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Herd Level Information and Bulk Tank Milk Analysis: Tools for Improving Milk Quality and Herd Udder Health

Bhushan M. Jayarao, *MVSc, PhD, MPH;* **Shreekumar R. Pillai,** *MVSc, PhD;*
David R. Wolfgang, *VMD;* **David R. Griswold,** *VMD, MS;* **Lawerence J. Hutchinson,** *DVM*
Department of Veterinary Science, 111 Henning Bldg., Pennsylvania State University, University Park, PA 16802

Abstract

When milk produced on a farm is examined for bacteriological milk quality and mastitis causing bacteria, it can disclose descriptive information about the general udder health status of the herd, milk hygiene and milking practices on the farm. Many dairy producers periodically receive information about their bulk tank milk with reference to standard plate counts and bulk tank somatic cell counts. Some dairy producers also receive a report on preliminary incubation counts. This information, when collected over a period of time, in combination with bulk tank mastitis culture reports can become a significant knowledge base. This comprehensive data, when interpreted in context with the farm's management practices, provides a rationale to determine current and potential milk quality and mastitis problems in a herd. This paper describes the process of collecting, analyzing, and interpreting bulk tank milk microbiology test results, and utilizing the information to make decisions on improving udder health of the herd and improving milk quality.

Résumé

L'analyse du lait produit à la ferme dans le but de déterminer la qualité bactériologique du lait et la présence de bactéries causant la mammite permet d'obtenir de l'information sur la santé du pis dans un troupeau et sur l'hygiène du lait et des pratiques de production de lait à la ferme. Plusieurs producteurs laitiers reçoivent périodiquement de l'information sur le comptage bactérien total et le nombre de cellules somatiques du lait de réservoir. Quelques producteurs reçoivent aussi un rapport préliminaire des comptes d'incubation. Cette information, lorsque ramassée sur une longue période de temps conjointement avec les rapports sur les cultures de bactéries causant la mammite provenant du lait de réservoir, peut améliorer les connaissances sur le troupeau. Ces données, interprétées à la lumière du mode de gestion du troupeau, peuvent fournir une base pour déterminer les problèmes actuels et éventuels de la qualité du lait et de la mammite dans un troupeau. Cette présentation se penche sur les processus de collecte, d'analyse et d'interprétation des rapports de tests microbiologiques du lait de réservoir et sur l'utilisation de l'information pour améliorer la qualité de santé des pis dans le troupeau et la qualité du lait.

Introduction

Bulk tank milk (BTM) from all dairy farms is periodically tested for antibiotic residues and bacterial contamination. Many progressive milk cooperatives and processors periodically test raw milk quality and also encourage milk producers to test their BTM for mastitis-causing bacteria. The concept of using BTM to identify mastitis pathogens began in California in the 1970s. Soon afterwards, researchers in Minnesota defined, developed and refined techniques for conducting BTM analysis for environmental pathogens.[14,15] Studies conducted over the last decade have shown that examination of BTM is useful for diagnosing multiple problems (current and potential) that might exist in a dairy herd related to milk quality and mastitis pathogens.[4,19]

Over the last five years, the use of BTM analysis as a tool to determine milk quality and troubleshoot herds with mastitis has received a lot of attention, especially from veterinarians and dairy health consultants who view milk quality and mastitis as an

important part of their consulting service for their clients.[6,7,11,27,30] Progressive milk cooperatives have begun to monitor BTM for milk quality and mastitis pathogens to reward dairy producers who excel at producing quality milk and have a low incidence of mastitis. In addition, milk producers and cooperatives view BTM analysis as an important part of their quality assurance program.

The original concept of doing BTM analysis arose from the idea of reducing the number of samples required to determine the number of cows in a herd with subclinical mastitis.[14,15] Over time, the understanding of the bacteriology of raw milk, mastitis, and farm management practices related to milking and milk hygiene has increased considerably, making it possible to formulate strategies to improve milk quality and reduce the incidence of mastitis in dairy herds.[2,3,13,18,25]

The benefits and limitations of using BTM analysis are outlined in Table 1. Briefly, the benefits of conducting a BTM analysis are: 1) it saves time (time needed to collect individual quarter milk samples, and time needed to conduct laboratory tests), 2) it is less expensive as compared to a whole herd culture, and 3) it is an important part of a quality assurance program. The key limitations of BTM analysis are: 1) it does not provide information on an individual cow basis for either milk quality or mastitis, 2) information about herd management practices related to milking and milk hygiene is needed to interpret the BTM analysis reports, 3) samples must be shipped on ice or icepacks, and 4) it must arrive at the laboratory within 36 h of collection. The focus of this paper is to provide the most recent available information about bulk tank milk analysis, and to discuss how it can be used in routine veterinary practice to improve milk quality.

A Systematic Approach to Conducting a Bulk Tank Milk Analysis

The first step is to define the need for doing BTM testing for the client. Bulk tank milk analysis can be effectively used for resolving nine important milk quality issues. This list of issues was developed by the Field Investigation Group at the Pennsylvania State University with input from veterinarians, dairy producers and sanitarians (Table 2). The second step is to set up a BTM sampling process. BTM should preferably represent one milking and should be collected 1-2 hours after that milking. Bulk tank milk containing more than one milking is difficult to interpret because some of the milk quality attributes, such as preliminary incubation counts, are strongly influenced by temperature and the length of time the milk has been held in the bulk tank.

Milk samples must be collected from the top of the bulk tank using a clean sanitized dipper. The National Mastitis Council[22] recommends that the milk be agitated for at least 10 minutes prior to sample collection. Milk collected from the outlet valve usually has a higher standard plate count and laboratory pasteurization count because of contamination with the soil type resident flora around the valve and its threading. Milk collected from the top of the bulk tank without agitation also gives erroneous results. This milk invariably has a very high fat content, high somatic cell count and high standard plate count. Two ounces of milk is adequate for conducting all tests listed under Step 3. Milk should be collected in sterile 2 ounce snap cap vials or in whirlpack bags, labeled with the correct information and immediately placed on ice.[22] The most frequently observed sampling

Table 1. Benefits and limitations of bulk tank milk analysis.

