

A Survey of Foodborne Pathogens in Bulk Tank Milk and Raw Milk Consumption Among Farm Families in Pennsylvania

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ABSTRACT

A 2-part study was conducted to determine the risk of exposure to human pathogens from raw milk. The first part of the study focused on determining raw milk consumption habits of dairy producers. A total of 248 dairy producers from 16 counties in Pennsylvania were surveyed. Overall, 105 (42.3%) of the 248 dairy producers consumed raw milk and 170 (68.5%) of the 248 dairy producers were aware of foodborne pathogens in raw milk. Dairy producers who were not aware of foodborne pathogens in raw milk were 2-fold more likely to consume raw milk compared with dairy producers who were aware of foodborne pathogens. The majority of dairy producers who consumed raw milk indicated that taste (72%) and convenience (60%) were the primary factors for consuming raw milk. Dairy producers who resided on the dairy farm were nearly 3-fold more likely to consume raw milk compared with those who lived elsewhere. In the second part of the study, bulk tank milk from the 248 participating dairy herds was examined for foodborne pathogens. *Campylobacter jejuni* (2%), Shiga toxin-producing *Escherichia coli* (2.4%), *Listeria monocytogenes* (2.8%), *Salmonella* (6%), and *Yersinia enterocolitica* (1.2%) were detected in the milk samples. *Salmonella* isolates were identified as *S. enterica* serotype Typhimurium (n = 10) and *S. enterica* serotype Newport (n = 5). Of the 248 bulk tank milk samples, 32 (13%) contained ≥ 1 species of bacterial pathogens. The findings of the study could assist in developing farm community-based educational programs on the risks of consuming raw milk.

Key words: bulk tank milk, dairy, foodborne pathogens, raw milk

INTRODUCTION

Pasteurization of commercially distributed milk has greatly reduced the risk of infection resulting from the

consumption of contaminated milk (Centers for Disease Control and Prevention, 1999; Cohen, 2000). However, a portion of the US population continues to consume raw milk and products made from it, namely, soft cheeses (Rohrbach et al., 1992; Headrick et al., 1997, 1998; Shiferaw et al., 2000; Hegarty et al., 2002). In the United States, consumption of raw bulk tank milk (BTM) is a common practice among farm families. Studies have reported that the most prevalent consumers of raw milk are dairy farm families and dairy farm employees (Shiferaw et al., 2000; Hegarty et al., 2002). A study by Rohrbach et al. (1992) reported that 34.9% of dairy producers in eastern Tennessee and southwest Virginia consumed raw milk. Jayarao and Henning (2001) reported that 60% of dairy producers in eastern South Dakota and western Minnesota consumed raw milk. Among the nonfarming population, a growing number of consumers are claiming that raw milk is healthier and are choosing raw milk over pasteurized milk (Potter et al., 1984; Bren, 2004). In California the sale of raw milk is legal, making the state the largest producer of “certified raw milk” in the United States (Headrick et al., 1997, 1998). Certified raw milk is unpasteurized milk with a total bacterial count below a specified standard, but this is not a guarantee that the milk is free of bacterial pathogens. A survey by Headrick et al. (1997) on raw milk consumption in California showed that 3.2% of the population surveyed consumed raw milk. Elsewhere, a survey of adults from 8 US states showed that less than 2% of the participants drank raw milk (Altekruse et al., 1999b). Although raw milk advocates claim that raw milk is healthier, research has shown no significant difference in the nutritional value of pasteurized and unpasteurized milk (Potter et al., 1984; Centers for Disease Control and Prevention, 1999; Bren, 2004).

Raw milk has been a known vehicle for pathogens for more than 100 yr (Potter et al., 1984; Centers for Disease Control and Prevention, 1999; Gillespie et al., 2003). Outbreaks associated with the consumption of raw milk occur routinely every year. In 1987 the FDA banned the interstate sale of raw milk; however, the sale of raw milk within state boundaries falls under the jurisdiction of each state’s government (Bren, 2004).

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As of 1995, the intrastate sale of raw milk for human consumption was legal in 28 states (Headrick et al., 1998). In Pennsylvania, the sale of raw milk is legal on dairy farms if producers have a permit from the Department of Agriculture (Commonwealth of Pennsylvania, 2005). In many states where the off-farm sale of raw milk is prohibited, people have circumvented the law through "cow-sharing" or "cow-leasing" programs. In such programs, people pay a fee to a farmer to lease a share of a cow in exchange for raw milk (Bren, 2004; Mazurek et al., 2004). Outbreaks of foodborne illness have been linked to raw milk obtained from these cow-leasing programs (Centers for Disease Control and Prevention, 2002). Consumption of certified raw milk has also been the source of outbreaks (Centers for Disease Control and Prevention, 1984a,b). Between 1973 and 1992, raw milk was associated with 46 outbreaks of foodborne illness in the United States, and it is significant to note that 40 (87%) of these outbreaks occurred in states where the intrastate sale of raw milk was legal at the time (Headrick et al., 1998). Consumption of raw milk is a high-risk behavior and will continue to cause morbidity and mortality until people stop consuming raw milk and raw milk products (Keene, 1999).

The risk of foodborne disease has increased over the last 20 yr (Oliver et al., 2005). Outbreaks of foodborne illnesses following consumption of raw milk and products made from raw milk caused by Shiga toxin-producing *Escherichia coli* (STEC; Keene et al., 1997; Wilson et al., 1998; Proctor and Davis, 2000), *Salmonella* spp. (Reed and Grivetti, 2000; Centers for Disease Control and Prevention, 2003; Mazurek et al., 2004), *Listeria monocytogenes* (Linnan et al., 1988; Centers for Disease Control and Prevention, 2001), and *Campylobacter jejuni* (Evans et al., 1996; Centers for Disease Control and Prevention, 2002; Peterson, 2003) have been reported in recent years. Gillespie et al. (2003) reported that between the years of 1992 and 2000, 52% of foodborne outbreaks in England and Wales were attributed to raw milk. Raw milk and products made from raw milk have been implicated in similar numbers of documented cases of foodborne illness in France (DeBuyser et al., 2001).

Gastroenteritis is the primary condition associated with cases of foodborne illness attributable to raw milk consumption. Although enteritis caused by foodborne pathogens such as STEC and *Salmonella* spp. is usually self-limiting, immunocompromised individuals are at a higher risk of serious illness. Foodborne *C. jejuni* and *Yersinia enterocolitica* illnesses are typically characterized by gastritis and enterocolitis; however, debilitating postinfection immunologic sequelae, including Guillain-Barré syndrome (Altekruse et al., 1999a; Oliver et al., 2005) and reactive arthritis (Schiemann, 1987), are

known to develop in some individuals following an episode of foodborne illness with these pathogens. Unlike other foodborne bacteria, which cause mainly gastritis and enteritis, *L. monocytogenes* causes listeriosis, which is characterized by septicemia and meningitis in humans (Oliver et al., 2005).

