M., Jansen, J., Van den Borne, B.H. and van Veersen, J. 2007. A structural approach of udder health improvement via private practioners: ups and downs. Proceeding of the NMC 46th Annual Meeting, San Antonio, Texas, USA, pp. 142-151.
- National Mastitis Council. 1987. Laboratory and filed handbook on bovine mastitis. National Mastitis Council, Inc.
- Østerås, O., Sølverød, L. and Reksen, O. 2006. Milk culture results in a large Norwegian survey-effects of season, parity, days in milk, resistance, and clustering. *J. Dairy Sci.* 89(3): 1010-1023.
- Quinn, P.J., Carter, M.E., Markey, B.K. and Carter, G.R. 1994. Clinical Veterinary Microbiology. Mosby-Year Book Europe Limited.
- Rajala-Schultz, P.J., Smith, K.L., Hogan, J.S. and Love, B.C. 2004. Antimicrobial susceptibility of

- mastitis pathogens from first lactation and older cows. Vet. Microbiol. 102(1-2): 33-42.
- Sampimon, O.C., Vernooij, J.C., Mevius, D.J. and Sol, J. 2007. Sensitivity to various antibiotics of coagulase-negative staphylococci isolated from samples of milk from Dutch dairy cattle. Tijdschr Diergeneeskd. 132(6): 200-204.
- Tenhagen, B.A., Koster, G., Wallmann, J. and Heuwieser, W. 2006. Prevalence of mastitis pathogens and their resistance against antimicrobial agents in dairy cows in Brandenburg, Germany. J Dairy Sci. 89(7): 2542-2551.
- Taponen, S. and Pyörälä. 2009. Coagulase-negative staphylococci as cause of bovine mastitis- not so different from *Staphylococcus aureus*? Vet. Microbiol. 134(1-2): 29-36.