Benefits	Limitations
1. Provides a logical approach for troubleshooting herds with multiple milk quality and mastitis related problems.	1. Does not provide information about milk quality and mastitis at individual cow level.
2. Less expensive than quarter milk sampling the whole herd.	2. Understanding milk quality and mastitis problems in a herd cannot be done effectively using a single BTM sample.
3. BTM analysis can be done in about 96 hours.	3. Information on herd management practices on milking cows, mastitis prevention, milk sanitation and general farm hygiene are required to interpret BTM analysis results.
4. A reliable tool for veterinarians to troubleshoot milk quality and herd level mastitis.	4. Proper interpretation of BTM milk analysis results is critical before implementing changes on the farm.
5. An important component of total herd health management or veterinary practice consultancy services.	5. BTM samples cannot be frozen, they must be shipped on ice or icepacks.
6. Bulk tank milk analysis report becomes documentary evidence of milk quality assurance protocol practiced on the farm.	6. BTM samples have to be processed within 36 hours of collection.

Table 2. How to conduct a bulk tank milk analysis.

STEP ONE: Identify the issue			**STEP TWO:** Collection of bulk tank milk sample	**STEP THREE:** Laboratory tests		
Issue	**Milk quality**	**Mastitis**		**What laboratory tests need to be done?**	**Milk quality**	**Mastitis**
Low or no premiums?	✓	✓	**When to collect?** At least 1-2 hours after milking, preferably should represent one milking.	Bulk tank milk somatic cell count	✓	✓
Persistent high bacterial counts?	✓	✓	**How to collect?** 1. Seek permission of owner. 2. Agitate milk in the bulk tank for 10 minutes. 3. Wear disposable gloves. 4. Collect 2 ounces of milk from the top of the bulk tank using a clean sanitized dipper. 5. Transfer the milk to a 2 ounce whirlpack bag or 2 ounce snap cap vial. 6. Label the sample (farm, date, and note temp. on the bulk tank).	Standard plate count	✓	✓
Education of milkers	✓	✓		Preliminary incubation count	✓	
Bulk tank somatic cell count >250,000 cells/ml?	✓	✓		Coliform count	✓	✓
More mastitis cases in the last month?		✓	**How many samples to collect?** Establish a bulk tank milk profile: 4- samples (1 sample/week)	*Staphylococcus aureus* count		✓
Buying the whole herd?	✓	✓		*Streptococcus agalactiae* count		✓
Monitor after herd expansion?	✓	✓	**How to transport the sample to laboratory?** Bulk tank milk samples must not be frozen, they should be shipped on ice or icepacks to the laboratory, such that the samples can be processed within 36 hours of collection.	Streptococci and Strep-like organisms		✓
Suspect Mycoplasma in the herd?		✓		Coagulase negative staphylococci		✓
			Information on farm management practices Use the standardized questionnaire to collect information on farm management practices.	Mycoplasma		✓
Monitor fat and protein in milk?	✓			Percent fat and percent protein	✓	

errors include: 1) milk stored in containers not suited for transport, 2) samples improperly labeled, 3) milk samples become too warm, 4) volume of milk is insufficient to conduct all of the tests, and 5) milk samples freeze during transport to the laboratory. Milk samples should arrive in the laboratory within 36 hours of collection.[23] Before conducting a BTM analysis, it is important that the veterinarian or dairy health professional communicate with the laboratory, and inquire if the tests listed under Step 3 (Table 2) can be performed by the laboratory.

Laboratory Tests

Tests necessary to assess milk quality and udder health status of the herd are listed in Table 2 and are described briefly as follows:

1. Bulk tank somatic cell counts

Bulk tank somatic cell counts (BTSCC) are valuable indicators of udder health and milk quality. Monitoring udder health status of a herd by BTSCC is very useful, especially in herds with contagious mastitis and herds experiencing clinical outbreaks due to environmental pathogens.[10,22] Milk from uninfected quarters generally has BTSCC less than 250,000 cells/ml.[28] Although no specific somatic cell count (SCC) minimum can be used for detection of an infection, the probability that an infection is present increases as the SCC increases. Research has shown that factors such as stage of lactation or age generally do not result in significant increases in SCC above 250,000 cells/ml if the gland is uninfected.[21] Most laboratories that conduct bacteriological milk quality analysis also test for somatic cells. Laboratories that handle large volumes of milk samples use automated electronic cell counters to estimate the number of somatic cells present in BTM. The dairy producer or the attending veterinarian can send bulk tank milk samples (~ 20 ml) to the Dairy Herd Improvement Association and request that somatic cells be counted. This information is very useful for evaluating the udder health status of the herd.[22]

2. Tests for milk quality

The procedures for conducting bacteriological tests for milk quality are described in the Standard Methods for Examination of Milk and Milk Products.[9,16,23,34] These procedures are industry accepted procedures, and all approved laboratories must follow the methods described. Bacteriological tests for milk quality include:

A. The Standard Plate Count (SPC) is the most commonly used test and is required by the FDA and the state regulatory agencies. The SPC is an estimate of the total aerobic bacteria present in milk. Milk samples are plated on a plate count agar and incubated for 48 hours at 90°F (32°C), after which bacterial colonies are counted and the counts expressed as the number of colony forming units (cfu) per milliliter (ml).

B. The Preliminary Incubation Count (PIC) is a test to estimate the number of pyschrotrophic (cold-loving) bacteria. The results of this test gives an indication of on-farm sanitation, holding temperature of milk in the bulk tank and is a general reflection of milk production practices on the farm. The test is done by holding milk at 55°F (12.8°C) for 18 hours, after which bacteria are enumerated using SPC.

C. The Laboratory Pasteurization Count (LPC), also known as the thermoduric count, is an estimate of the number of bacteria that can survive laboratory pasteurization at 143°F (62.8°C) for 30 minutes. This process destroys most of the mastitis causing pathogens, selecting for those bacteria that can survive pasteurization temperatures (thermoduric bacteria). This is not a regulatory test required by state or federal agencies, however, many progressive milk processors perform this test to ensure quality of the final product. Bacteria not killed by pasteurization are enumerated using the SPC technique.

D. The Coliform Count (CC) is a test that estimates the number of bacteria that originate from manure or a contaminated environment. Milk samples are plated on MacConkey's agar and incubated for 48 hours at 90°F (32°C), after which typical coliform colonies are counted. The counts are expressed as the number of cfu per ml.

