Somatic cell counts and bacteriological status in quarter foremilk samples of cows in Hesse, Germany—A longitudinal study

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ABSTRACT

Somatic cell counts (SCC) are generally used as an indicator of udder health. Currently in Germany, 100,000 cells/mL is the threshold differentiating infected and noninfected mammary glands. The aim of our study was the detailed analysis of udder health in a representative part of the dairy cow population in Hesse, Germany. Between 2000 and 2008, 615,187 quarter foremilk samples were analyzed. In addition to evaluation of distribution of SCC and prevalence of mastitis pathogens, pathogen prevalence was also calculated depending on SCC. The data indicated that 38% of all samples had SCC >100,000 cells/mL and 62% showed SCC ≤100,000 cells/mL; 31% of all samples revealed SCC ≤25,000 cells/mL. Coagulase-negative staphylococci were the dominant pathogens in the Hessian quarter foremilk samples (17.17% of all samples) followed by *Corynebacterium* spp. (13.56%), *Streptococcus uberis* (8.7%), and *Staphylococcus aureus* (5.01%). Mastitis pathogens were detected in 83% of all samples with SCC >100,000 cells/mL. However, the prevalence of mastitis pathogens in the SCC range from 1,000 to ≤100,000 cells/mL was 8.5% (5.51% minor pathogens, 2.01% major pathogens, and 0.98% other pathogens). For farms producing high quality milk, exceptional hygiene management is compulsory. One of the farms randomly selected showed clearly different results from the Hessian survey. Fifteen percent more samples lay in the SCC range ≤100,000 cells/mL with a lower prevalence of mastitis pathogens of 1.91% (1.03% minor pathogens, 0.83% major pathogens, and 0.05% other pathogens). Based on these results, inflammatory processes can obviously be detected in mammary glands

of udder quarters healthy according to the current definitions. However, we argue that such inflammation can be detected by examination of the relationship of immune cells in milk.

Key words: low somatic cell count, prevalence of mastitis pathogens, udder health status

INTRODUCTION

Mastitis is the most costly disease in milk production worldwide, and approximately 70 to 80% of the financial losses are caused by subclinical mastitis (Reneau and Packard, 1991; Seegers et al., 2003). Subclinical mastitis is characterized by decreased milk production, altered composition of the secretion, and presence of bacteria, but without any visible changes in milk and udder (Harmon, 1994).

For mastitis diagnosis, traditional and well-established tests including SCC and microbial culture-based methods are standard (Viguier et al., 2009). Several SCC cut-offs distinguishing infected and noninfected mammary glands and indicating IMI have been evaluated and discussed in the literature (Schepers et al., 1997; Schukken et al., 2003). According to current definitions of udder health in Germany, quarter foremilk samples with SCC ≤100,000 cells/mL are in the physiological range (DVG, 2002). It is well known that the crossover of normal cellular defense in the mammary gland into an inflammatory reaction starts at a level of >100,000 cells/mL (Harmon, 1994; DVG, 2002). However, SCC vary with status of lactation, age, stress of the animals, time and frequency of milking, season, and, mainly, udder infection status (Dohoo and Meek, 1982; Harmon, 1994).

The implementation of classical mastitis prevention programs (Neave et al., 1969) in combination with the introduction of penalty limits for bulk milk SCC have led to substantial progress in controlling subclinical

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mastitis worldwide. The geometric bulk milk SCC in Hesse, Germany, was 255,000 cells/mL in 1993 and decreased to 200,000 cells/mL in 2008 (HVL, 1994, 2009). A decrease in the geometric bulk milk SCC from 600,000 cells/mL in 1971 to 200,000 cells/mL in 2002 was observed in the Netherlands (Sampimon et al., 2005). In Finland, the geometric bulk milk SCC decreased from 325,000 cells/mL to 132,000 cells/mL between 1988 and 2002 (Myllys et al., 1998; Pitkälä et al., 2004). A moderate decrease of the geometric bulk milk SCC from 356,000 cells/mL in 1998 to 316,000 cells/mL in 2005 was described evaluating the data of one large US milk cooperative in New York State (Nightingale et al., 2008). In addition to mastitis control programs, the selection of animals showing low SCC in animal breeding programs might have had a strong influence on the development of mastitis pathogens. In particular, the prevalence of *Staphylococcus aureus* decreased. Data for *Staph. aureus* are also available for the German federal state of Hesse, Finland, the Netherlands, and the state of Wisconsin (United States). In Hesse, *Staph. aureus* showed a prevalence of 13.0% in 1995 and 2.9% in 2008 (LHL, 2009). In Finland, the prevalence of the bacteria was reduced from 5.1% in 1988 (Myllys et al., 1998) to 3.4% in 2001 (Pitkälä et al., 2004). The prevalence of *Staph. aureus* in the Netherlands decreased from 6.2% in 1973 to 1.8% in 2003 (Sampimon et al., 2009). In Wisconsin, the proportion of *Staph. aureus* in all culture-positive samples decreased from 17.7% in 1994 to a value of 9.7% in 2001 (Makovec and Ruegg, 2003). In contrast, CNS are currently the most isolated pathogens from milk samples in many countries (Pitkälä et al., 2004; Piepers et al., 2007; Sampimon et al., 2009).

The objective of our research was a detailed evaluation of the udder health situation in a representative part of the dairy cow population in Hesse, Germany, analyzing the distribution of SCC, prevalence of mastitis pathogens, and prevalence of mastitis pathogens in dependence of SCC. Additionally, for a more detailed evaluation of udder health in cows with low SCC and for comparison with the total Hessian average, a single farm practicing exceptional hygiene management was analyzed. However, because mastitis prevention programs have been implemented in Hesse, Germany, for many years, a high level of udder health could be expected.

MATERIALS AND METHODS

Animals and Farms

From 2000 to 2008, 615,187 quarter foremilk samples were taken from dairy cows in the German federal state of Hesse and analyzed by the Animal Health Services

(Hesse, Germany). This random test represented approximately 12.5% of all Hessian dairy cows. Samples were collected from farms producing high quality milk ($n = 98,430$; 16% of all samples), from conventional producing farms ($n = 412,175$; 67% of all samples), and from farms with severe udder health problems ($n = 104,852$; 17% of all samples). Almost all lactating cows of the farms were tested.

