

## The Management Practices Associated With Prevalence and Risk Factors of Mastitis in Large Scale Dairy Farms in Thuringia-Germany 1: Environmental Factors Associated With Prevalence of mastitis

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**Abstract:** The present field study was conducted to investigate the factors that influence the udder health status of dairy cows in Thuringia-Germany. A total of 64542 udder quarter and whole milk samples of which 56950 were udder quarter samples and 7592 were whole milk samples, from 10741 dairy cows in 48 dairy farms in the state of Thuringia were collected and subjected to microbiological investigations. The prevalence of infection was 27.57% of the quarters and 49.59% of the whole milk samples. *Staphylococcus aureus* and Coagulase Negative Staphylococci (CNS) were the most frequently isolated contagious pathogens. Whereas, *Streptococcus dysgalactiae* and Esculin Positive Streptococci (EPS, environmental pathogens), showed a prevalence in the udder and udder quarter samples of 12.90/13.90% and 9.0/10.60% respectively. The study found that the risk of developing mastitis was increased when the attacking pathogen was contagious ( $p < 0.001$ ; OR, 3.11), in small herds ( $p < 0.001$ ; OR, 2.91) and in purchased heifers ( $p < 0.001$ ; OR, 2.97). Cows in early stage of lactation as well as summer calving cows were at higher risk of developing mastitis (OR, 2.64 and 2.51 respectively).

**Keywords:** Odds ratio, mastitis, risk factors, prevalence, dairy cows, contagious and environmental pathogens.

### INTRODUCTION

In recent years the demand for liquid milk increased tremendously worldwide, due to increased population growth. Consumers in the industrialized countries demanding food not only to be economical, but also safe and sound in respect to animal welfare and the environment. In Germany it was found that the number of dairy farms decreased at the time the herd size grew-up (Klaas, 2000). Mastitis is the costliest disease of dairy industry today with an annual losses in USA estimated to be 200 \$ per cow (David and Shearer, 1996). Whereas, in Germany mastitis losses/cow/year estimated to be 285 \$ (Hamann, 2001). Infectious mastitis is present when the pathogen and the inflammatory changes were detected in the secretion. Whereas, non specific mastitis is present when there were inflammatory changes but no pathogen in the secretion and a latent infection is present when the secretion contained pathogens but had normal cell count (IDF, 1987). Frequent isolation of minor environmental pathogens and increased incidence rate of mastitis was reported when farmers used straw yard housing for milking cows (compared with cubicle housing), mucked out the calving area less frequently than once per month, when they had greater than 50% replacement rate and when always practiced post-milking teat disinfection (Linde *et al.*, 1980; Brooks and Brnum, 1984; Pankey *et al.*, 1985; Watts, 1988; Woodward *et al.*, 1988; Barkema *et al.*, 1999 and Peeler *et al.*, 2000). Introduction of improved control measures were found to reduce mastitis caused by contagious pathogens particularly *S. aureus*, *St. agalactiae* and *St. dysgalactiae* (Wilesmith *et al.*, 1986). Smith *et al.* (2000) stated that small herds reported more cows leaving for mastitis than high, medium and low medium herd size. In a recent study in Ethiopia, Biffa *et al.* (2005) found that Prevalence of mastitis was significantly influenced by season of calving ( $\lambda_2 = 28.7$ ;  $p < 0.001$ ).

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The primary objective of the study was to investigate and explore environmental factors most likely influencing the prevalence and risk factors associated with mastitis in large scale dairy farms.

### MATERIALS AND METHODS

The study covered the period from June 1998 to April 2000. Farms included in the study (48 large scale dairy farms) are among 572 dairy farms in the state of Thuringia-Germany which are members of the MLP-organization (Milchleistungsprüfung). A total of 64542 milk samples, of which 56950 were udder quarter samples and 7592 were whole milk samples from 10742 cows were regularly tested by the LKV (Landeskontrollverband). Herd size, herd management, housing, feeding regime and husbandry system data were obtained using a questionnaire. Milk performance data was supplied by the national data center (VIT) at Verden. Regularly collected milk samples were subjected to bacteriological investigations at the Animal Health Service-Mastitis Laboratory at Bad Langensalza (Thuringia). The three sources of the data were merged into one data set (SAS, 1996). Milk samples, which were collected before and after calving, were subjected to bacteriological investigations and the pathogens isolated were categorized into two groups (contagious and environmental). Table 1 summarizes the factors distribution of the data.