3. Tests for isolation and identification of mastitis causing bacteria

The National Mastitis Council, after critical evaluation of peer reviewed research articles, has recommended isolation and identification procedures for mastitis causing bacteria. These procedures have been described by Hogan and co-workers in the laboratory handbook on bovine mastitis.[22] Briefly, 0.01 ml of milk is streaked vertically across the diameter of an agar plate. The inoculum is then evenly spread over the entire surface of the plate by a back and forth motion at right angles to the central streak, using the same loop that was used for inoculation. Milk samples are plated on MacConkey's agar to detect coliforms and gram-negative bacteria, modified Edward's media for Streptococci and Streptococci-like organisms, Vogel Johnson agar for Staphylococci and Modified Hayflick's medium for Mycoplasma organisms. Plates are incubated at 98.6°F (37°C) for 48 hours. Mycoplasma medium is incubated under modified atmospheric conditions.

Interpretation of Tests Used to Assess BTM Quality

Bulk tank milk SPC of < 1000 cfu /ml is an indication that milk from clean and healthy cows has been collected under hygienic conditions. Under current conditions it is extremely difficult to totally prevent contamination of milk, but SPC counts of less than 5,000 cfu/ml can be achieved. Realistically, SPC of < 10,000 can be achieved by most farms. High SPC in raw milk can be due to improper cleaning of the milking system or presence of *Streptococcus agalactiae* mastitis infection in a herd (Table 3a). Milking cows with soiled udders and teats or mastitis caused by environmental streptococci or coagulase negative staphylococci, unclean or unsanitized milking equipment, and the inability to cool milk rapidly to less than 40°F (4.4°C) can increase the SPC of raw milk.[31,32]

A laboratory pasteurization count of > 200 cfu/ml is considered high (Table 3a). High LPC is most often seen with persistent cleaning problems, leaky pumps, old pipe line gaskets, inflations and other rubber parts, and milkstone deposits. Significant levels of contamination from soiled cows can also contribute to high LPC.[2,31]

Preliminary incubation counts are generally higher than SPCs. A 3-4 fold higher PIC count than the SPC is suggestive of potential problems related to cleaning and sanitation of the milking system or poor udder preparation before milking. Failure to cool milk rapidly, marginal cooling or prolonged storage times may also result in high PIC (Table 3b). A PIC equal or slightly higher than a high SPC (> 50,000 cfu/ml) may suggest that the high SPC is possibly due to mastitis.[2,13,31]

Coliform counts reflect the hygiene and sanitation practices followed on the farm. Interpretation of CC is shown in Table 3b. Coliform counts > 50 cfu/ml suggest poor milking practices, dirty equipment, contaminated water, dirty milking facilities or cows with subclinical or clinical coliform mastitis.[2,31,32]

Interpretation of Tests for Assessing Udder Health Status of a Dairy Herd

Bulk tank milk analysis for BTSCC along with BTM culturing for mastitis pathogens can be used effectively to monitor herd udder health status.

Table 3a. Interpretation of raw milk bacterial counts and suggested guidelines for troubleshooting high counts.

Test and suggested counts	If counts are high for 3 out of 4 samples then the likely problem can be:	What to look for on the farm?
Standard Plate Count (SPC)	1. Improper cooling of milk	1a. Temperature of milk 2 h after milking (must be ≤40°F) 1b. Check the bulk tank temperature indicator/ thermometer for accuracy 1c. Ask if any recent changes have been made to cooling system
Low (Good) < 5,000 cfu/ml		
Medium (Acceptable) < 10,000 cfu/ml	2. Poor milking practices	2a. Evaluate udder preparation and milking procedures (See Table 7)
High (Concern) > 10,000 cfu/ml	3. Unclean or unsanitized milking equipment	3a. Check for detergent and or sanitizer left in the containers 3b. Check water temp used for cleaning 3c. Enquire about water quality (chlorination, well water management, coliform count, pH, water hardness) 3d. Ask if equipment is sanitized between milkings 3e. If pails and buckets are used, ask how they are cleaned
	4. Mastitis	4a. Check for history for *Streptococcus agalactiae* mastitis 4b. Check for cows with subclinical mastitis (perform California Mastitis Test)
Lab. Pasteurization Count (LPC)	1. Unclean milking equipment and utensils	1a. Persistent cleaning failure in some area of the milking system 1b. Same as SPC (3a-3e)
Low (Good) < 100 cfu/ml	2. Faulty milking machine and worn out parts	2a. Check for leaky pumps, old pipeline gaskets, inflations and other rubber parts, and milk stone deposits 2b. Check air lines and moisture traps
Medium (Acceptable) 100 - 200 cfu/ml		
High (Concern) > 200 cfu/ml	3. Extremely dirty cows	3a. Check for soiled udder and teats at time of milking. Determine if udders are flamed singed or clipped

Bulk tank somatic cell counts

Bulk tank somatic cell counts in milk are comprised mostly of leucocytes that enter the udder primarily to destroy mastitis causing bacteria and to repair damaged udder tissue.[33] All milk samples will contain some somatic cells, however their numbers increase considerably when the udder is infected, or there is trauma to the udder. Injured mammary tissue and a high number of somatic cells following an intramammary infection can clog the tiny milk ducts in the udder, which in turn results in lowered milk secretion and production. The number of somatic cells vary significantly among both infected and uninfected cows.[33] Variation in SCC is also influenced by the stage of lactation, season of the year and individual cow responses to infection.[21] *Considering these factors, BTSCC should only be used as a guideline to indicate the overall udder health of a dairy herd.* Persistent elevated BTSCC from at least 4 samples taken over a 4-week period should be used to determine the udder health status of the herd (Table 2). Research done on herds in the Quinte

Table 3b. Interpretation of raw milk bacterial counts and suggested guidelines for troubleshooting high counts.