In addition, one farm (A) with exceptional hygiene management for the production of high quality milk was randomly selected to evaluate its cytotubacteriological status; 12,660 quarter foremilk samples were taken on that farm from 1995 to 2003. According to the legislative requirements in Germany, farms producing high quality milk have to perform monthly SCC and bacteriological examinations of quarter foremilk samples from all lactating cows.

On farm A, 60 Holstein-Friesian cows were housed in a pen barn and milked twice a day in a milking parlor. The average herd milk yield averaged 9,000 kg/yr.

Milk Sampling

Quarter foremilk samples were obtained according to DVG (2000) standards. Before milking, teat ends were scrubbed with 70% ethanol and the first 2 squirts of milk were discarded. A volume of 10 mL of milk per udder quarter was collected in a sterile 14-mL plastic sample tube (Greiner Bio-one, Frickenhausen, Germany).

Laboratory Analysis

Somatic cells were determined using a Fossomatic 5000 (Foss Electric, Hillerød, Denmark) and used as guideline for the selection of udder quarters for bacteriological analysis. Quarter foremilk samples showing $\text{SCC} > 100,000$ cells/mL were cultured categorically. All samples of a herd with $\text{SCC} \leq 100,000$ cells/mL were only examined bacteriologically when highly contagious mastitis pathogens such as *Staph. aureus*, *Streptococcus agalactiae*, or group G streptococci were present. Furthermore, all samples from farms producing high quality milk were analyzed bacteriologically independent from SCC.

Culture and isolate identification were performed according to IDF (1981) standards. Promptly after collecting the quarter foremilk samples and cooled transportation to the laboratory, about 10 μL of milk was streaked onto a quadrant of a 7% bovine blood agar plate containing 0.05% esculin (Merck, Darmstadt, Germany) and incubated for 48 h at 37°C; the plates were examined after 24 and 48 h of incubation. Isolates were classified into (1) major pathogens: *Staph. aureus*, *Strep. agalactiae*, *Streptococcus dysgalactiae*, *Streptococ-*

cus uberis, *Escherichia coli*, coliforms (e.g., *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*); (2) minor pathogens: CNS, *Corynebacterium* spp.; and (3) other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci (predominantly *Streptococcus canis*), yeasts, and fungi. Contaminated samples were defined as a mixture of at least 2 environmental type organisms without isolation of a major mastitis pathogen.

Statistical Analysis

The data sets of the results of the SCC examinations in Hesse and on farm A were divided into 15 different SCC ranges from $\leq 1,560$ to $>12,800,000$ cells/mL. Their limits were calculated using a logarithmic partition to the base 2 of the SCC scale according to the SCS standard [$SCS = \log_2 (SCC/100,000) + 3$; Ali and Shook, 1980]. All records with a missing SCC value or a value $<1,000$ cells/mL were excluded from the data sets (Hessian survey: $n = 27,281$; farm A: $n = 122$). These records were not included in the total numbers (Hessian survey: $n = 615,187$; farm A: $n = 12,660$). For a more practical interpretation, a wider scale range was determined according to the SCS standard defining 4 SCC groups: group I: $\leq 6,250$ cells/mL; group II: $>6,250$ to $\leq 25,000$ cells/mL; group III: $>25,000$ to $\leq 100,000$ cells/mL; and group IV: $>100,000$ cells/mL.

Distributions of SCC in Hesse and on farm A were tested for Gaussian distribution using the Shapiro and Wilk test as well as the Chi-squared test (BMDP Statistical Solutions Ltd., Cork, Ireland). Because of technical reasons, random tests of 2,000 records of each data set were used for the calculation of the Shapiro and Wilk test, whereas the whole data sets of both Hesse and dairy farm A were used for the Chi-squared test.

Additionally, a subset comprising data of Hesse from 2003 ($n = 79,204$) was analyzed to identify the effect of fixed and random effects on SCC by applying a linear mixed model. Selection of data for verification of results, as presented in Figure 1, was done because of computational limitations. The SCC were log-transformed according to Ali and Shook (1980) to obtain better statistical properties. The data subset was analyzed using the MIXED procedure of SAS 9.1 (SAS Institute Inc., Cary, NC) and the following statistical model

$$Y_{ijk} = \mu + H_i + Q_j + e_{ijk},$$

where Y_{ijk} = observed value for SCS in herd i and udder quarter j of cow k ; μ = overall mean; H_i = random

effect of herd i ($i = 1$ to 1,338); Q_j = fixed effect of quarter j ($j = 1$ to 4); and e_{ijk} = random error term.

In addition to the SCC data ($n = 235,556$), the results of the bacteriological examinations of 145,965 quarter foremilk samples analyzed in Hesse were available for the period 2000 to 2003. On farm A, the bacteriological data could be evaluated for the period 1995 to 2003 ($n = 12,660$). For statistical calculation, the classification of the bacteriological results into major, minor, and other pathogens was carried out according to Reneau (1986). The classification and calculation of the frequency of each pathogen was performed independently from SCC and depending on 15 SCC ranges defined above using SAS 9.1 (SAS Institute Inc.).

The data subset comprising results from 2003 ($n = 79,204$) was also analyzed to identify the impact of fixed and random effects on specific pathogens by applying a generalized linear mixed model. Because pathogens are classified as binary traits, the residuals cannot be normally distributed. The best function to describe the relationship between the dependent and independent variables is not linear but S-shaped. This is the primary reason why a linear logistic model with mixed effects was used. As described by König et al. (2005), the probability of observing the event of interest (e.g., prevalence of the pathogen) was

$$\pi_i = \text{Prob}(Y_i = 1|\theta),$$

where θ is a parameter vector including fixed and random effects. The logit of the observation Y_i was

$$\log \left[\frac{\pi_i}{1 - \pi_i} \right] = \eta_i.$$

Because π is the probability of $Y = 1$, it follows that $1 - \pi$ is the probability of $Y = 0$ and so $\frac{\pi}{1 - \pi}$ is the ratio of the 2 probabilities, which, when stated in the form of odds, gives the odds of having $Y = 1$. Analysis of variance of the measurements was carried out using logistic models implemented in the SAS glimmix macro (Wolfinger and O'Connell, 1993) that included the fixed effects of the udder quarter and the defined SCC group and the random effect of the herd. The F -ratios used in the ANOVA are identical to the Wald/rank(K) F -statistics as defined by Littell et al. (1999). Wald-type tests were also used to identify significant fixed effects on different pathogens (type III tests of fixed effects).