**Table 1:** Factors distribution of the data

| Factor               | No. of classes | Definition  |
|----------------------|----------------|---|
| Herd size            | 3              | Small (< 200 cows), medium (200-400 cows), large (> 400 cows)   |
| Origin of the heifer | 2              | Farm bred and purchased   |
| Stage of lactation   | 3              | Early (1-100 days), medium (101-200 days) and late (>200 days)  |
| Season of the year   | 4              | Spring (March-May), Summer (June-August), Autumn (September-November) and Winter (December-February ) |

The frequencies of contagious and environmental groups of bacteria were computed (SAS, 1996).  $\lambda^2$ -test was performed to test the significance of bacteria encountered in the udder quarters and the udder in accordance with the factors studied.

For the investigation of the relationship between the probabilities of occurrence of mastitis in response to environmental factors, logistic regression analysis was performed in which logistic regression model (events/trials) was used. In the model each dependent variable can accept any value, whereas, the independent variable (explanatory) values ranged between 0 and 1. The unknown parameter  $\beta$  was estimated by the method of maximum likelihood. The procedure logistic (SAS, 1996) was employed to estimate the unknown parameter.  $\lambda^2$ -test was used to examine the statistical significance. Based on many trials conducted the following model was chosen:

$$\text{Logit}(\rho_i) = b_o + \sum b_i a_i + \sum p a_i + \text{Anim}_o$$

Where,

- $\rho_i$  = Probability of occurrence of mastitis,
- $b_o$  = Intercept,
- $b_i$  = Regression coefficient of the  $i^{\text{th}}$  risk factor,
- $\sum p a_i$  = fixed effect of the  $i^{\text{th}}$  risk factor,
- $\text{Anim}_o$  = Random effect of the  $o^{\text{th}}$  animal,

(i=bacterial group, herd size, origin of the cow, stage of lactation, season of the year).

### RESULTS AND DISCUSSIONS

The analysis of factors associated with intra-mammary infection on the quarter level is always confronted with the complex interrelationship of quarters and the effect of one pathogen on the risk of infection with another. To cope with the first obstacle analysis was performed on both the quarter and cow levels. The close-to-perfect agreement of the differences between groups indicates that for the purpose of this study, the interaction between quarters of individual cows was of minor importance. A possible interaction between pathogens was ignored in this study. This seemed reasonable, because the risk of new infections in cows over

time but in different groups of cows was not studied. Because about 72% of the quarters were negative, the possible effect of the interaction was probably small. Figure 1 displays the frequency distribution of individual bacterial types that have been discovered in the udder quarter and whole milk samples.