Test and suggested counts	If counts are high for 3 out 4 samples then the likely problem can be:	What to look for on the farm ?
Preliminary Incubation Count (PIC) **Low (Good)** < 10,000 cfu/ml **Medium (Acceptable)** 10,000 - 50,000 cfu/ml OR < 3x to 4x SPC **High (Concern)** > 50,000 cfu/ml OR > 3x to 4x SPC	1. Unclean milking equipment and utensils 2. Marginal cooling of milk 3. Poor udder preparation before milking	1a. Same as SPC (3a- 3e) 2a. Check temperature of milk in the bulk tank 2 hours after milking 3a. How are the cows cleaned and sanitized before milking? 1. Use of an approved pre-dip? 2. Teats dipped using a dip cup or spray? 3. Are cows fore-stripped ? 4. Individual paper or cloth towels? 5. Teat and teat ends thoroughly clean and dry before attaching the milking unit? 6. Use of an approved post-dip?
Coliform Counts (CC) **Low (Good)** < 10 cfu/ml **Medium (Acceptable)** 10 - 50 cfu/ml **High (Concern)** > 50 cfu/ml	1. Poor udder preparation before milking 2. Herd history with regard to coliform mastitis	1a. Same as PIC (3a) 1b. Does the claw fall in manure during milking? 1c. Look for wet udders during milking 1d Look for worn rubber hoses and gaskets 1e. Check milk filter after milking for fecal matter 2a. Inquire if herd has had many cases of clinical coliform mastitis? If yes, 1. What kind of bedding is used (sawdust, dried manure, washed sand)? 2. How is the bedding managed? 3. How frequently is it replaced? 4. Freestall area is wet all the time, 4-6 inches of manure in the alleys? (Socks-like appearance on the feet of cows)

area of Ontario, Canada indicated that the ability of bulk tank counts to predict the quarter infection rate of herds was approximately doubled (45.5% versus 80%) when the interpretation was based on 6 previous monthly bulk tank samples instead of a single test.[33]

Bulk tank somatic cell counts allow practitioners to assess the overall udder health status of a dairy herd. A dairy herd that has an excellent udder health program in place will have the BTSCC and incidence of mastitis as shown in Table 4. An estimate of milk production based on the BTSCC is shown in Table 5. Based on this table, herds with somatic cell counts over 500,000 could be producing from 6 to 29% below potential because of the presence of sub-clinical mastitis infections. This clearly suggests that owners or managers of herds with over 500,000 cells/ml of BTM should be concerned about herd udder health and should initiate mastitis control and prevention practices.[21,33] Several useful guides on interpreting BTSCC and individual cow SCC are available.[1,8,10,28,33]

Mastitis causing bacteria in BTM

Mastitis causing organisms in BTM can be classified into one of the two groups, contagious and environmental. The most common contagious organisms are *Staphylococcus aureus*, *Streptococcus agalactiae* and Mycoplasma species. The environmental organisms are coagulase negative Staphylococci, Streptococci and Streptococci-like organisms, coliforms and gram-negative non-coliform bacteria. Several useful guides on interpreting BTM mastitis culture results and

Table 4. Criteria that define excellent udder health status of individual cows and the herd.

Criteria	Ideal udder health targets
Bulk Milk Somatic Cell Count	< 250,000 cells/ml
Herd average (actual)	< 200,000 SCC
Herd average (DHI Linear Score)	< 3.0 LS SCC
100% of first calvers (DHI)	< 100,000 SCC
> 85% of herd	< 200,000 SCC
> 95% of herd	< 500,000 SCC
Incidence of clinical mastitis	< 25 cases /100 cows per year
Number of culls due to mastitis or other udder health problems	< 5 cases/ 100 cows a year

Taken from Leslie [28]

Table 5. Estimates of percent infected quarters and losses in milk production due to elevated BTSCC

BTSCC/ml	Percent quarters infected	Percent production loss
200,000	6	0
500,000	16	6
1,000,000	32	18
1,500,000	48	29

Taken from Harmon [21]

implementing mastitis control programs are available.[1,5,15,17] Tables 6a and 6b show comprehensive guidelines for interpreting counts of mastitis causing bacteria, and control measures that can be implemented to lower the incidence of mastitis in the herd.

Contagious mastitis pathogens

The primary habitat of *Staphylococcus aureus* is the infected udder. It readily colonizes the skin of the teats and teat ends when there is damage to the skin surface (chapped, frostbite, cuts, scabs, warts). *S. aureus* infections are usually chronic or subclinical, occasionally showing mild clinical signs.[22] *S. aureus* has been shown to produce extracellular enzymes that allow it to penetrate deep into the mammary tissue. Abscesses that form as a result of the infection are very difficult to treat with routine antimicrobial therapy, thus making it a very challenging organism to control in a dairy herd. A large majority of newly infected animals (~ 70%) do not show signs of clinical mastitis, however cows that are chronically infected show signs of clinical mastitis.[10] A guide for interpreting *S. aureus* counts is shown in Table 6a. The presence of *S. aureus* in successive BTM samples is a good indicator that cows with *S. aureus* infection are present in the herds. However, when implementing a mastitis control program for *S. aureus*, BTM analysis has been shown to be less useful, as this organism is shed infrequently and in low numbers, making it difficult to monitor *S. aureus* with current isolation and detection protocols. The general history of a herd experiencing a problem with *S. aureus* is an elevated BTSCC of 500,000 to 600,000 (range of 350,000 - 1 million), depending on the number of animals infected.[10] The type of herd (closed or open) and milking practices followed on the farm influence the persistence and spread of *S. aureus* in the herd. Measures for controlling *S. aureus* infection in a herd have been listed in Table 6a.

Streptococcus agalactiae is a highly contagious pathogen, and very quickly spreads through the herd. Cow to cow transmission at the time of milking is responsible for the rapid spread of the organism. Unlike *S. aureus*, *S. agalactiae* are shed in large numbers from infected quarters and can be easily cultured from BTM. A guide for interpreting *S. agalactiae* counts in BTM is shown in Table 6a. The most frequent method of introducing *S. agalactiae* into a clean herd is by purchasing adult cows without prior testing. Herds with cows that have *S. agalactiae* infections should be considered as possibly having poor mastitis prevention and control practices.

Mycoplasma intramammary infections in a herd also can cause an increase in BTSCC count. The presence of Mycoplasma in a herd can be detected by bulk tank culture, even if there are very few cows with Mycoplasma mastitis. A guide for interpreting the presence of Mycoplasma in BTM is shown in Table 6b. Herds that have numerous cows with Mycoplasma mastitis generally have either a previous history of Mycoplasma pneumonia in the herd, or a high number of calves with respiratory diseases. Another characteristic feature of Mycoplasma mastitis is that it does not respond to conventional antibiotic therapy. Further, the infected quarter becomes progressively worse, and in most instances the infection spreads to other quarters of the same cow even when she has been treated. Aggressive culturing and culling is necessary to reduce the Mycoplasma mastitis rate in a herd.[22]

Corynebacterium bovis is a highly contagious organism, and weakly pathogenic in nature. Intramammary infections are usually subclinical and rarely cause clinical mastitis. Reports suggest that cows with *C. bovis* infection generally have lower milk production. In a herd where many cows are infected with *C. bovis*, an elevated BTSCC is commonplace. *Corynebacterium bovis* is a resident of the teat canal, and has a particular affinity to keratin present in the teat canal. Fore-stripping of cows before milking can considerably reduce the risk of *C. bovis* infection in the herd.[22] A guide for interpreting *C. bovis* counts is shown in Table 6b. Effective teat-dipping and

Table 6a. Interpretation of results for contagious mastitis pathogens in bulk tank milk.