Limited epidemiological data exist on raw milk consumption and the corresponding risk of foodborne illness. Furthermore, the long-term effects on human health of continued exposure to milkborne pathogens are unknown. The purpose of this study was to ascertain information on: 1) raw milk consumption practices of dairy producers, and 2) the prevalence of foodborne pathogens in BTM in Pennsylvania.

MATERIALS AND METHODS

Participants

Dairy producers (n = 914) from 51 counties in Pennsylvania were mailed a letter soliciting their participation in the study. A total of 374 dairy producers (271 and 103 respondents on the first and second mailings, respectively) consented to participate in the study. Of the 374 dairy producers who consented to the study, 265 participated in the first part of the study (a self-administered questionnaire survey), and 248 participated in both the first and second parts of the study (the analysis of BTM for foodborne pathogens).

Questionnaire Survey

The self-administered questionnaire survey used by Jayarao and Cassel (1999) was modified to collect information on demographics (income, education, residence, raw milk use by employees, source of milk, knowledge of foodborne pathogens) and on attributes that influence raw milk consumption practices (taste, health, convenience, cost, and other reasons). The responses to each of the questions were entered into a Microsoft Excel spreadsheet. For each question, a test of significance (χ^2 test) was applied to determine the difference in responses to the questions between dairy producers who consumed raw milk and those who did not. Epi-info-2002 (Centers for Disease Control and Prevention, Atlanta, GA), a database and statistical system for epidemiology on microcomputers, was used for performing χ^2 tests and odds ratio analyses. Statistical significance was accepted at $P < 0.05$.

Collection of BTM

Bulk tank milk from the dairy herds (n = 248) was examined for *C. jejuni*, STEC, *L. monocytogenes*, *Salmonella* spp., and *Y. enterocolitica*. A single BTM sample

(approximately 120 mL) was collected in a sterile snap-cap milk collection vial from each of the 248 dairy producers between July 1, 2001, and June 30, 2002. Milk samples were collected by county extension agents, dairy producers, or laboratory personnel following the National Mastitis Council (1999) standards for BTM sample collection and handling. The county extension agent provided on-farm instruction to the dairy producer on BTM collection and handling procedures as described by the National Mastitis Council. Briefly, milk in the bulk tank was agitated before collection, and samples were taken from the top of the bulk tank using a sanitized dipper. Within 36 h of collection, all milk samples from the region were shipped on ice overnight to the laboratory. On receipt of the sample in the laboratory, only those samples that recorded a temperature of $<7^{\circ}\text{C}$ were processed.

Isolation of Foodborne Pathogens from BTM

Isolation of *C. jejuni* from raw milk was performed as described by Hunt et al. (2001). Enriched broth and plates were incubated in a microaerophilic atmosphere that was created by using a gas-generating kit for *Campylobacter* in an anaerobic jar (Oxoid Ltd., Basingstoke, UK). All presumptive *Campylobacter* isolates were identified to genus level as described by Hunt et al. (2001) and then speciated using the API-CAMPY identification kit (BioMérieux, Hazelwood, MO).

Escherichia coli was isolated from raw milk as described by Jayarao and Henning (2001) with the following modifications. Briefly, 10 mL of raw milk was added to 90 mL of trypticase soy broth (Difco Laboratories, Detroit, MI) supplemented with 20 $\mu\text{g}/\text{mL}$ of novobiocin (Sigma Chemical Co., St. Louis, MO) and incubated overnight at 37°C . *Escherichia coli* were isolated by subculturing 50 μL of enriched sample onto MacConkey agar supplemented with 4-methylumbelliferyl-beta glucuronide (Difco Laboratories). The plates were incubated for 48 h at 42°C . At least 5 to 10 fluorescent colonies from MacConkey-methylumbelliferyl-beta glucuronide plates were selected for further characterization. For detection of *E. coli* O157:H7, 50 μL of enriched sample was spread on sorbitol MacConkey agar (Difco Laboratories) supplemented with cefixime (0.05 mg/L) and potassium tellurite (2.5 mg/L; Dynal Inc., Lake Success, NY) and incubated for 48 h at 42°C . At least 510 sorbitol-negative colonies from sorbitol MacConkey agar were tested with O157 antigen by latex agglutination (Unipath Co., Ogdensburg, NY). Isolates identified as belonging to the genus *Escherichia* were screened for the presence of Shiga toxin 1 (*stx*₁) and Shiga toxin 2 (*stx*₂) genes as described by Meng et al.

(1997). Isolates that carried the Shiga toxin genes were speciated using API 20E (BioMérieux).

Listeria monocytogenes was isolated from raw milk as described by Jayarao and Henning (2001). All isolates were examined for Gram's reaction, hemolysis, and the Christie, Atkins, Munch-Petersen reaction on 5% sheep blood agar, catalase production, nitrate reduction, and motility on sulfide-indole production-motility medium (Becton, Dickinson & Co., Cockeysville, MD). All isolates were confirmed to species by use of the API-Listeria identification kit (BioMérieux). Isolates identified as *L. monocytogenes* were serotyped using O antisera (Difco Laboratories).

Isolation of *Salmonella* from raw milk was performed as described by Jayarao and Henning (2001) with modifications. Briefly, 25 mL of milk was added to 225 mL of lactose broth and incubated for 24 h at 37°C . Approximately 0.1 and 1 mL of preenriched samples were transferred to Rappaport-Vassiliadis medium and tetrathionate broth, respectively (Difco Laboratories), followed by 24 h of incubation at 42 and 37°C , respectively. The enrichments were streaked on Hektoen enteric agar (Difco Laboratories) and xylose lysine desoxycholate agar (Unipath Co.) and incubated for 24 h at 35°C . All presumptive *Salmonella* colonies were inoculated on triple sugar iron agar and urease agar (Difco Laboratories) incubated at 37°C for 24 h, and were tested with *Salmonella* polyvalent O antiserum (Difco Laboratories). Organisms that gave typical reactions for *Salmonella* were then speciated using an API-20E identification kit (BioMérieux). Isolates were then tested by seroagglutination using *Salmonella* O group (A-I) antisera (Difco Laboratories).