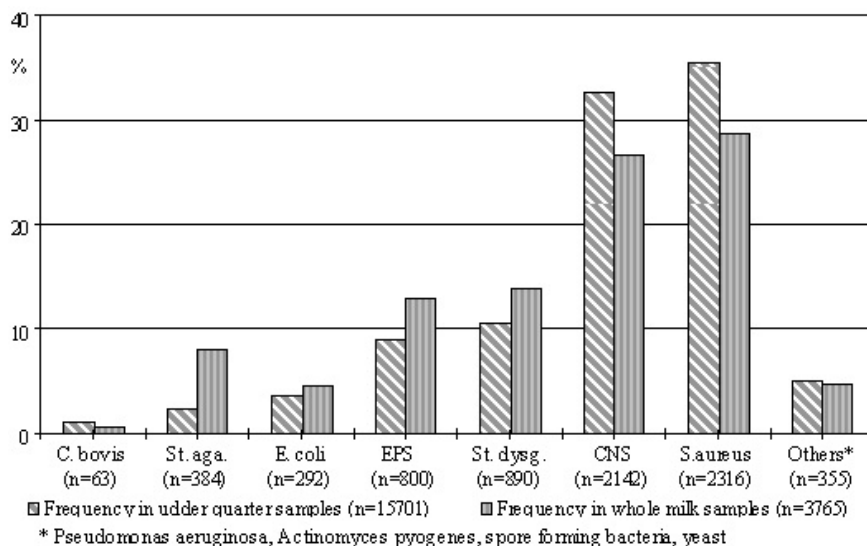


Fig 1: Frequency distribution of bacterial types

Total positive finding was estimated to be 15701 of quarter samples and 3765 of whole milk samples, which represent 27.57% and 49.59% respectively. *S. aureus* (*Staphylococcus aureus*) and CNS (Coagulase Negative Staphylococci) were the most frequently isolated pathogens from the quarter and udder samples (35.50/28.70% and 32.70/26.60%, respectively). This result is in line with Trinidad *et al.* (1990); Nickerson *et al.* (1995); Wilson *et al.* (1997); Waage *et al.* (1999); Dingwell *et al.* (2002) and Cremonesi *et al.* (2006). However, *St. dys.* (*Streptococcus dysgalactiae*) and EPS (Esculin Postive Streptococci) scored a higher frequency in the whole milk samples than in quarter samples (13.90/12.90 vs. 10.60/9.0%, respectively). Distribution of contagious and environmental pathogens shown in Figure 2; revealed that contagious pathogens

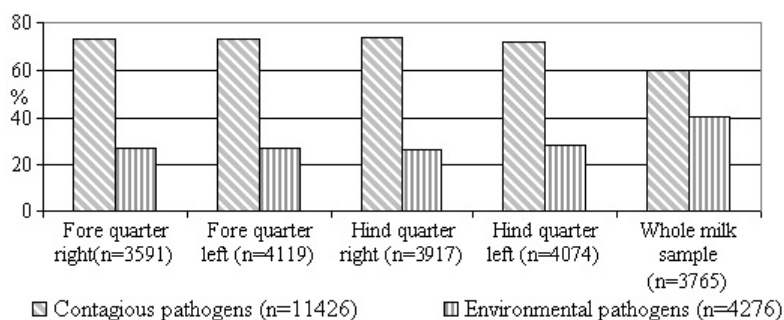


Fig. 2: Distribution of contagious and environmental pathogens

discovered in the udder quarters as well as in the udder samples scored high frequency (72.79%) compared to the environmental pathogens (27.21%). The difference was highly significant ( $p < 0.001$ ). The study also showed that contagious pathogens were highly significantly different ( $p < 0.001$ ) in the chance of causing mastitis than environmental pathogens (Odds ratio "OR", 3.11; 95% CI, 3.08-3.14  $p < 0.001$ ) as shown in Table 2. The result is consistent with the conclusion of Chrystal *et al.* (1999), who stated that nearly all cases of intra-mammary infection occurs as a result of the contagious microorganisms passing through the teat canal. However, different conclusion was declared by Peeler *et al.* (2003) who stated that the importance of environmental vs. contagious pathogens has increased. And as the environmental pathogens are those present in the environment of the animal and could easily be controlled by improving the management practices, the management of the studied farms will be following the management procedure hypothesized that less problem is expected.

Table 2 summarizes the risk factors associated with prevalence of mastitis. Farms studied are fairly of large herd size and to test their effect they were classified into three classes according to the number of the lactating cows they owned. Prevalence of mastitis varied significantly ( $p < 0.001$ ) between the study herds (Table 3).