Mastitis causing bacteria and suggested counts	If counts are high for 3 out of 4 samples then check for the following:	Suggested control measures
Staphylococcus aureus **Low** < 1 cfu/ml **Medium** 100 - 500 cfu/ml **High** > 500 cfu/ml	1. **Type of herd:** closed herd, suggests the presence of chronic infection; open herd, suggests the likelihood of newly purchased animals as one of the possible source of *Staph. aureus*. 2. **BTSCC** in a herd with high *Staph. aureus* infection generally ranges from 350,000 - 1000,000 cells/ ml (most occasions 500,000-600,000 cells/ml). 3. **Management practices** that allow spread of *Staph. aureus* in the herd: 1. Milking cows without gloves 2. Cloth towels reused without proper cleaning 3. Milking infected cows along with uninfected cows 4. Poor fly control during summer 5. During winter, milking cows with chapped teats 6. Milking cows with teat and teat end injuries	1. CMT all newly purchased animals, and cows in milk with high SCC (> 250,000 cells/ml) 2. Detect cows early with *Staph. aureus* mastitis by doing milk culture testing 3. Milk all infected cows last 4. Post milking teat-dip cows 5. Dry-cow therapy 6. Use of individual paper towel or cloth towel 7. Back flush milking units using a sanitizing solution 8. Cull cows with chronic infection 9. Segregate herd if possible
Streptococcus agalactiae **Low** < 1 cfu/ml **Medium** 1000 - 5000 cfu/ml **High** > 6000 cfu/ml	1. **Type of herd:** closed herd, suggests presence of chronic infection; open herd, suggests both the likelihood of newly purchased animals bringing in the infection. 2. **BTSCC** in a herd with high *Strep. agalactiae* infection generally ranges from 500,000-600,000 cells/ml, with high SPC (50,000 to > 100,000 cfu/ml). 3. **Management practices** that allow spread of *Strep. agalactiae* in the herd: 1. Milking cows without gloves 2. Cloth towels reused without proper cleaning 3. Milking infected cows along with uninfected cows 4. No or inadequate teat-dipping practices	1. CMT all newly purchased animals, and cows with high SCC (> 250,000 cells/ml) 2. Detect cows early with *Strep. agalactiae* mastitis by miik culture 3. Teat-dip cows 4. Dry-cow therapy 5. Use of individual paper towels 6. Don't feed *Strep. agalactiae* containing milk to calves 7. Back flush milking units using a sanitizing solution 8. Milk all infected cows last

milk hygiene practices can be used effectively to reduce the incidence of *C. bovis* mastitis.

Environmental mastitis pathogens

Bacterial species of environmental origin found in BTM are listed in Table 6c. Bacteria of environmental origin that cause mastitis can be placed into four categories: 1) Streptococci and Streptococci-like organisms (SSLO), 2) coagulase negative Staphylococci (CNS), 3) coliforms, and 4) gram-negative non-coliforms.

Streptococci and Streptococci-like organisms (SSLO) consist of a large heterogeneous group of organisms. Current isolation methods and presumptive biochemical and enzymatic tests do not allow clear differentiation of any one specific genera, therefore they are referred to collectively as SSLO. Organisms belonging to the genera Streptococci, Enterococci, Lactococci and Aerococci have been isolated previously from BTM. The predominant mastitis causing environmental streptococci consist of *Streptococcus uberis* and *Streptococcus dysgalactiae*. *Streptococcus uberis* is a common cause of mastitis in cows during early lactation and the dry period. Cows are infected with *S. uberis* from environmental sources. *Streptococcus dysgalactiae* is found in infected udders and the dairy environment. It is spread from cow to cow during milking or from environmental

Table 6b. Interpretation of results for contagious mastitis pathogens in bulk tank milk.

Mastitis causing bacteria and suggested counts	If counts are high for 3 out of 4 samples then look for the following:	Suggested control measures
Mycoplasma	1. **Type of herd:** closed herd, suggests the presence of chronic infections in the herd that would include animals of all ages; open herd, suggests the likelihood of newly purchased animals as one of the possible sources of Mycoplasma, which is the most frequent cause of a Mycoplasma outbreak in a herd that has expanded recently. 2. **BTSCC** is generally > 500,000 cells/ ml when there are more than 5 to 10% of the cows with Mycoplasma infection. 3. **Management practices:** poor herd health management practices with a history of Mycoplasma pneumonia in the herd including calves; hygroma in adult cattle; cows treated for clinical mastitis do not respond to treatment; cloth towels and cannulas are reused without proper cleaning and disinfection; and the herd has expanded recently.	1. Perform whole herd culture 2. Segregate infected and non-infected cows 3. Attention needs to be given to newly purchased animals 4. Improve milking hygiene 5. Cow to cow transfer of infection can occur while milking, therefore, cows with Mycoplasma IMI should be milked last 6. Milkers should wear rubber gloves 7. Use individual paper towels 8. Cull cows that are infected with Mycoplasma 9. Monitor bulk tank milk monthly for Mycoplasma
Corynebacterium bovis **Low** < 500 cfu/ml **Medium** 500 - 1000 cfu/ml **High** >1000 cfu/ml * *C. bovis* is infrequently found in bulk tank milk, is highly contagious, but less of a pathogen	1. **Type of herd:** little information is available as to the type of herd and the occurrence of *C. bovis* mastitis. It can be presumed to be similar to other contagious bacteria. 2. **BTSCC** do not generally increase, but when there are many cows with *C. bovis* subclinical mastitis the BTSCC may be around 350,000 - 500,000 cells/ml. 3. **Management practices:** when teat-dipping and dry-cow therapy are not practiced or improperly implemented, *C. bovis* is frequently isolated. Teat-end lesions can cause *C. bovis* to colonize and allow their spread in the herd.	1. All cows should be pre-stripped before milking 2. Evaluate teat-dipping practices 3. All cows to be dried off, should be dry-cow treated 4. Treat cows for teat end lesions

sources. Depending on the rate of infection in the herd, BTSCC is frequently elevated, ranging from 250,000 to 450,000 cells/ml.[10] In herds with persistently high (>1000 cfu/ml) counts of SSLO in BTM, a higher incidence of clinical and subclinical mastitis due to *S. uberis* and or *S. dysgalactiae* can be expected.