Yersinia enterocolitica was isolated from raw milk as described by Weagant et al. (2001). Briefly, 10 mL of milk was added to 90 mL of peptone sorbitol bile broth and incubated at 10°C for 10 d. On d 10, the enriched broth was treated with 0.5% KOH and plated on MacConkey's agar and cefsulodin-irgasan-novobiocin agar (Difco Laboratories). After 48 h of incubation at room temperature, all presumptive *Yersinia* colonies were examined on lysine arginine iron agar, Christensen's urea agar, and bile esculin agar (Difco Laboratories). Organisms identified as *Yersinia* were speciated using the API-20E identification kit (BioMérieux). Isolates of *Y. enterocolitica* were presumed to be pathogenic based on a positive reaction to the autoagglutination test described by Laird and Cavanaugh (1980).

Epi-info-2002 (Centers for Disease Control and Prevention, Atlanta, GA), a database and statistical system for epidemiology on microcomputers, was used to perform χ^2 tests and odds ratio analyses on the occurrence of pathogenic bacteria in raw milk in relation to milk

Table 1. Demographic indicators of dairy producers who participated in the study

Indicators	Consume raw milk (%)		Total (%)
	Yes (n = 105)	No (n = 143)	
Income			
<\$10,000	21 (20)	14 (9.8)	35 (14.1)
\$10,000–19,999	29 (27.6)	31 (21.6)	60 (24.2)
\$20,000–34,000	40 (38)	52 (36.3)	92 (37.1)
\$35,000–75,000	16 (15)	33 (23)	49 (19.8)
>\$75,000	9 (8.6)	13 (9)	22 (8.8)
Education			
High school	73 (69.5)	80 (56)	153 (62)
Trade school	7 (6.7)	21 (14.6)	28 (11.3)
College	14 (13.4)	23 (16)	37 (15)
Other	11 (10.4)	19 (13.2)	40 (16.1)
Residence ¹			
On dairy farm	71 (68)	62 (43)	133 (54)
Away from farm	34 (32)	81 (57)	115 (46)
Employees consume raw milk			
Yes	19 (18)	41 (28.6)	60 (24.2)
No	86 (82)	102 (71.4)	188 (76)
Milk source ²			
Own farm	105 (100)	72 (50.4)	177 (71.4)
Grocery store	7 (6.7)	71 (49.6)	78 (31.4)
Aware of foodborne pathogens ³			
Yes	61 (58)	109 (76.2)	170 (68.5)
No	44 (42)	34 (23.7)	78 (31.4)

¹Residence: test of significance, χ^2 (P) = 13.37 (0.0002); odds ratio (confidence interval) = 2.73 (1.46 – 4.78).

²Milk source: test of significance, χ^2 (P) = 53.69 (0.0000); odds ratio (confidence interval) = 14.79 (6.12 – 37.41).

³Aware of pathogens: test of significance, χ^2 (P) = 8.41 (0.0037); odds ratio (confidence interval) = 2.31 (1.29 – 4.15).

consumption practices. Statistical significance was accepted at $P < 0.05$.

RESULTS

A total of 248 (28%) of the 914 dairy producers from 16 counties in Pennsylvania participated in the study. Of the 248 dairy producers surveyed, 105 (42.3%) dairy producers reported that they consumed raw milk. There were no significant differences in the income and educational level between dairy producers who consumed raw milk and those who did not. Dairy producers who resided on the dairy farm premises were nearly 3-fold more likely to consume raw milk than those who did not reside on the dairy farm ($\chi^2 = 13.37$, $P < 0.0005$; Table 1). All of the 105 dairy producers who consumed raw milk obtained the raw milk from their own farms. Over half (68.5%) of the 248 dairy producers surveyed were aware of the fact that raw milk could contain disease-causing bacteria. Dairy producers who were not aware of foodborne pathogens in raw milk were 2-fold more likely to consume raw milk than producers who were aware of foodborne pathogens ($\chi^2 = 8.41$, $P < 0.005$). About 24% of the dairy producers indicated that their employees were allowed to take raw milk that

was produced on the farm (Table 1). The relevance of raw milk attributes (taste, health and nutritional value, convenience, and cost) and their influence on raw milk consumption were evaluated (Table 2). Dairy producers who consumed raw milk indicated that convenience (60%, availability of raw milk) and taste (72%) were the most important attributes.

Farm BTM samples from 248 dairy herds in 16 counties in Pennsylvania were examined for foodborne pathogens. Pathogenic organisms were isolated from 26 (10.5%) of 248 BTM samples. *Campylobacter jejuni*, STEC, *L. monocytogenes*, *Salmonella* spp., and *Y. enterocolitica* were detected in 2.0, 2.4, 2.8, 6.0, and 1.2% of BTM samples, respectively (Table 3). Overall, 32 (13%) of the 248 BTM samples had ≥ 1 bacterial pathogen. *Salmonella* isolates were identified as *S. enterica* serotype Typhimurium ($n = 10$) and *S. enterica* serotype Newport ($n = 5$). All *L. monocytogenes* ($n = 3$) were identified as O antigen type 1. The *stx*₂ gene was present in 5 of the 6 STEC isolates, and 1 strain encoded for the *stx*₁ gene. *Escherichia coli* O157:H7 was not isolated from the BTM samples. Based on an autoagglutination test, we concluded that all *Y. enterocolitica* ($n = 3$) were pathogenic.

Table 2. Reasons cited by farm families for consuming milk

Attributes	Response to raw milk consumption (n = 105)				
	Taste	Health	Convenience	Cost	Other ¹
Taste alone	13				
Health alone		3			
Convenience alone			13		
Cost alone				1	
Other ¹ alone					5
Taste + health	11	11			
Taste + health + convenience	9	9	9		
Taste + health + convenience + cost	25	25	25	25	
Taste + cost	4			4	
Taste + convenience	12		12		
Convenience + cost			3	3	
Other ¹ + taste, health, convenience, or cost	2	1	1	4	6
Total	76	49	63	37	11
%	72	47	60	35	10

¹Safe (2), no chemicals (3), clean (2), refreshing and homemade (4).

In this study, 105 (42.3%) of the 248 dairy producers who participated in the BTM pathogen survey reported that they consumed raw milk. Bacteriologic analysis of BTM showed that 15 (14.2%) of the 105 producers who consumed raw milk had ≥ 1 bacterial pathogen isolated from their bulk tank (Table 3). There was no significant difference in the incidence of pathogenic bacteria in the raw milk of dairy producers who did and did not consume raw milk ($P > 0.05$).