**Table 2:** Risk factors associated with mastitis, estimated coefficient (b), standard error (S.E.),  $\chi^2$ , Odds ratio (OR) and 95% confidence interval (95%CI)

| Risk factor              | b     | S.E.  | $\chi^2$ | OR   | 95% CI    |
|--------------------------|-------|-------|----------|------|-----------|
| Intercept                | -3.23 | 0.21  |          |      |           |
| Environmental pathogens  |       |       |          | 1    |           |
| Contagious pathogens     | 0.476 | 0.010 | 22.31 *  | 3.11 | 3.08-3.14 |
| Herd size >200 cows      |       |       |          | 1    |           |
| Herd size <200 cows      | 0.101 | 0.010 | 112.50*  | 2.91 | 2.39-3.42 |
| Farm bred heifers        |       |       |          | 1    |           |
| Purchased heifers        | 0.387 | 0.032 | 13.36*   | 2.97 | 3.07-3.88 |
| Late stage of lactation  |       |       |          | 1    |           |
| Early stage of lactation | 0.014 | 0.013 | 0.20+    | 1.14 | 0.99-1.04 |
| Winter                   |       |       |          | 1    |           |
| Summer                   | -0.09 | 0.011 | 07.57*   | 2.51 | 2.44-2.58 |

\* $p \leq 0.001$  (highly significant)

**Table 3:** Prevalence and risk factors of contagious and environmental pathogens as influenced by herd size

| Herd size        | OR (95% CI)      | positive samples | Contagious    |          | Environmental |          |
|------------------|------------------|------------------|---------------|----------|---------------|----------|
|                  |                  |                  | Relative      | Absolute | Relative      | Absolute |
| Small (n=5069)   | 2.91 (2.39-3.42) | 1708 (33.70)     | 1283 (75.12 ) | (25.31)  | 425 (25.31)   | (08.38)  |
| Medium (n=21393) | 1                | 5772 (26.98 )    | 4037 (69.94 ) | (18.87 ) | 1735 (30.06 ) | (08.11)  |
| Large (n=37722)  | 1                | 11884 (31.50 )   | 8324 (70.04 ) | (22.07 ) | 3560 (29.96)  | (09.43)  |
| $\chi^2$         | 112.50*          | 106.80*          |               |          |               |          |

OR= Odds ratio CI= 95% Confidence Interval

\*  $p \leq 0.001$  (highly significant)

Numbers in parentheses indicate percentage

It was higher in small dairy herds (33.70 %; OR, 2.91; 95% CI, 2.39-3.42); relatively lower in medium size herds (26.98 %; OR, 1.00). The result also revealed that contagious pathogens were most frequently isolated. This result indicating that small herds are at a relatively high risk of contracting mastitis than large herds, comparable findings are those of Wilesmith *et al.* (1986); Lafi *et al.* (1994) and Norman *et al.* (2000). This may indicate that large herd size farms were subjected to some modernization in husbandry systems and milking systems and changes of management practices resulted in reducing the frequency of pathogens, specifically the contagious pathogens. Table 4 indicated that purchased heifers were about three times (37.53%; OR, 2.97; 95% CI, 3.07-3.88) at greater risk of acquiring mammary gland infection than farm bred heifers (29.87%; OR, 1.00). This could demonstrate the association between mastitis in the purchased heifers and the incidence of mastitis in the herds from which they were originated. Similar emphases were those of Waage *et al.* (1998); Peeler *et al.* (2000) and González and Wilson (2003). This may explore that foreign animals were more prone to infection than farm bred animals, which may signify the fact that new comers could harbor and bring the pathogens. This fact was also stated by Jones and Bailey (1998). Table 5 presents the prevalence of mastitis causing pathogens (contagious and environmental) within lactation. Early stage and the period of involution of the mammary glands were the most susceptible stages with prevalence of (30.90 %; OR, 1.14; 95% CI, 0.99-1.04) and (29.70%, OR, 1.00) respectively, and lower in mid lactation (23.24 %; OR, 1.00); indicating the possibility of encountering mastitis in early stage of lactation. These results are consistent with previous studies (Kehrli and Shuster, 1994; Radostits *et al.*, 1994; Jones and Bailey, 1998). Stage of lactation investigated as a risk factor was found similar in all of its stages ( $p > 0.05$ ), similar to the result obtained by Biffa *et al.*, (2005). Season of the year was found to exert a significant effect on prevalence of mastitis (Table 6); higher positive findings mostly contagious pathogens were obtained in spring and autumn (32.45%, 31.90% respectively), that was significantly ( $p < 0.001$ ) different than those encountered in summer and winter (28.64%, 23.78%, respectively). Logistic regression analysis revealed that the probability of occurrence of mastitis was fairly high in summer seasons than in winter ( $p < 0.001$ ; OR, 2.51; CI, 2.44-2.58). This finding is in accordance with Waage *et al.* (1998); Waage *et al.* (1999); Petkow *et al.* (2001) and Olivo *et al.* (2005).