The CNS are normal residents of the skin surface. The CNS reported to cause mastitis are listed in Table 6c. The CNS are an opportunistic group of bacteria, which gain access into the teat canal and the gland from skin sources. The infection is usually mild and transient in nature, however clinical mastitis due to CNS has been widely reported in literature. In herds where CNS is the predominant mastitis causing bacteria, the BTSCC frequently ranges from 350,000 to 500,000 cells/ml.[10]

Coliform organisms include *Escherichia coli*, Klebsiella spp., Enterobacter spp. and Citrobacter spp. These environmental organisms are frequently isolated from bulk tank milk.[24] A high proportion of new infections with coliforms occur approximately 2 weeks before and 2 weeks after drying off. During lactation susceptibility to infection is highest at calving and decreases considerably as lactation progresses. During hot and humid weather conditions, cows are at higher risk of developing coliform mastitis.[22] Herds with a high incidence of

Table 6c. Interpretation of results for environmental origin bacteria in bulk tank milk.

	If counts are high (> 1000 bacteria/ml) for 3 out of 4 samples then the following corrective actions can be done:	
Bacteria and their counts	**Most frequently observed problems**	**Corrective actions**
1. Streptococci and Strep-like organisms (SSLO) 2. Coagulase negative Staphylococci (CNS) 3. Coliforms (*E. coli,* Klebsiella, Enterobacter, Citrobacter) 4. Non-coliforms (Pseudomonas, Serratia, Proteus, Acientobacter, Moraxella, Hafnia) **SSLO, CNS, Non-coliforms** **Low** < 500 cfu/ml **Medium** 500 - 1000 cfu/ml **High** > 500 cfu/ml **Coliforms** **Low** < 10 cfu/ml **Medium** 10-50 cfu/ml **High** > 50 cfu/ml * See Table 6d for list of bacterial species isolated from bulk tank milk.	**BTSCC** 1. Streptococci and Strep-like organisms: 250,000-450,000 cells/ml 2. CNS: 350,000-500,000 cells/ml 3. Coliforms: < 300,000 cells/ml 4. Non-coliforms: No data **Milking Procedures** 5. No established milking protocol 6. Milking procedures not consistent (varies from milker to milker and shift to shift) 7. Poor udder surface (clipping or flaming not done) 8. Teat and teat-ends not thoroughly cleaned 9. Milking done on wet teats 10. Cloth towels reused without cleaning **Farm Hygiene** 1. Poor bedding management 2. Manure removal not done regularly	**Milking procedures** (See Table 7 for recommended milking procedures) 1. Pre-dipping and post-dipping practices on the farm (use of approved teat-dip, teat-dip concentration, application method, and application time of pre-dip need to be evaluated) 2. Cleaning teat and teat-ends before milking a. Individual paper towels recommended, if cloth towels are used, clean and sanitized towels should be used only once during the milking operation b. Teat and teat-ends must be thoroughly cleaned, and must be dry before applying the milking unit. Wet and unclean teat and teat-ends increase the risk of mastitis in the herd. **Udder health** 1. Perform California Mastitis Test on a regular basis for cows in early (by 6th milking) and late lactation (0-3 days before drying off) 2. Dry-treat all cows before drying off 3. Frequently examine teat and teat-end condition. Cows with bruised teats and teat-end injuries are more likely to get infected with mastitis causing bacteria. **Milking machine** Milking machine needs to be inspected periodically by a professional. Attention needs to be given to liner slips and vacuum levels. Excessive liner slips and faulty vacuum levels can predispose cows to mastitis. **Bedding in tie or free stalls** Wet and dirty bedding harbor the environmental bacteria and between milkings can get inside the teat canal, or on the surface of teat and teat-ends. Advocate dry and clean bedding that is worked on at least twice a day.

coliform mastitis usually have a BTSCC that is < 200,000 cells/ml.[10] The BTSCC does not increase as most milk from cows with coliform mastitis is discarded due to the nature of the secretions.

Gram-negative non-coliform organisms, such as *Pseudomonas* and *Serratia,* can cause severe mastitis, including outbreaks of clinical mastitis.[12,26] These organisms have been isolated from BTM.[24] The interaction between these organisms and the mammary gland, the BTSCC following a herd infection, and the number of organisms shed in milk are not fully characterized.

The most frequently observed problems related to high counts (> 1000 cfu/ml) from successive bulk tank samples include: 1) absence of an established milking

Table 6d. Bacterial species isolated from bulk tank milk.

Group of bacteria	Species (species indicated in bold associated with subclinical and clinical mastitis)
Coagulase-negative Staphylococci	*S. caprae,* ***S. chromogenes, S. cohnii,*** *S. epidermidis, S. hominis,* ***S. hyicus, S. intermedius, S. lentus, S. simulans, S. warneri, S. xylosus***
Streptococci and Strep-like organisms	*S. mutans, S. salivarius S. alactolyticus,* ***S. bovis, S. equinus, S. uberis, E. faecalis, E. faecium,*** *E. saachrolyticus, E. avium, E. durans, E. hirae, E. malodoratus,* ***S. agalactiae, S. dysgalactiae, S. equi, S. equisimilis, S. zooepidemicus,*** *S. downei, S. acidominimus, S. thermophilus.*
Coliforms	***Escherichia coli,*** *Citrobacter fruendii,* **Klebsiella spp,** Enterobacter spp.
Gram-negative non-coliform bacteria	**Acinetobacter, Hafnia,** Moraxella, **Pseudomonas, Serratia**

protocol, 2) poor udder condition and health, and 3) deficient management practices related to general farm hygiene and stall maintenance (Table 6c). Suggested corrective actions includes establishing a milking protocol that is practiced consistently. A recommended milking practice is described in Table 7. Udder health must be monitored periodically by: 1) doing the California Mastitis Test, 2) examining udder and teat-end condition, 3) giving extra attention to cows that are in early and late lactation and 4) dry-treating all cows at dry-off. When practiced consistently and over a period of time, these practices help to improve herd udder health (Table 6c). Improving overall farm hygiene and implementing proper bedding management practices is critical to the overall goal of reducing the incidence of environmental mastitis in the herd (Table 6c).