DISCUSSION

In the United States, foodborne illnesses are estimated to cause 76 million illnesses and 5,000 deaths each year (Mead et al., 1999). The FDA banned the interstate sale of raw milk in 1987; however, the intrastate sale of raw milk for human consumption was legal in 28 states as of 1995 (Headrick et al., 1998). Despite

the known association of raw milk with pathogenic organisms, some consumers believe raw milk is of better quality than pasteurized milk (Potter et al. 1984; Bren, 2004). In the United States, more than 500 people became ill in 2001 and 2002 from drinking raw milk or from consuming soft cheeses made from raw milk (Bren, 2004).

Consumption of raw milk is practiced by dairy producers for several reasons. Many farm families consume raw milk simply because it is a traditional practice and less expensive to take milk from the bulk tank than to buy retail pasteurized milk. In this study, 42.3% of dairy producers surveyed reported drinking raw BTM, and reported taste and convenience as the primary reasons for choosing to consume raw milk over retail pasteurized milk. A survey of Irish dairy producers reported that many farmers believed raw milk had higher nutritional value than pasteurized milk (Heg-

Table 3. Prevalence of pathogens in bulk tank milk

Organism	No. (%) of isolates from bulk tank milk (n = 248)	Combination of pathogens		Consume raw milk ¹ (n = 248)	
		Organism(s)	Bulk tanks (n = 248)	Yes (n = 105)	No (n = 143)
<i>Salmonella</i> spp.	15 (6.0)	<i>Salmonella</i>	11	7	4
		+ <i>E. coli</i>	2	1	1
		+ <i>L. monocytogenes</i>	1	1	—
		+ <i>Y. enterocolitica</i>	1	1	—
<i>Listeria monocytogenes</i>	3 (1.2)	<i>L. monocytogenes</i>	2	1	1
<i>Campylobacter jejuni</i>	5 (2.2)	<i>C. jejuni</i>	3	2	1
		+ <i>E. coli</i>	2	1	1
<i>Yersinia enterocolitica</i>	3 (1.2)	<i>Y. enterocolitica</i>	2	—	2
STEC ²	6 (2.4)	<i>E. coli</i>	2	1	1
Total	32 (13%)		26	15	11

¹Consumption of raw milk: test of significance, χ^2 (P) = 2.15 (0.1429); odds ratio (confidence interval) = 2.00 (0.82 – 4.92).

²Shiga toxin-producing *Escherichia coli*.

arty et al., 2002). Multistate surveys on food consumption behaviors have reported that raw milk consumption is most common among men, younger people (ages 18 to 29 yr), people of Hispanic descent, people earning less than \$15,000/yr, and those who live on a farm or in a rural area (Yang et al., 1998; Shiferaw et al., 2000). In our study, dairy producers who resided on the dairy farm premises were nearly 3-fold more likely to consume raw milk than those who resided away from the farm. Previous studies have reported that people with less than a high school education are more likely to consume raw milk than those who have completed high school, suggesting that the level of education may influence raw milk consumption habits (Headrick et al., 1997; Yang et al., 1998; Altekruuse et al., 1999b; Shiferaw et al., 2000). However, we found no significant difference based on education level or income in this study.

Dairy farms are considered reservoirs of many foodborne pathogens, including *Salmonella*, *Listeria*, *Campylobacter*, and STEC (Oliver et al., 2005). The presence of these and other pathogens in BTM is the result of fecal contamination on teats, udder surfaces, and milking machines (Jayarao and Wang, 1999). In this study, *C. jejuni*, STEC, *L. monocytogenes*, *Salmonella* spp., and *Y. enterocolitica* were detected in 2.0, 2.4, 2.8, 6.0, and 1.2% of BTM samples, respectively. Similar isolation rates have been reported in studies of BTM (Rohrbach et al., 1992; Jayarao and Henning, 2001). Van Kessel et al. (2004) reported results on the prevalence of foodborne pathogens in BTM as part of the USDA's National Animal Health Monitoring System Dairy 2002 Survey. They observed a higher isolation rate for *L. monocytogenes* (6.5%) and a lower rate for *Salmonella* spp. (2.6%) in BTM compared with our study (Van Kessel et al., 2004).

Each year, an estimated 2.1 to 2.4 million cases of human campylobacteriosis occur, making it the most commonly reported bacterial cause of foodborne infection in the United States (Altekruuse et al., 1999a; Mead et al., 1999). *Campylobacter jejuni* is found in many foods of animal origin and has frequently been isolated from raw BTM (Rohrbach et al., 1992; Jayarao and Henning, 2001; Peterson, 2003). In this study *C. jejuni* was found in 2% of the BTM samples. Previous studies have reported the prevalence of *C. jejuni* in raw milk samples as ranging from <1 to 12% (Rohrbach et al., 1992; Jayarao and Henning, 2001). Outbreaks of *C. jejuni* enteritis caused by drinking raw milk are often associated with youth activities such as school trips to farms (Wood et al., 1992; Altekruuse et al., 1999a).

Dairy cattle can serve as reservoirs of STEC strains that can cause illnesses in humans through contaminated milk, from meat supplied through cull animals,

and by direct contact with cattle or the dairy farm environment. Shiga toxin-producing *E. coli* are highly pathogenic in humans with low infectious doses (Nataro and Kaper, 1998; Hussein and Sakuma, 2005). Among the STEC, O157:H7 is the classical serotype associated with enterohemorrhagic diseases. Non-O157 STEC strains are only recently becoming recognized as important human pathogens (Nataro and Kaper, 1998; Hussein and Sakuma, 2005). Consumption of raw milk has been implicated as the cause in several outbreaks of disease caused by *E. coli* O157:H7 (Keene et al., 1997; Proctor and Davis, 2000) and by non-O157 STEC (Wilson et al., 1998; Hussein and Sakuma, 2005). Shiga toxin-producing *E. coli* excrete potent Shiga toxins that are encoded by the *stx*₁ and *stx*₂ genes, respectively (Hussein and Sakuma, 2005). The STEC isolates in this study predominantly carried the *stx*₂ gene. Epidemiological data suggest that *stx*₂ is more important than *stx*₁ in the development of hemolytic uremic syndrome, a life-threatening illness associated with STEC infection in children (Nataro and Kaper, 1998).