**Conclusion:**

The study concluded that herds monitored in this study had a controlled environmental mastitis but still at high risk of intra-mammary infection due to the effect of the contagious pathogens. Herd size was proving to affect the degree of infection to the extent that small herd size was more threatened with udder infection than large herd size. To reduce the prevalence of mastitis, breeding programs should consider risk factors to reduce the causal agents.

**Table 4:** Prevalence and risk factors of contagious and environmental pathogens as influenced by origin of the heifer.

| Origin of the heifer | OR (95% CI)       | positive samples | Contagious    |          | Environmental |          |
|----------------------|-------------------|------------------|---------------|----------|---------------|----------|
|                      |                   |                  | Relative      | Absolute | Relative      | Absolute |
| Farm bred (n=50106)  | 1                 | 14966 (29.87)    | 10817 (72.28) | (21.59)  | 4149 (27.72)  | (08.28)  |
| Purchased (n=5720)   | 2.97 ( 3.07-3.88) | 2147 (37.53)     | 1395 (64.97)  | (24.38)  | 752 (35.03)   | (13.15)  |
| $\chi^2$             | 13.36*            | 248.94*          |               |          |               |          |

OR= Odds ratio CI= 95% Confidence Interval

\* p ≤ 0.001 (highly significant)

Numbers in parentheses indicate percentage

**Table 5:** Prevalence and risk factors of contagious and environmental pathogens as influenced by stage of lactation

| Stage of lactation                 | OR (95% CI)      | positive samples | Contagious   |          | Environmental |          |
|------------------------------------|------------------|------------------|--------------|----------|---------------|----------|
|                                    |                  |                  | Relative     | Absolute | Relative      | Absolute |
| Early stage of lactation (n=44199) | 2.64 (2.49-2.54) | 13657 (30.90)    | 9621 (70.45) | (21.77)  | 4036(29.55)   | (09.13)  |
| Mid stage of lactation (n=2082)    | -                | 486 (23.34)      | 277 (57.00)  | (13.30)  | 209 (43.00)   | (10.04)  |
| Late stage of lactation (n=4195)   | 1                | 1246 (29.70)     | 869 (69.74)  | 20.71    | 377 (30.26)   | (08.99)  |
| $\chi^2$                           | 0.20+            | 21.90 *          |              |          |               |          |

OR= Odds ratio CI= Confidence Interval

\* p ≤ 0.001 (highly significant) + p> 0.05 (not significant)

Numbers in parentheses indicate percentage

**Table 6:** Prevalence and risk factors of contagious and environmental pathogens as influenced by season of the year

| Season           | OR (95% CI)      | positive samples | Contagious   |          | Environmental |          |
|------------------|------------------|------------------|--------------|----------|---------------|----------|
|                  |                  |                  | Relative     | Absolute | Relative      | Absolute |
| Spring (n=12308) | -                | 3994 (32.45)     | 2792 (69.90) | (22.68)  | 1202 (30.10)  | (09.77)  |
| Autumn (n=17640) | -                | 5627 (31.90)     | 3982 (70.77) | (22.57)  | 1645 (29.23)  | (09.33)  |
| Summer (n=15931) | 2.51 (2.44-2.58) | 4562 (28.64)     | 3331 (73.01) | (20.91)  | 1231 (26.98)  | (07.73)  |
| Winter (n=20900) | 1                | 4970 (23.78)     | 3385 (68.11) | (16.20)  | 1585 (31.89)  | (07.58)  |
| $\chi^2$         | 7.57*            | 21.92*           |              |          |               |          |

OR= Odds ratio CI= Confidence Interval

\*p ≤ 0.001 (highly significant)

Numbers in parentheses indicate percentage

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