Putting it all Together

The SPC, PIC, and BTSCC, in combination with other milk quality measures, and contagious and environmental mastitis counts can be used effectively to identify current and potential milk quality and mastitis problems in a herd. A scheme for identifying current and potential milk quality and mastitis problems in a herd is shown in Figure 1. In this figure, the first step is to determine if the SPC counts, are > 5,000 cfu/ml. If so, the second step is to determine if the PIC counts are

Table 7. Recommended milking procedures in stanchion/tie stall or parlors.

Stanchion / Tie stall		Parlor	
Step	**Operation**	**Step**	**Operation**
1	Wear gloves	1	Wear gloves
2	Wipe off excess dry manure, straw and bedding	2	Wipe off excess dry manure, straw and bedding
3	Strip each teat into a stripcup	3	Strip each teat into a stripcup
4	Dip teats with an approved pre-dip. Allow the pre-dip to react for at least 30 sec.	4	Dip teats 3-4 cows with an approved pre-dip. Allow the pre-dip to react for at least 30 sec.
5	Clean teat and teat-ends using single paper towel or individual cloth towel	5	Return to the first cow and clean teat and teat ends using a single paper towel or individual cloth towel
6	The teats must be dried for at least 15 sec.	6	The teats must be dried for at least 15 sec
7	Attach milking machines immediately after teats are dried	7	Attach milking machines immediately after teats are dried
8	Dip teats with post-dip immediately after milking	8	Dip teats with post-dip immediately after milking

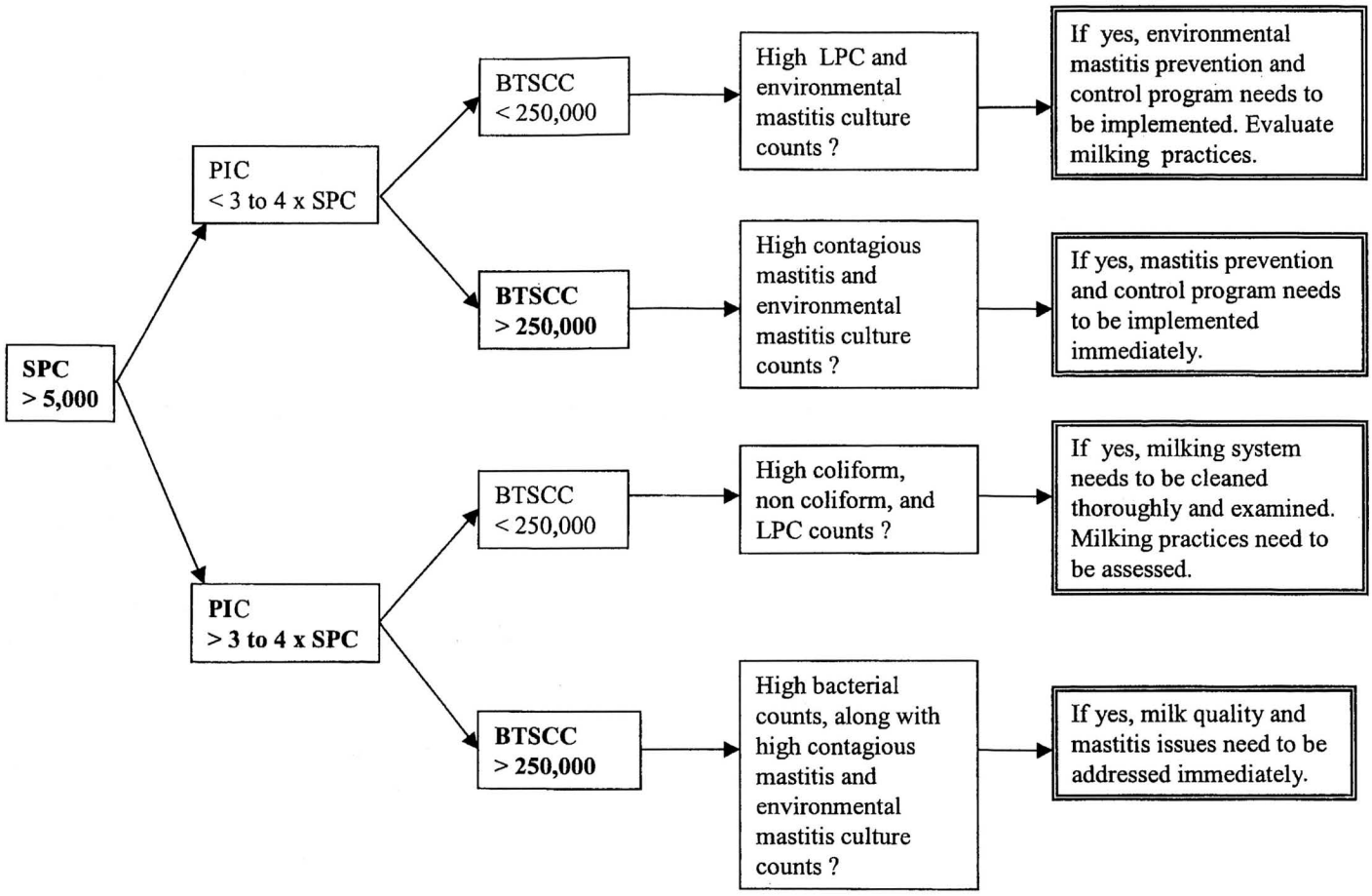

Figure 1. A schematic diagram for identifying current and potential milk quality and mastitis problems in a herd.

> or < 3 to 4 x the SPC. The third step is to link this finding with the BTSCC (> or < 250,000 cells/ml). This information, when used with other milk quality measures and mastitis culture counts, aids in identification of likely problems in the herd.