Contamination with *L. monocytogenes* was the leading cause of food recalls by the FDA from 1994 to 1998 (Wong et al., 2000). In this study, all *L. monocytogenes* belonged to O antigen type 1. The O antigen includes serotypes 1/2a, 1/2b, and 1/2c; and serotypes 1, 1/2a, 1/2b, and 4b have been found in raw milk (Jayarao and Henning, 2001; Van Kessel et al., 2004). Outbreaks of *L. monocytogenes* have been associated with homemade Latin-style soft cheeses made from unpasteurized raw milk. These outbreaks occur primarily in Hispanic communities, with many of the cases involving pregnant women (Linnan et al., 1988; Centers for Disease Control and Prevention, 2001). Listeriosis is a major public health concern in these communities because pregnant women are especially susceptible to *L. monocytogenes*, which can cross the placenta and cause spontaneous abortions and stillbirths (Oliver et al., 2005).

Generally, dairy cattle are not considered reservoirs of pathogenic *Y. enterocolitica* (Jayarao and Henning, 2001). In humans, *Y. enterocolitica* is an important foodborne pathogen and is mainly transmitted through consumption of contaminated pork, milk, or water. Raw milk frequently contains *Y. enterocolitica*, but outbreaks of foodborne infection are not commonly reported (Schiemann, 1987; Jayarao and Henning, 2001). In this study, all *Yersinia* isolates were pathogenic based on the autoagglutination test described by Laird and Cavanaugh (1980).

An estimated 1.4 million cases of salmonellosis occur annually in the United States (Mead et al., 1999). Sales of raw milk directly to the public have resulted in foodborne outbreaks of multidrug-resistant salmonellosis in California and Washington (Reed and Grivetti, 2000),

Ohio, Illinois, Indiana, and Tennessee (Centers for Disease Control and Prevention, 2003). In this study, *Salmonella* was the predominant organism isolated from BTM. The *Salmonella* isolates were identified as *S. enterica* serotype Typhimurium ($n = 10$) and *S. enterica* serotype Newport ($n = 5$). The emergence of multidrug-resistant *S. enterica* serotype Typhimurium definitive type 104 and multidrug-resistant *S. enterica* serotype Newport is of particular concern to public health agencies (Keene, 1999; Jayarao and Henning, 2001; Oliver et al., 2005).

The isolation of foodborne pathogenic bacteria in raw milk has been reported extensively in Canada and United States (Rohrbach et al., 1992; Jayarao and Henning, 2001; Oliver et al., 2005). The isolation rate reported over the last 2 decades varies considerably. We feel that this variation could be attributed partly to the techniques used for isolation and identification of the pathogens, their true prevalence, sample size, season, geographic area, farm size, the number of animals on the farm, hygiene, and farm management practices. In this study, only a single BTM sample was examined for each farm. It seems likely to assume that repeated sampling would result in a higher overall incidence of pathogen detection at the farm level. Our findings clearly suggest that pathogens do occur in BTM and may pose a health hazard if milk is consumed raw. In this study, 105 (42.3%) of the 248 dairy producers who participated in the BTM pathogen survey reported that they consumed raw milk. Bacteriologic analysis of BTM showed that 15 (14.2%) of the 105 producers who consumed raw milk had ≥ 1 bacterial pathogen isolated from their bulk tank. Other studies have reported that 26.6 (Jayarao and Henning, 2001) and 25% (Rohrbach et al., 1992) of producers who consumed raw milk had ≥ 1 bacterial pathogen in their BTM. There was no significant difference in the incidence of pathogenic bacteria in raw milk of the dairy producers who did and did not consume raw milk ($P > 0.05$). Similar results were seen by Jayarao and Henning (2001).

Two-thirds (68.5%) of the 248 dairy producers surveyed were aware of the fact that raw milk could contain disease-causing bacteria, meaning roughly one-third of the dairy producers surveyed were not aware of the risk of foodborne pathogens in their BTM. However, out of the 105 dairy producers who drank raw milk, 61 (58.1%) reported that they continued to drink raw milk despite knowing that foodborne pathogens could be found in their raw BTM. The continuation of this high-risk behavior in spite of their awareness is a concern.

CONCLUSIONS

The findings of this study suggest that pathogenic bacteria of human health significance can be found in

raw milk. This observation, along with previously reported data on consumption of raw milk, indicates that dairy producers and farm families are at risk for exposure to foodborne pathogens when consuming raw milk. Consumption of raw milk is a preventable cause of foodborne illness, making pasteurization of raw milk an important public health tool for foodborne disease prevention. Yet one of the most surprising findings of this study was the fact that 31.4% of the dairy producers surveyed were unaware that their raw BTM could contain disease-causing microorganisms. These findings suggest a lack of knowledge among dairy producers about the risks associated with raw milk consumption. One way to approach this problem would be to develop educational outreach programs for dairy producers, as well as for the general public, that focus on issues related to the consumption of raw milk. This study shows that *C. jejuni*, STEC, *L. monocytogenes*, *Salmonella* spp., and *Y. enterocolitica* can be found in BTM. The prevalence rates were similar to other reports from the United States of foodborne pathogens in BTM. This study suggests a risk of exposure to foodborne pathogens among the dairy farm families who consume unpasteurized BTM.

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Guidelines for Monitoring Bulk Tank Milk Somatic Cell and Bacterial Counts

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ABSTRACT

This study was conducted to establish guidelines for monitoring bulk tank milk somatic cell count and bacterial counts, and to understand the relationship between different bacterial groups that occur in bulk tank milk. One hundred twenty-six dairy farms in 14 counties of Pennsylvania participated, each providing one bulk tank milk sample every 15 d for 2 mo. The 4 bulk tank milk samples from each farm were examined for bulk tank somatic cell count and bacterial counts including standard plate count, preliminary incubation count, laboratory pasteurization count, coagulase-negative staphylococcal count, environmental streptococcal count, coliform count, and gram-negative noncoliform count. The milk samples were also examined for presence of *Staphylococcus aureus*, *Streptococcus agalactiae*, and *Mycoplasma*. The bacterial counts of 4 bulk tank milk samples examined over an 8-wk period were averaged and expressed as mean bacterial count per milliliter. The study revealed that an increase in the frequency of isolation of *Staphylococcus aureus* and *Streptococcus agalactiae* was significantly associated with an increased bulk tank somatic cell count. Paired correlation analysis showed that there was low correlation between different bacterial counts. Bulk tank milk with low (<5000 cfu/mL) standard plate count also had a significantly low level of mean bulk tank somatic cell count (<200,000 cells/mL), preliminary incubation count (<10,000 cfu/mL), laboratory pasteurization count (<100 cfu/mL), coagulase-negative staphylococci and environmental streptococcal counts (<500 cfu/mL), and noncoliform count (<200 cfu/mL). Coliform count was less likely to be associated with somatic cell or other bacterial counts. Herd size and farm management practices had considerable influence on somatic cell and bacterial counts in bulk tank milk. Dairy herds that used automatic milking detachers, sand as bedding ma-

terial, dip cups for teat dipping instead of spraying, and practiced pre- and postdipping had significantly lower bulk tank somatic cell and/or bacterial counts. In conclusion, categorized bulk tank somatic cell and bacterial counts could serve as indicators and facilitate monitoring of herd udder health and milk quality.