The final generalized linear model used to determine the impact of fixed and random effects on prevalence of pathogens was

Figure 1. Statistical distribution of SCC of all quarter foremilk samples analyzed in the German federal state of Hesse taken from 2000 to 2008 ($n = 615,187$) by classification into 15 SCC ranges using a logarithmic partition to the base 2 of the SCC scale according to the SCS standard. The bar charts show the distribution of SCC observed in Hesse, and the line shows the distribution of SCC expected by the exact Gaussian distribution.

$$\text{logit}(\pi_{rstu}) = \log \left[\frac{\pi_{rst}}{1 - \pi_{rst}} \right] = \eta_{rst} = \varphi + \gamma_r + \lambda_s + \varepsilon_t,$$

where π_{rstu} = probability of occurrence of a pathogen in udder quarter s and SCC group r for cow u in herd t ; φ = overall mean effect; γ_r = fixed effect of SCC-group; λ_s = fixed effect of udder quarter; and ε_t = random effect of the herd

RESULTS

Evaluation of Udder Health in Hessian Dairy Farms

For the evaluation of udder health in Hesse, Germany, a data set with the results of 615,187 quarter foremilk samples was used. The samples were derived from farms with different levels of udder health. In general, farms producing high quality milk showed a bulk tank SCC of 180,000 cells/mL. The bulk tank SCC of conventional milk producers averaged 200,000 cells/mL, whereas the bulk tank SCC in farms with massive udder health problems was higher (400,000 cells/mL).

Distribution of SCC. Somatic cell counts of the quarter foremilk samples analyzed ranged from 1,000 to 30,000,000 cells/mL. The distribution of all quarter foremilk samples in the 15 SCC ranges is illustrated in Figure 1. In total, 38% of all quarter foremilk samples indicated an inflammatory reaction (SCC >100,000 cells/mL) according to the current DVG definitions (DVG, 2002). A high frequency (62%) of the samples had SCC in a physiological range ($\leq 100,000$ cells/mL). Thirty-one percent of all samples had SCC between 25,000 and 100,000 cells/mL, and 31% had SCC $\leq 25,000$ cells/mL.

For a more practical interpretation, 4 SCC groups (I to IV) were defined. Six percent of the quarter foremilk samples analyzed in Hesse belonged to group I ($\leq 6,250$ cells/mL), 25% belonged to group II ($>6,250$ to $\leq 25,000$ cells/mL), 31% belonged to group III ($>25,000$ to $\leq 100,000$ cells/mL), and 38% belonged to group IV ($>100,000$ cells/mL).

The distribution of the logarithm of SCC in Hesse was similar to a Gaussian distribution (Figure 1). However, the goodness of fit test for normality according to the Shapiro and Wilk test as well as the chi-squared

Table 1. Least squares means and standard error of mean for SCS in different udder quarter positions from 79,204 quarter foremilk samples analyzed in Hesse, Germany, in 2003

Udder quarter	SCS	
	LSM	SEM
Front right	2.45 ^a	0.02
Rear right	2.52 ^b	0.02
Front left	2.31 ^c	0.02
Rear left	2.45 ^a	0.02

^{a-c}Least squares means with different letters differ significantly at $P < 0.05$.

test showed a significant deviation ($P < 0.001$) from the exact Gaussian distribution. In particular, in both tails of the distribution deviations from normality could be observed (Figure 1). More samples than expected were observed on the left side of the middle area of the Gaussian distribution. In contrast, fewer samples than expected were observed on the right side of the distribution.

To evaluate the effect of quarter position on SCC, a data set including 79,204 quarter foremilk samples tested in 2003 was analyzed as an exemplar using a statistical model. The data from 2003 were representative of the whole data set from 2000 to 2008. For statistical reasons, SCC were transformed to SCS. The analysis indicated a highly significant ($P < 0.001$) effect of quarter position on SCS. The mean SCS value of the front left quarter was significantly ($P < 0.05$) lower than that of the other 3 quarters (Table 1). In contrast, the highest mean SCS value could be calculated for the rear right quarter. In total, the mean SCS of both front quarters of 2.38 (SEM 0.01) was significantly (P

< 0.001) lower than in both rear quarters (2.49; SEM 0.01).

Distribution of Bacteriological Results. A high frequency of the quarter foremilk samples (62%) taken between 2000 and 2008 lay in a physiological range with $SCC \leq 100,000$ cells/mL and offered a proper standard of udder health. Bacteriological results were available for 145,065 of the 235,556 quarter foremilk samples taken from 2000 to 2003. The prevalence of mastitis pathogens in the samples examined was 51.96% (Table 2). The most frequently isolated pathogens were CNS (33.05%), followed by *Corynebacterium* spp. (26.1%), *Strep. uberis* (16.75%), *Staph. aureus* (9.63%), coliforms (2.21%), *Strep. dysgalactiae* (1.61%), *E. coli* (1.17%), and *Strep. agalactiae* (0.55%). Other pathogens were detected in 4.31% of all culture-positive samples. A proportion of 4.62% of all culture-positive samples was contaminated.