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References

1. Bagley CV: Helping dairy producers reduce SCC. http://www.ext.usu.edu/ag/dairy/factsht/factindx.htm.
2. Bramley AJ, McKinnon CH, Staker, RT, Simpkin DL: The effect of udder infection on the bacterial flora of the bulk milk of ten dairies. *J Appl Bacteriol* 57:317-323, 1984.
3. Bramley AM, McKinnon CH: The microbiology of raw milk. in Robinson RK (ed.): *Dairy Microbiology,* Vol 1. London, Elsevier Science Publishers, 1990, pp 163-208.
4. Bray DR, Shearer JK: Trouble-shooting a mastitis problem herd. University of Florida Cooperative Extension Circular 1162. *Dep Dairy Poultry Sci.,* Florida Coop Ext Serv, Inst Food Agric Sci Univ Florida, Gainesville, 1996.
5. Britt JS, Hartmann F, Reinemann D: Use of microbiology and strategic site sampling at strategic times to solve high bacteria count problems in bulk tank milk, "An actual case history". *Proc 36th Natl Mastitis Council* 80- 90, 1997.
6. Britten AM, Emerson T: A bulk tank culturing program for monitoring milk quality and udder health. *Proc 35th Natl Mastitis Council* 149-150, 1996.
7. Britten AM: Is Strep mastitis causing high bacteria counts in your bulk tank? *Proc 37th Natl Mastitis Council* 26-34, 1998.
8. Chambers JV, Hockney MD, Dillon WM: Making leucocytes count - a guide to identifying and controlling mastitis problems in a dairy herd. http://hermes.ecn.perdue.edu/agcomm/Pubs/DH/DH-127.htm.
9. Christen GL, Davidson PM, McAllister JS, Roth LA: Coliform and other indicator bacteria. in Marshall RT (ed): Standard Methods for Examination of Dairy Products. 16th ed, Washington DC, APHA, 1992, pp 247-269.
10. Corbett RB: The use of somatic cell counts in mastitis management. *Proc 37th Natl Mastitis Council* 51-55,1998.
11. Emerson T: Bulk tank milk bacterial culturing - an aid to quality milk production. *Proc 28th Natl Mastitis Council* 49-53,1989.
12. Erskine RJ, Unflat JG, Eberhart RJ: Pseudomonas mastitis: difficulties in detection and elimination from contaminated wash-water systems. *JAVMA* 191:811-815, 1987.
13. Fenlon DR, Logue DN, Gunn J, Wilson J: A study of mastitis bacteria and herd management practices to identify their relationship to high somatic cell counts in bulk tank milk. *Brit Vet J* 151:17-25, 1995.

14. Farnsworth JR: The current status of the use of bulk tank milk cultures in milk quality and mastitis control procedures. *Agri Pract* 13:5-8, 1992.
15. Fransworth JR: Microbiological examination of bulk tank milk, in Anderson KL, (ed): Update on bovine mastitis, *Vet Clin North Am Food Anim Pract* 9:469-474, 1993.
16. Frank JL, Christen Gl, Bullerman LB: Test for groups of microorganisms, in Marshall RT (ed): Standard Methods for Examination of Dairy Products. 16th ed, Washington DC, *APHA,* 1992, pp 271-286.
17. Gangwer M, Gamroth M, Hansen D: Interpreting bulk tank milk culture. http://www.inform.umd.edu/EdRes/Topic/Agr/interpreting_bulk_milk_culture.html.
18. Gehringer G: Multiplication of bacteria during farm storage, in Factors influencing the bacteriological quality of raw milk. IDF Bulletin No. 120, 1980.
19. Godkin MA, Leslie KE: Culture of bulk tank as a screening test: a brief review. *Can Vet J* 34:601-605, 1993.
20. Guterbock WM, Blackmer PE: Veterinary interpretation of bulk tank milk. *Vet Clin North Am Food Anim Pract* 6:257-268, 1984.
21. Harmon RJ: Somatic cell counts: myths vs reality. *Proc 37th Natl Mastitis Council* 40-50, 1998.
22. Hogan JS, Gonzalez RN, Harmon RJ, *et al*: *Laboratory Handbook on Bovine Mastitis*. Madison, WI, National Mastitis Council, Inc. 1999, pp 1-188.
23. Houghtby GA, Maturin LJ, Koeing EK: Microbiological count methods, in Marshall RT (ed): *Standard Methods for Examination of Dairy Products*. 16th ed, Washington DC, APHA, 1992, pp 213-246.
24. Jayarao B M, Wang L: A study on the prevalence of coliforms and non-coliforms in bulk tank milk. *J Dairy Sci* 82:2620-2624, 1999.
25. Jeffrey DC, Wilson J: Effect of mastitis-related bacteria on the total bacteria counts of bulk tank milk supplies. *J Dairy Soc Technol* 40:23-26, 1987.
26. Kamarudin MI, Fox LK, Gaskins CT, Gay JM: Environmental reservoirs for *Serratia marcescens* intramammary infections in dairy cows. *JAVMA* 208:555-558, 1996.
27. Keeter A: Udder health management in large dairy herd - maintaining control. *Proc 36th Natl Mastitis Council* 140-144, 1997.
28. Leslie KE: Somatic cells counts: interpretation for individual cows. http://www.gov.on.ca/OMAFRA/english/ livestock/dairy/facts/84-12.htm. 1984.
29. McKinnon CH, Rowlands GJ, Bramley AJ: The effect of udder preparation before milking and contamination from the milking plant on the bacterial numbers in bulk milk of eight dairy herds. *J Dairy Res* 57:307-318, 1990.
30. Mickelson A, Hansen L, Morris N: The impact of environmental mastitis on milk quality in the pacific northwest. *Proc 37th Natl Mastitis Council* 26-34, 1998.
31. Murphy SC: Raw milk bacteria tests: standard plate count, preliminary incubation count, lab, pasteurization count and coliform count—what do they mean for your farm? *Proc Reg Natl Mastitis Council* 34-41, 1997.
32. Reinemann DJ, Mein GA, Bray DR, *et al*: Troubleshooting high bacteria in farm milk. *Proc 36th Natl Mastitis Council* 65-73, 1997.
33. Stiles R, Rodenburg J: Bulk tank somatic cell counts. http://www.gov.on.ca/OMAFRA/ english/ livestock/ dairy/facts/84_031.htm. 1984.
34. White CH, Bishop JR, Morgan DM: Microbiological methods for dairy products. in Marshall RT (ed): *Standard Methods for Examination of Dairy Products*. 16th ed, Washington DC, *APHA,* 1992, pp 287-325.