(**Key words:** bulk tank milk, somatic cell, bacterial count, milk quality)

Abbreviation key: BTM = bulk tank milk, BTSCC = bulk tank somatic cell count, CC = coliform count, ES = environmental streptococci, LPC = laboratory pasteurization count, NC = noncoliform count, PIC = preliminary incubation count, SA = *Staphylococcus aureus*, SAG = *Streptococcus agalactiae*, SPC = standard plate count.

INTRODUCTION

Since the early 1990s, researchers have used bulk tank milk (BTM) to diagnose multiple problems (current and potential) that might exist in a dairy herd related to milk quality and mastitis pathogens. Progressive dairy producers, veterinarians, and dairy health consultants are interested in BTM analysis as a tool to determine milk quality and troubleshoot herds with mastitis. Many quality-conscious milk cooperatives have implemented BTM analysis to reward dairy producers who excel at producing high 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 (Emerson, 1989; Farnsworth, 1993; Bray and Shearer, 1996; Britten and Emerson, 1996; Keeter, 1997; Mickelson et al., 1998; Jayarao et al., 2001).

Successful milk quality assurance programs start with farm BTM free of antibiotic residues and with low somatic cell and bacterial counts, resulting in better quality products with longer shelflife (Boor et al., 1998; Ma et al., 2000; Reugg and Tabone, 2000). Many dairy producers also receive premiums from their milk cooperative for producing milk with low somatic cell and bacterial counts. Several guidelines have been proposed

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to interpret BTM milk bacterial counts (Bray and Shearer, 1996; Britt et al., 1997; Murphy, 1997; Jones and Sumner, 1999; Edmondson, 2000; Jayarao et al., 2001; Jayarao and Wolfgang, 2003). However, many of the guidelines are based on individual or collective experience, or extrapolations from other scientific studies. Further, many of the interpretive guidelines lack validation and provide little insight into the interrelationship between different groups of bacteria found in BTM. An extension and research study, conducted in Pennsylvania from April 2000 through March 2001, focused on BTM analysis. The findings of the milk quality survey were used to establish guidelines for interpreting BTM counts and also to understand the relationship between different bacterial groups that occur in BTM.

MATERIALS AND METHODS

Dairy Herds

The veterinary extension group at Pennsylvania State University with the support of the county extension agents implemented the study. A total of 12 county extension agents and 1 milk cooperative participated in the study. Each participating county extension agent/milk cooperative enrolled 7 to 11 dairy producers from its county or region. Dairy producers who participated in the study were solicited by county extension agents through their extension newsletter or announcements about the study during a monthly dairy extension meeting. For a given county, participation in the study was open to all dairy producers, and the first 12 dairy producers who responded to the invitation were included in the study.

Dairy producers who opted to participate in the program answered a self-administered questionnaire. The questionnaire sought information on the following aspects of the dairy herd: 1) herd size, 2) milk production, 3) milking frequency, 4) milkings per tank pickup, 5) type of milking facility, 6) change in milking facility, 7) use of automatic milking detachers, 8) type(s) of bedding, 9) animals purchased, 10) residue violations in the past 6 mo, 11) milk quality premiums in the past 6 mo, 12) type of milk equipment cleaning system, 13) mastitis prevention and control practices, and 14) milking procedures. The questionnaire used in this study has been successfully used previously (Jayarao and Cassel, 1999). The responses to the questions were analyzed to determine if any of these practices were associated with bulk tank somatic cell count (BTSCC) or bacterial counts.

Collection and Processing of BTM

The county extension agent provided on-farm instruction on BTM collection and handling procedures as described by National Mastitis Council BTM sample collection and handling guidelines (NMC, 1999). Dairy producers collected the sample in the first and third week of each month for 2 mo (4 samples total). Sampling kits containing gloves, racks, tubes (50 mL sterile screw cap tubes), and labels were provided. Bulk tank milk samples were collected in sterile 50-mL screw-cap centrifuge tubes. Within 24 hr of collection, all milk samples were shipped on ice overnight to the laboratory. On receipt of the sample in the laboratory, only those samples that recorded a temperature of $<7^{\circ}\text{C}$ were processed. The BTM in the 50-mL centrifuge tube was mixed thoroughly several times, and 20 mL of the milk was transferred to a snap-cap vial containing a preservative and sent to the Dairy One Laboratory in State College, PA, for determination of BTSCC. The remainder of the milk sample was used for bacteriological analysis.

Bacteriological Analysis of BTM

The BTM samples were examined for standard plate count (SPC), preliminary incubation count (PIC), laboratory pasteurization count (LPC), CNS count, environmental streptococci (ES) count, coliform count, and gram-negative noncoliform (NC) count. Bacteriological tests for milk quality were done as described by the American Public Health Association (Marshall, 1992). The milk samples were mixed thoroughly by gently inverting the milk vial 20 to 25 times. One milliliter of milk was transferred to a sterile tube containing 9 mL of quarter-strength Ringer's solution (Oxoid, Unipath Ltd., UK). The 10-fold diluted sample was vortexed at high speed for 15 s, and 50 μL was plated on selective and nonselective media using a spiroplater (Autoplate 4000, Spiral Biotech, Bethesda, MD). Plate count agar was used for enumeration of SPC, PIC, and LPC. The numbers of ES and *Streptococcus agalactiae* (SAG) in BTM samples were estimated using modified Edward's agar supplemented with colistin sulfate and oxolinic acid (Sawant et al., 2002). MacConkey's agar no. 3 (Oxoid) was used to determine coliform and noncoliform counts. Baird Parker's agar (Difco) was used to determine the number of CNS and presence of *Staphylococcus aureus* (SA). Plates for enumeration of SPC, PIC, and LPC were incubated at 32°C for 48 h. Plates for enumeration of CNS, ES, coliform count (CC), and NC were incubated at 37°C for 48 h. The Autoplate 4000 user guide (Spiral Biotech) was used to enumerate bacterial counts.