Approximately 40% ($n = 60,327$) of all quarter foremilk samples with $SCC \leq 100,000$ cells/mL were analyzed bacteriologically (Table 3). One fraction ($n = 30,759$) of these samples was cultured because of the presence of highly contagious mastitis pathogens such as *Staph. aureus*, *Strep. agalactiae*, or group G streptococci in a herd. The remaining fraction ($n = 29,568$) originated from high quality milk producers whose samples were generally analyzed bacteriologically. Pathogens were found in 8.5% of these quarter foremilk samples. Minor pathogens (*Corynebacterium* spp., CNS) as well as major pathogens (*Staph. aureus*, *Strep. uberis*) were detected beginning at an SCC level of $\leq 1,560$ cells/mL (Table 3). Within the SCC groups I to III, the frequencies of minor pathogens (5.51%), major pathogens (2.01%), and other pathogens (0.98%)

Table 2. Prevalence and distribution of mastitis pathogens in quarter foremilk samples analyzed bacteriologically in the German federal state of Hesse between 2000 and 2003 ($n = 145,065$)

Item	n	All quarters (%) ¹	Culture-positive (%) ²
Quarters analyzed bacteriologically	145,065		
Culture-negative quarters	69,691	48.04	
Culture-positive quarters	75,374	51.96	
<i>Staphylococcus aureus</i>	7,262	5.01	9.63
<i>Streptococcus agalactiae</i>	414	0.29	0.55
<i>Streptococcus dysgalactiae</i>	1,211	0.83	1.61
<i>Streptococcus uberis</i>	12,623	8.70	16.75
<i>Escherichia coli</i>	882	0.61	1.17
Coliforms ³	1,664	1.15	2.21
CNS	24,913	17.17	33.05
<i>Corynebacterium</i> spp.	19,673	13.56	26.10
Other pathogens ⁴	3,246	2.24	4.31
Contamination	3,486	2.40	4.62

¹Percentage of all quarters analyzed bacteriologically.

²Percentage of all culture-positive quarters.

³Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.

⁴Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

were higher than expected. Concerning the total number of quarter foremilk samples with SCC values $\leq 100,000$ cells/mL analyzed from 2000 to 2003 ($n = 150,818$), no mastitis pathogens were detected in 94 to 98% of the cases within the individual SCC ranges (Table 3).

Mastitis pathogens were detected in 82.91% of the 84,738 quarter foremilk samples with SCC values $>100,000$ cells/mL (group IV) analyzed from 2000 to 2003 (Table 3); CNS were the most frequently isolated pathogens. Their prevalence decreased from a maximum of 30.77% (SCC 100,000 to 200,000 cells/mL) to 12.03% (SCC of $>12,800,000$ cells/mL). *Staphylococcus aureus* were predominantly identified in samples with SCC $>400,000$ cells/mL (10.49 to 14.29%). *Corynebacterium* spp. were isolated with higher frequencies in samples with SCC between 100,000 and 800,000 cells/mL. *Streptococcus uberis* showed the highest prevalences (20.34 to 24.54%) in samples with SCC $>1,600,000$ cells/mL. The prevalence of *Strep. agalactiae* was generally very low ($<1\%$). However, *Strep. agalactiae* was more frequently detected (0.53 to 0.80%) in the SCC range from 400,000 to 12,800,000 cells/mL. The major pathogen *Strep. dysgalactiae* was predominantly identified (3.01 to 6.55%) in samples with SCC $>800,000$ cells/mL. The prevalence of *E. coli* increased from 0.47% at SCC of $>100,000$ to 200,000 cells/mL to a maximum of 8.29% at SCC of $>12,800,000$ cells/mL. Coliforms were predominantly isolated in samples showing SCC from 800,000 to $>12,800,000$ cells/mL. The prevalence of other pathogens in group IV ranged between 2 and 8%. The proportion of contaminated samples decreased at increasing SCC.

As an exemplar, a data set including all quarter foremilk samples ($n = 79,204$) tested cytot bacteriologically in 2003 was evaluated applying a generalized linear mixed model for the identification of fixed and random effects on the prevalence of the individual pathogens. The analysis indicated that the position of the udder quarter influenced the prevalence of most pathogens significantly (Table 4). The major pathogen *Staph. aureus* was significantly ($P < 0.001$) more frequently isolated from the front right quarter than from the other 3 quarters (Table 5). The highest prevalences for *Strep. agalactiae* and *Corynebacterium* spp. were found in front quarters, whereas *Strep. dysgalactiae* and CNS were predominantly isolated from rear quarters. *Streptococcus uberis* and other pathogens showed the highest prevalences in the front right and rear left quarters. Coliforms and *E. coli* were predominantly found in right quarters. Most contaminated samples originated from rear quarters. The proportion of samples without any isolation of pathogens was significantly ($P < 0.001$) higher in the front left quarter and lower in the rear left quarter than in right quarters.

A significant ($P < 0.001$) influence on the prevalence of all pathogens with the exception of *Strep. agalactiae* and *Strep. dysgalactiae* could be calculated for the 4 SCC groups defined (Table 4). The prevalence of most pathogens in groups I and II was significantly lower than in those of groups III and IV (Table 6). The proportion of samples without any isolation of pathogens decreased significantly ($P < 0.001$) with increasing SCC groups. The distribution of *Strep. agalactiae*, *Strep. dysgalactiae*, and *E. coli* depending on the 4 SCC groups could not be evaluated because of their extremely low prevalences in groups I to III.

Evaluation of Udder Health on Farm A

Farm A was chosen randomly to show the distribution of SCC and bacteriological results in a single farm practicing exceptional hygiene. The evaluation of udder health in this farm was based on the results of 12,660 quarter foremilk samples analyzed.

Distribution of SCC. Somatic cell counts of all quarter foremilk samples taken on farm A lay in a range between 1,000 and 24,000,000 cells/mL; the distribution of SCC on farm A is presented in Figure 1. A total of 23% of all quarter foremilk samples on farm A indicated an inflammatory reaction (SCC $>100,000$ cells/mL) according to the current DVG definitions (DVG, 2002) of udder health, representing 15% fewer samples than in the Hessian survey. The high frequency (77%) of samples with SCC $\leq 100,000$ cells/mL became evident on this farm, too, and was 15% above the Hessian average. Forty percent of all samples revealed SCC between 25,000 and 100,000 cells/mL and 37% revealed SCC $\leq 25,000$ cells/mL, ranging 9 and 6%, respectively, above the Hessian average.

Dividing SCC frequencies of farm A into the same SCC groups (I to IV) used for the Hessian surveillance, 9% of the quarter foremilk samples belonged to group I, 28% to group II, 40% to group III, and 23% to group IV.

Again, the goodness-of-fit test for normality according to the Shapiro and Wilk test as well as the Chi-squared test applied to the distribution of SCC on farm A (Figure 2) showed a significant deviation ($P < 0.001$) from the exact Gaussian distribution, as already seen for the Hessian samples and for the same reasons. However, the distribution observed was close to the normal distribution (Figure 2).

Distribution of Bacteriological Results. Bacteriological results were available for all 12,660 quarter foremilk samples taken from 1995 to 2003. The prevalence of mastitis pathogens was 21.65% (Table 7). Pathogens isolated most frequently were CNS (35.32%), followed by *Corynebacterium* spp. (29.92%), *Strep. ub-*

Table 3. Prevalence and distribution of mastitis pathogens (%) isolated from quarter foremilk samples obtained in the German federal state of Hesse within 2000 to 2003 (n = 235,556) in 15 different SCC ranges

Group/SCC range (×1,000 cells/mL)	n	Major pathogens ¹						Minor pathogens				
		<i>Staph. aureus</i>	<i>Strep. agalactiae</i>	<i>Strep. dysgalactiae</i>	<i>Strep. uberis</i>	<i>E. coli</i>	Coliforms	CNS	<i>Coryne- bacterium</i> spp.	Others ²	No pathogens	Contam. ³
Group I												
≤1.56	415	0.48	0	0	0.24	0	0	0.48	0.24	0	98.56	0
>1.56–3.13	3,470	0.35	0	0	0.09	0	0.03	0.58	0.35	0.06	98.40	0.14
>3.13–6.25	12,681	0.54	0	0	0.13	0	0.04	0.66	0.28	0.13	98.09	0.13
Group II												
>6.25–12.5	26,815	0.42	0.01	0.01	0.14	0	0.04	0.83	0.48	0.14	97.79	0.14
>12.5–25	33,837	0.43	0.02	0.01	0.15	0	0.06	0.78	0.94	0.20	97.19	0.22
Group III												
>25–50	36,246	0.47	0.03	0.02	0.18	0.01	0.06	0.92	1.39	0.13	96.59	0.20
>50–100	37,354	0.56	0.06	0.02	0.41	0.01	0.09	1.79	1.95	0.15	94.65	0.31
Group IV												
>100–200	30,406	3.73	0.21	0.36	10.77	0.47	1.32	30.77	27.21	2.54	18.26	4.36
>200–400	21,421	6.46	0.37	0.92	14.29	0.65	1.51	30.10	22.53	2.64	16.48	4.05
>400–800	13,426	10.49	0.64	1.69	16.13	0.89	1.43	26.95	18.95	3.40	16.19	3.24
>800–1,600	8,513	13.83	0.80	3.01	16.43	1.52	2.69	22.62	15.11	4.26	16.43	3.30
>1,600–3,200	5,270	13.78	0.70	3.29	20.34	2.03	4.22	20.53	11.37	6.09	14.86	2.79
>3,200–6,400	3,011	11.94	0.53	3.19	24.54	2.56	3.26	17.36	9.48	7.25	17.70	2.19
>6,400–12,800	1,943	14.29	0.88	4.23	21.71	5.00	4.18	15.11	6.03	8.10	18.82	1.65
>12,800	748	11.36	0.53	6.55	23.26	8.29	3.48	12.03	3.74	7.49	21.68	1.59

¹Major pathogens: *Staphylococcus aureus*; *Streptococcus (Strep.) agalactiae*; *Strep. dysgalactiae*; *Strep. uberis*; *Escherichia coli*; Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.

²Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

³Contaminated samples.

Table 4. Results of ANOVA for the prevalence of the individual pathogens in 79,204 quarter foremilk samples analyzed in Hesse, Germany, in 2003¹

Pathogen	Effect	
	Udder quarter	SCC group
<i>Staphylococcus aureus</i>	***	***
<i>Streptococcus agalactiae</i>	*	NC
<i>Streptococcus dysgalactiae</i>	**	NC
<i>Streptococcus uberis</i>	***	***
<i>Escherichia coli</i>	NS	***
Coliforms ²	***	***
CNS	***	***
<i>Corynebacterium</i> spp.	NS	***
Contamination	***	***
Other pathogens ³	NS	***
No pathogens	***	***

¹Analyzed factors were quarter positions (front right, rear right, front left, and rear left) and SCC group (I ≤6,250 cells/mL, II >6,250 to ≤25,000 cells/mL, III >25,000 to ≤100,000 cells/mL, IV >100,000 cells/mL); NC = not calculated.

²Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.

³Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

****P* < 0.001; ***P* < 0.01; **P* < 0.05; NS = *P* > 0.05

eris (21.71%), *Staph. aureus* (5.29%), coliforms (1.86%), *Strep. dysgalactiae* (0.62%), and *E. coli* (0.51%). Other pathogens were detected in 0.80% of all culture-positive samples. A proportion of 3.98% of all culture-positive samples was contaminated. The major pathogen *Strep. agalactiae* could not be isolated from any sample taken on farm A.

Mastitis pathogens were found in 1.91% of 9,715 quarter foremilk samples showing SCC ≤100,000 cells/mL (Table 8). Minor pathogens (CNS) were detected at a minimum of ≤1,560 cells/mL, whereas major pathogens (*Staph. aureus*) could be isolated beginning at a level of >3,130 to 6,250 cells/mL. The frequencies of minor pathogens (1.03%), major pathogens (0.83%), and other pathogens (0.05%) were clearly lower within the SCC groups I to III compared with the Hessian averages. No mastitis pathogens were detected in 96 to 99% of the cases within the individual SCC ranges (Table 8).

Mastitis pathogens were detected in 87.95% of the 2,905 quarter foremilk samples with SCC values >100,000 cells/mL (group IV) analyzed within this period (Table 8); CNS were the most frequently isolated pathogens. Their prevalence decreased from a maximum of 33.62% (SCC 100,000 to 200,000 cells/mL) to 17.64% (SCC of 6,400,000 to >12,800,000 cells/mL) and increased again to a level of 33.33% at SCC >12,800,000 cells/mL. *Staphylococcus aureus* was predominantly isolated in samples with SCC from >200,000 to 6,400,000 cells/

mL (3.85 to 10.16%). The prevalence of *Corynebacterium* spp. decreased constantly from 31.83% at SCC of >100,000 to 200,000 cells/mL to 8.35% at SCC of >12,800,000 cells/mL. *Streptococcus uberis* showed prevalences varying between 14.83% and 35.29% in the SCC range >100,000 cells/mL. The prevalence of *Strep. dysgalactiae* increased from 0.06% at SCC of >100,000 to 200,000 cells/mL to 8.33% at SCC of >12,800,000 cells/mL. *Streptococcus agalactiae* was not detected. The prevalence of *E. coli* in group IV ranged between 0 and 8.33%. The prevalence of coliforms increased from 1.05% at SCC of >100,000 to 200,000 cells/mL to 8.33% at SCC of >12,800,000 cells/mL, except that no coliform bacteria were isolated in samples showing SCC between 6,400,000 and 12,800,000 cells/mL. The prevalence of other pathogens in group IV lay between 0 and 12%. The proportion of contaminated samples ranged between 0 and 5%.