Colonies suggestive of SAG from modified Edward's agar supplemented with colistin sulfate and oxolinic acid were randomly selected and streaked on 5% sheep blood agar and incubated for 48 h at 37°C. All isolates were examined for gram's reaction and catalase production, serotyped (Streptex, Oxoid), and identified using API 20 STREP (BioMérieux, Hazelwood, MO) (Sawant et al., 2002). Colonies suggestive of SA from Baird Parker agar were randomly selected, streaked on 5% sheep blood agar, and incubated for 48 h at 37°C. The isolates were examined for hemolysis, catalase production, and coagulase production, and identified using API-STAPH (BioMérieux) (NMC, 1999).

Isolation of *Mycoplasma* was done as described by Gonzalez et al. (1995), with modifications. Briefly, 500 μ L of BTM was pre-enriched in modified Hayflick's broth and incubated for 48 h at 37°C in a moist 10% CO₂ incubator. One hundred microliters of the pre-enriched broth was streaked on modified Hayflick's agar and incubated for 7 d at 37°C in a moist 10% CO₂ incubator. *Mycoplasma* colonies were viewed under a low-power microscope. *Mycoplasma* was differentiated from *Acholeplasma laidlawii* using the digitonin inhibition test as described by Thurmond et al. (1989).

Data Analysis

A total of 149 dairy herds elected to participate in the study of which 4 herds opted not to participate during the course of the study. Of the 145 herds, 7 dairy herds were unable to provide information on farm management practices, and 12 dairy producers, on 2 consecutive occasions, supplied contaminated bulk tank milk, or the milk that was received for analysis had a temperature in excess of 7°C. A total of 126 dairy herds with complete data sets were used for data analysis.

Answers to the questionnaire were transferred to Microsoft Excel and grouped by their categorical response (e.g., *yes*, *no*). To estimate if a response had an influence on the mean BTSCC, SPC, PIC, LPC, SA, CNS, ES, CC, and NC counts for each group within a response were compared with the 3 categories (low, medium, high) within each bacterial count using one-way ANOVA. A *P*-value of < 0.05 was considered a significant association between the response and a category of the count. All statistical analyses were performed using JMP software version 4.0 (SAS Inst., Inc., Cary, NC). BTSCC and bacterial counts from the 4 BTM samples from each farm were transformed to log₁₀ values. The log₁₀ transformed BTSCC and bacterial counts (SPC, PIC, LPC, SA, CNS, ES, CC, and NC) from the 4 bulk tank samples from each farm were averaged and subjected to correlation coefficient analysis (SAS Inst. Inc.).

The BTSCC, SPC, PIC, LPC, SA, CNS, ES, CC, and NC counts were each classified as low, medium, or high. These 3 categories are the suggested interpretive criteria for monitoring BTM (see Table 2). The average counts for each of the 3 groups were compared using the Tukey-Kramer (equal variance) or Dunnett's T3 (unequal variance) procedures. These 2 procedures were used due to unequal sample sizes observed in the 3 categories of a given count. The Tukey-Kramer procedure performs all pair-wise comparisons, testing whether the 3 means are significantly different. The Dunnett's T3 procedure performs all comparisons with a control category. In our study, the second category (medium) was used as the control because the sample mean falls in this category. *P* < 0.05 was considered significant. Epi-info-2002 (Centers for Disease Control and Prevention, Atlanta, GA), a database and statistics system for epidemiology on microcomputers, was used for performing χ^2 -square tests and odds ratio analysis.

RESULTS

Dairy Herds

The responses to the 14 questions on the questionnaire were grouped based on herd size (Table 1). Nearly 71% of the farms had fewer than 100 lactating cattle, typical of farm families engaged in milking cows in Pennsylvania. Farm management practices changed as the herd size increased. This observation can be supported by change in the management practices such as 1) number of milkings per day, 2) type of milking facility, 3) use of automatic milking detachers, 4) type of cow bedding, 5) number of animals purchased, 6) milk equipment cleaning system, 7) mastitis prevention and control, and 8) milking practices. For the majority of the dairy herds, cows were milked twice a day (88%) in stanchion barns (61%) and/or parlors (39%). About 45% of dairy herds had automatic milking detachers. As the herd size increased, so did the use of automatic milking detachers. Sawdust was used as bedding on 44% of the farms surveyed. Nearly 5% of the respondents to the questionnaire indicated antibiotic residues in the last 6 mo. A majority of the dairy producers practiced dry cow treatment (88%), whereas 73% of the dairy producers who teat dipped their cows practiced both pre- and postdipping. Milking practices varied considerably within a given herd size and between the 4 herd size categories (Table 1). Significant differences were observed with respect to the type of bedding used (*P* ≤ 0.000), antibiotic residues in bulk tank milk (*P* ≤ 0.043), type of milking equipment cleaning system (*P* ≤ 0.022), dry cow treatment (*P* ≤ 0.043), teat-dipping practices (*P* ≤ 0.031), stripping practices before milking (*P* ≤ 0.028), and towel type (*P* ≤ 0.011) (Table 1).

Table 1. Characteristics of the dairy herds that participated in the study.

Query	Herd size				Total (n = 126)	χ^2 ($P \leq 0.005$)	
	< 50 (n = 35)	50 to 99 (n = 55)	100 to 199 (n = 30)	>200 (n = 6)			
No. of cows in milk (average)	38	67	136	294	87		
Milk produced per cow (lb)	33	34	33	30	32.5		
Times milked (%)							
Two	97	91	83	17	88	4.99 (0.111)	
Three	3	9	17	83	12		
No. of milkings in bulk tank (%)							
Two	0	10	24	17	11	6.63 (0.011)*	
Three	0	4	10	0	4		
Four	97	82	62	50	80		
>Four	3	4	3	13	5		
Milking facility							
Stanchion	94	75	10	0	61	0.28 (0.632)	
Parlor	6	25	90	100	39		
Change in milking facility in last 6 mo (%)	14	20	30	33	21	6.89 (0.078)	
Automatic milking detachers (%)	26	52	47	83	45	4.54 (0.122)	
Cow bedding (%)							
Combination (>1 type bedding, c-i)	11	9	3	0	8	11.86 (0.000)*	
Corn fodder	3	4	0	0	1		
Hay	0	4	0	0	2		
Mats	3	7	0	0	4		
Newspaper	6	11	7	0	8		
Sand	3	6	17	0	7		
Sawdust	37	33	63	87	44		
Shavings	14	11	3	13	10		
Straw	23	19	7	0	16		
Animals purchased (%)							
Dry cows	9	9	30	33	15		2.70 (0.145)
Milking cows	11	16	40	50	22		
Spring heifers	20	11	33	50	21		
Antibiotic residues in last 6 mo	3	9	4	0	5	11.3 (0.043)*	
Milk premiums in last 6 months	74	47	55	60	58	1.04 (0.382)	
Milk equipment cleaning system (%)							
Automatic	74	96	93	100	88	7.64 (0.022)*	
Manual	3	2	0	0	2		
Semi-automatic	23	2	7	0	10		
Mastitis prevention and control (%)							
Dry cow therapy (always)	86	88	93	100	88	11.37 (0.043)*	
Teat dipping	74	67	87	83	73	14.54 (0.031)*	
Predipping only	3	7	0	17	6	12.01 (0.007)*	
Postdipping only	18	33	10	0	22		
Pre- and postdipping	79	60	90	83	72		
Milking practices							
Written protocols	3	9	7	17	7	9.73 (0.052)	
Check for mastitis	38	36	58	80	42	0.62 (0.486)	
Wear gloves	18	12	27	50	19	4.96 (0.112)	
Strip before milking	63	63	77	83	67	15.63 (0.028)*	
Type of towel							
Common wash cloth	3	4	7	0	4	6.69 (0.011)*	
Individual wash cloth	11	18	30	67	21		
Paper towel	71	62	53	33	61		
Medicated towel	15	16	10	0	14		
Milkers							
Employees	3	2	17	17	7	1.92 (0.195)	
Family members	49	31	20	0	31		
Self and employees	9	20	43	67	25		
Self	40	37	20	17	37		