DISCUSSION

Udder health status in the German federal state of Hesse was evaluated on the basis of quarter foremilk samples taken from 2000 to 2008. Quarter foremilk samples are accepted for the evaluation of udder health because a high correlation between SCC in foremilk samples and in total representative samples was reported (Dohoo and Meek, 1982). In general, foremilk refers to the first secretions removed from the teat at milking time and represents a portion of cisternal milk (Stockler et al., 2009). It is known that the milk

Table 5. Least squares means for the prevalence of pathogens in different udder quarter positions from 79,204 quarter foremilk samples analyzed in Hesse, Germany, in 2003

Pathogen	Udder quarter ¹			
	FR	RR	FL	RL
<i>Staphylococcus aureus</i>	1.08 ^a	0.77 ^b	0.86 ^b	0.86 ^b
<i>Streptococcus agalactiae</i>	0.30 ^{ab}	0.22 ^a	0.39 ^b	0.29 ^{ab}
<i>Streptococcus dysgalactiae</i>	0.51 ^a	0.64 ^{ab}	0.55 ^a	0.82 ^b
<i>Streptococcus uberis</i>	1.01 ^a	0.82 ^b	0.85 ^{bc}	0.98 ^{ac}
<i>Escherichia coli</i>	0.38 ^a	0.36 ^{ab}	0.25 ^b	0.28 ^{ab}
Coliforms ²	0.42 ^a	0.60 ^b	0.37 ^a	0.40 ^a
CNS	4.17 ^a	4.46 ^b	4.09 ^a	4.81 ^c
<i>Corynebacterium</i> spp.	3.75 ^{ab}	3.73 ^{ab}	3.87 ^a	3.62 ^b
Contamination	0.68 ^a	0.89 ^b	0.76 ^a	0.93 ^b
Other pathogens ³	0.07 ^a	0.06 ^a	0.06 ^a	0.08 ^a
No pathogens	83.65 ^a	83.82 ^a	84.79 ^b	82.22 ^c

^{a-c}Least squares means within the same row with different letters differ significantly at *P* < 0.05.

¹Udder quarter position: FR = front right, RR = rear right, FL = front left, RL = rear left.

²Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.

³Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

Table 6. Least squares means for the prevalence of pathogens in the 4 SCC groups defined from 79,204 quarter foremilk samples analyzed in Hesse, Germany, in 2003

Pathogen	SCC group ¹			
	I	II	III	IV
<i>Staphylococcus aureus</i>	0.37 ^{ab}	0.35 ^a	0.57 ^b	7.71 ^c
<i>Streptococcus agalactiae</i>	—	—	—	—
<i>Streptococcus dysgalactiae</i>	—	—	—	—
<i>Streptococcus uberis</i>	0.03 ^a	0.03 ^a	0.07 ^a	1.04 ^b
<i>Escherichia coli</i>	—	—	—	—
Coliforms ²	0.25 ^a	0.21 ^a	0.31 ^a	2.19 ^b
CNS	1.58 ^a	1.88 ^a	3.55 ^b	27.81 ^c
<i>Corynebacterium</i> spp.	0.83 ^a	2.30 ^b	3.78 ^c	22.78 ^d
Contamination	0.42 ^a	0.49 ^a	0.50 ^a	4.11 ^b
Other pathogens ³	0.003 ^a	0.08 ^a	0.07 ^a	1.27 ^b
No pathogens	95.78 ^a	94.00 ^b	90.05 ^c	17.52 ^d

^{a-d}Least squares means within the same row with different letters differ significantly at $P < 0.05$.

¹SCC groups: I = $\leq 6,250$ cells/mL; II = $> 6,250$ to $\leq 25,000$ cells/mL; III = $> 25,000$ to $\leq 100,000$ cells/mL; IV = $> 100,000$ cells/mL.

²Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.

³Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

Figure 2. Statistical distribution of SCC of all quarter foremilk samples analyzed on farm A taken from 1995 to 2003 ($n = 12,660$) by classification into 15 SCC ranges using a logarithmic partition to the base 2 of the SCC scale according to the SCS standard. The bar charts show the distribution of SCC observed on farm A, and the line shows the distribution of SCC expected by the exact Gaussian distribution.

Table 7. Prevalence and distribution of mastitis pathogens isolated from quarter foremilk samples taken on farm A between 1995 and 2003 (n = 12,660)

Item	n	All quarters (%) ¹	Culture-positive (%) ²
Quarters analyzed bacteriologically	12,660		
Culture-negative quarters	9,919	78.35	
Culture-positive quarters	2,741	21.65	
<i>Staphylococcus aureus</i>	145	1.15	5.29
<i>Streptococcus agalactiae</i>	0	0	0
<i>Streptococcus dysgalactiae</i>	17	0.13	0.62
<i>Streptococcus uberis</i>	595	4.70	21.71
<i>Escherichia coli</i>	14	0.11	0.51
Coliforms ³	51	0.40	1.86
CNS	968	7.65	35.32
<i>Corynebacterium</i> spp.	820	6.48	29.92
Other pathogens ⁴	22	0.17	0.79
Contamination	109	0.86	3.98

¹Percentage of all quarters analyzed bacteriologically.²Percentage of all culture-positive quarters.³Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.⁴Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.

fraction has a substantial affect on SCC because milk secreted during the milking process has lower SCC than milk from pre- or postmilking (Olde Riekerink et al., 2007). However, the volume of foremilk varies from 5 to 60 mL (Fernando and Spahr, 1983; Vangroenweghe et al., 2002; Sarikaya and Bruckmaier, 2006). Sarikaya and Bruckmaier (2006) found a significant decrease of SCC comparing the first 10 mL of milk with the follow-

ing 140 mL in udder quarters secreting >100,000 cells/mL, whereas this decrease was only moderate for udder quarters with a SCC <100,000 cells/mL. Comparing SCC in the first 5 mL of milk with the following 20 mL in infected and uninfected mammary glands, Fernando and Spahr (1983) reported slightly higher values for the first 5 mL of milk. In our study, foremilk was defined as the first 10 mL of milk collected from the teat after the