* $P \leq 0.05$.

Bulk Tank Somatic Cell Counts

The mean BTSCC (315,190 cells/mL) varied significantly with respect to the herd size. Fifty percent of the

BTM samples had a BTSCC <348,000 cells/mL. Paired correlation analysis showed that there was low correlation between BTSCC and different bacterial counts (Table 2). Bulk tank somatic cell counts were categorized

Table 3. Categorization of mean bulk tank somatic cell and bacterial counts.¹

Bulk tank	Proposed interpretive criteria		N ²	BTSCC	SPC	PIC	LPC	CNS	ES	CC	NC
	Category	Count (cfu/mL)									
BTSCC	Low	<200	19	179,390	2290	6540	90	360*	390	30	120
	Medium	200,000–400,000	55	283,320*	4140	10440	130	680	760	60	290
	High	≥400,000	52	497,310	5970	14960	160	940	1080	70	220
SPC	Low	<5,000	70	310,900	1950	6170*	80*	440*	490*	40	130*
	Medium	5,000–10,000	24	303,601	7470*	16870	280	1170	1220	70	350
	High	≥10,000	32	415,040	17680	31290	280	1410	1690	90	470
PIC	Low	<10,000	60	313,610	2370*	3660	80	470*	460*	40	120
	Medium	10,000–20,000	20	337,640	5100	13290*	130	1030	1170	60	200
	High	≥20,000	26	358,110	9280	44230	290*	1040	1350	70	440
LPC	Low	<100	52	307,860	2720	6130*	32	470	530	50	140
	Medium	100–200	21	313,700	2900	11870	140*	630	780	40	270
	High	≥200	53	368,430	8340*	20140	540	1120*	1190*	80	280
CNS	Low	<500	46	277,520*	2490	5810	80	260	450	50	120
	Medium	500–1000	40	350,140	3690	10370*	110	720*	820	50	180
	High	≥1000	40	390,760	10140*	26400	290*	2160	1470*	80	460
ES	Low	<500	33	284,960	1940	5090	60*	380	190	41	140
	Medium	500–1000	35	311,490	3480*	10110*	150	540	690*	60	170
	High	≥1000	58	378,970	8090	18970	200	1190*	1970	70	310
CC	Low	<50	57	307,990	3130	8520	110	600	620	20	140
	Medium	50–100	28	356,160	4980	11630	160	1030	950	70*	290
	High	≥100	41	354,710	6510	16330	170	700	980	220	290
NC	Low	<200	53	303,570	2980*	6790	100	560	650	40	60
	Medium	200–400	44	359,240	4770	12640	140	660	790	60	270*
	High	≥400	29	351,750	7970	24030*	240	1220	1140	90	1300

¹See Table 2 for abbreviation definitions.²N, number of bulk tanks**P* ≤ 0.05.

into 3 groups (low, <200,000; medium, 200,000 to 400,000; and high, >400,000 cells/mL) (Table 3). Mean CNS count was significantly associated with mean BTSCC (Table 3). A BTM with a mean BTSCC >200,000 cells/mL was 5 times more likely to have high CNS (>500 cfu/mL) counts compared with BTM with BTSCC <200,000 cells/mL (Table 4). Dairy producers who received milk premiums had significantly lower BTSCC (291,300 cfu/mL) compared with the BTSCC (378,090 cells/mL) in BTM of those dairy producers who did not receive premiums [χ^2 (*p*) = 3.27(0.0014)]. BTSCC was significantly lower when cows were milked using automatic milk detachers as compared with BTM from herds that milked cows without automatic milk detachers. The same observation was made with herds that

teat dipped the cows with a dip cup instead of using a spray. Interestingly, BTSCC was significantly higher in herds that practiced fore-stripping before milking compared with BTM from herds that did not. Dairy farms that used sand as bedding had significantly lower BTSCC in their BTM compared with dairy producers who used organic bedding such as shavings, newspaper, and straw (Table 5).

Standard Plate Count

For the 126 dairy herds in the study, the mean SPC for an 8-wk period was 4320 cfu/mL. The herd size did not influence the mean SPC of BTM. Fifty percent of BTM samples had a SPC <4120 cfu/mL. Paired correla-

Table 4. Odds ratio (confidence interval) estimates for somatic cell and bacterial counts.¹

Counts	BTSCC >200,000 cells/mL	SPC >5,000 cfu/mL	PIC >10,000 cfu/mL	LPC >100 cfu/mL	CNS >500 cfu/mL
BTSCC >200,000 cells/mL	—	—	—	—	—
SPC >5,000 cfu/mL	—	—	—	—	—
PIC >10,000 cfu/mL	—	9.55 (3.86–24.15)	—	—	—
LPC >100 cfu/mL	—	4.89 (2.07–11.75)	3.02 (1.36–6.77)	—	—
CNS >500 cfu/mL	5.04 (2.11–12.97)	5.86 (2.32–15.12)	3.13 (1.37–7.17)	3.71 (1.56–8.94)	—
ES >500 cfu/mL	—	6.80 (2.23–22.12)	4.22 (1.64–11.12)	2.93 (1.20–7.23)	5.75 (2.25–14.94)
NC >200 cfu/mL	—	6.14 (2.54–15.12)	3.73 (1.66–8.47)	—	—

¹See Table 2 for abbreviation definitions.