Table 8. Prevalence and distribution of mastitis pathogens (%) isolated from quarter foremilk samples obtained in dairy farm A within the years 1995 to 2003 (n = 12,660) in 15 different SCC ranges

Group/SCC range (×1,000 cells/mL)	n	Major pathogens ¹						Minor pathogens				
		<i>Staph.</i> <i>aureus</i>	<i>Strep.</i> <i>agalactiae</i>	<i>Strep.</i> <i>dysgalactiae</i>	<i>Strep.</i> <i>uberis</i>	<i>E. coli</i>	Coliforms	CNS	<i>Coryne-</i> <i>bacterium</i> spp.	Others ²	No pathogens	Contam. ³
Group I												
≤1.56	40	0	0	0	0	0	0	2.50	0	0	97.50	0
>1.56–3.13	283	0	0	0	0	0	0	0.71	0.35	0	98.94	0
>3.13–6.25	807	0.37	0	0	0	0	0	0.25	0	0	99.38	0
Group II												
>6.25–12.5	1,459	1.24	0	0	0.14	0	0	0.34	0.14	0	98.14	0
>12.5–25	2,052	0.34	0	0	0.19	0	0	0.58	0.34	0	98.55	0
Group III												
>25–50	2,564	0.55	0	0	0.08	0	0	0.74	0.31	0	98.32	0
>50–100	2,550	0.71	0	0	0.47	0	0.04	0.98	0.63	0.08	96.97	0.12
Group IV												
>100–200	1,618	1.29	0	0.06	14.83	0.31	1.05	33.62	31.83	0.62	11.77	4.62
>200–400	752	3.85	0	0.80	26.33	0.53	1.60	29.65	23.01	0.53	11.58	2.12
>400–800	273	5.50	0	1.47	28.94	0.73	1.47	27.47	20.15	0	11.71	2.56
>800–1,600	128	10.16	0	1.56	17.19	0	7.03	23.44	20.31	0	15.62	4.69
>1,600–3,200	65	6.16	0	1.54	21.54	1.54	6.15	23.08	13.85	3.08	18.44	4.62
>3,200–6,400	40	7.50	0	2.50	30	2.50	7.50	20	12.50	0	15	2.50
>6,400–12,800	17	0	0	5.88	35.29	0	0	17.64	11.76	11.76	17.67	0
>12,800	12	0	0	8.33	33.33	8.33	8.33	33.33	8.35	0	0	0

¹Major pathogens: *Staphylococcus aureus*; *Streptococcus* (*Strep.*) *agalactiae*; *Strep. dysgalactiae*; *Strep. uberis*; *Escherichia coli*; Coliforms = *Klebsiella* spp., *Serratia* spp., *Enterobacter* spp., other *Enterobacteriaceae*.²Other pathogens: *Arcanobacterium pyogenes*, *Proteus*, *Prototheca zopfii*, *Pseudomonas* spp., group G streptococci, yeasts, and fungi.³Contaminated samples.

first 2 squirts were discarded. Therefore, we presume that our SCC values are representative for the analysis of the udder health status. Foremilk samples were taken from farms producing high quality milk, from conventional farms, and from farms with severe udder health problems. This random test of 615,187 quarter foremilk samples represented 12.5% of all Hessian cows. The large data set generated indicated that approximately two-thirds of all quarter foremilk samples lay in the physiological range of $\text{SCC} \leq 100,000$ cells/mL. This high frequency confirmed a proper level of udder health in the Hessian dairy cow population. The geometric mean of SCC in bulk milk samples in Hesse showed a value of 200,000 cells/mL in 2008 (HVL, 2009). Comparable data are available from the Netherlands, Belgium, and Finland. In the Netherlands, a geometric mean of SCC in bulk milk samples of 200,000 cells/mL was observed in 2002 (Sampimon et al., 2005). The geometric mean of SCC of bulk milk samples in Belgium was 221,000 cells/mL in 2006 (Piepers et al., 2007). In Finland, the geometric mean of bulk milk SCC was 132,000 cells/mL in 2002 (Pitkälä et al., 2004).

The statistical distribution of the logarithm of SCC of all quarter foremilk samples tested in the years 2000 to 2008 in Hesse did not exactly show a Gaussian distribution. The reasons for the deviation of the empirical distribution curve may be due to the very large sample size and the origin of the samples. The data were collected from dairy farms offering different levels of udder health. Using a logarithmic partition for the SCC scale, the empirical distribution of SCC was still skewed to the right. Fewer samples at the level of $\leq 3,130$ cells/mL and a level of $>50,000$ to 1,600,000 cells/mL were observed than expected by exactly normally distributed values. In contrast, more samples lay in the range from $>3,130$ to 50,000 cells/mL and at $\text{SCC} > 1,600,000$ cells/mL than expected by normal distribution.

The distribution of SCC on dairy farm A showed 15% more samples in the SCC range $\leq 100,000$ cells/mL than the Hessian average. This represented a very high standard of udder health in this high quality milk-producing farm and is due to exceptional hygiene management. The distribution of SCC on farm A differed from a Gaussian distribution for the same reasons as determined for the total Hessian random test, but the deviation was reduced.

The results of our study indicated that SCC in front quarters were significantly lower than in rear quarters, which is in agreement with the results of Schepers et al. (1997). Because high SCC is usually an indicator of an immune response to IMI with bacterial pathogens (Dohoo et al., 1984), a higher prevalence of pathogens should be expected in rear quarters. However, in our data set, the prevalence of bacteria differed between

the individual quarters but these differences did not explain the higher SCC in rear quarters. Rear quarters might be more susceptible to infections than front quarters, as published previously (Pearson and Mackie, 1979), because of larger capacity and mass, greater vulnerability to direct trauma (e.g., horning), and greater exposure to environmental effects. In addition, teats of the rear quarters are frequently nearer the floor, especially in older cows, and would thus be contaminated or subjected to injury more readily.

In the Hessian survey in the period 2000 to 2003, SCC was used as a guideline for the selection of quarter foremilk samples for bacteriological analysis. The data showed 51.96% of samples to be culture-positive. Our results are comparable to those obtained from the Netherlands (Sampimon et al., 2009), Belgium (Piepers et al., 2007), and Finland (Pitkälä et al., 2004). Similar to results from Hesse (17.17%), CNS were the most frequently isolated pathogens in the Netherlands, Belgium, and Finland (9.7 to 16.61%). However, it is not clear whether IMI with CNS will always result in inflammation (Sampimon et al., 2009). High frequencies of isolation of *Corynebacterium* spp. have been noted in Hesse (13.56%) and Finland (11.52%), in contrast to the Netherlands and Belgium, where only 2.5 and 0.1% of the samples showed *Corynebacterium* spp., respectively. These differences were probably related to changes in herd management and bacteriological ecology in the herd environment (Pitkälä et al., 2004). The prevalence of *Staph. aureus* in the Netherlands, Belgium, and Finland (1.8 to 3.4%) was lower than in Hesse (5.01%). Implementation of standard mastitis prevention programs (Neave et al., 1969) led to a clear decrease of mastitis caused by *Staph. aureus* in many countries. In contrast to the low prevalence of *Strep. uberis* in the Netherlands and Finland (1.1 and 0.65%, respectively), the prevalence of this species was higher in Belgium (2.7%) and Hesse (8.7%). These differences may originate from different hygiene and management systems in these countries, because *Strep. uberis* is an environmental pathogen. *Streptococcus agalactiae* showed a low prevalence in Hesse (0.29%), Belgium (0.1%), and Finland (0.02%), but was not detected in any of the samples taken in the Netherlands in 2003. The implementation of mastitis prevention programs led to a clear reduction of the prevalence of *Strep. agalactiae*, too. The prevalence of *Strep. dysgalactiae* in Hesse (0.83%) was similar to that in the Netherlands (1.2%), but differed from that in Belgium (0.4%) and Finland (0.05%). The epidemiology of this pathogen is intermediate between contagious and environmental. When it is known which factors increase the incidence of IMI with these pathogens, the control program for *Strep. dysgalactiae* may need to be adjusted (Sampimon

et al., 2009). The low prevalences of coliforms and *E. coli* observed in Hesse (1.15 and 0.61%, respectively), Belgium (0.1% coliforms), and Finland (0.14% coliforms) were expected because these species are generally involved in acute clinical mastitis of short duration (Todhunter et al., 1991). In the Dutch study, *E. coli* was considered in the group of other bacteria because of only 2 isolations.

In our investigation, major pathogens (*Staph. aureus*, *Strep. uberis*) as well as minor pathogens (*Corynebacterium* spp., CNS) were detectable starting from an SCC level of 1,000 cells/mL. Similar data were found in a Dutch study (ten Napel et al., 2009). However, *Staph. aureus* is a natural species of mammalian skin and mucous epithelia (Sutra and Poutrel, 1994). *Streptococcus uberis* is considered an environmental pathogen (Radostits et al., 2007). Both major pathogens cause mastitis, which could take subclinical or clinical courses. *Corynebacterium* spp. readily colonize the teat canal of dairy cows (Brooks and Barnum, 1984). Coagulase-negative staphylococci are part of the normal teat skin flora and can colonize the teat canal (Devriese and De Keyser, 1980); some CNS species are detectable in the environment (White et al., 1989). In case of both minor pathogens, it is possible that they can contaminate the milk samples but not cause IMI (Linde et al., 1980). However, it is not clear whether the pathogens that we isolated from quarter foremilk samples with SCC values from 1,000 to 100,000 cells/mL originated from contamination or whether they caused an IMI. In any case, no signs of an IMI were visible based on SCC. Further research is needed for a detailed evaluation of immunological processes in such udder quarters.

The results of the bacteriological examinations on farm A were similar to the Hessian survey, and the low prevalence of mastitis pathogens in the SCC range $\leq 100,000$ cells/mL could be expected because of the exceptional level of hygiene management.

Somatic cell counts $>100,000$ cells/mL are normally related to inflammatory processes inside the mammary gland. In the Hessian study, bacterial diagnosis was possible in approximately 83% of these cases. Negative bacteriological results in these cases could depend on spontaneous elimination of infection (Eberhart et al., 1979; Smith et al., 1985), intermittent shedding of the pathogens (Sears et al., 1990), or presence of antimicrobials or other inhibitors in milk (Reiter, 1978). At the time of examination, pathogens could also be ingested by phagocytes or survive intracellularly in the host (Newbould and Neave, 1965; Hill et al., 1978). Exudations of too-low masses of the pathogens or ceased growth of the pathogens are further reasons for negative results (Sears et al., 1990). Negative results

following antibiotic therapies should be excluded in our study because of withdrawal period regulations.

An SCC range of $\leq 100,000$ cells/mL is within the physiological range, but could be related to subclinical mastitis in the presence of pathogens. However, SCC values $\leq 100,000$ cells/mL are normally not taken into account to evaluate inflammatory processes. Bacteria detected could also be derived from contamination of the skin, teat canal, or environment. We presume that not all of these bacteria contaminated the milk samples. Therefore, it is obvious that they originated from inside the mammary gland and might cause an IMI even in the SCC range $<100,000$ cells/mL.

Our data showed that in 62% of all udder quarters SCC $\leq 100,000$ cells/mL were detected, suggesting a high standard of udder health in the Hessian dairy cow population analyzed. In view of the mastitis pathogens, prevalences were clearly lower in samples with $\leq 100,000$ cells/mL than in samples with $>100,000$ cells/mL. This finding confirmed the 100,000 cells/mL threshold differentiating infected from noninfected mammary glands. However, our data also indicated that minor and major pathogens were detected even at a minimum of 1,000 cells/mL. This result led us to suspect inflammatory processes in the SCC range $\leq 100,000$ cells/mL. We assert that these inflammatory processes could be elucidated by examination of the relationship of immune cells in milk. Differential cell counts might be a better indicator than SCC for a profound evaluation of inflammation, especially at SCC levels $\leq 100,000$ cells/mL.

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