Genetic parameters for clinical mastitis in Czech Holstein cattle

Ludmila Zavadilová, Eva Kašná, Zuzana Krupová, Michaela Brzáková

Institute of Animal Science, Prague – Uhříněves, Czech Republic

*Corresponding author: zavadilova.ludmila@vuzv.cz


Abstract: Genetic parameters were estimated for clinical mastitis in Czech Holstein cattle. The datasets included 14,329 cows with 28,626 lactations. Clinical mastitis was defined as 0/1 occurrence per lactation. Single- or multi-trait repeatability linear animal models were employed for estimation of (co)variances and prediction of conventional or genomic breeding values. The inclusion of the random herd-year-month effect in the model was analysed. The estimated heritability for clinical mastitis ranged from 2.10% to 2.72%, while permanent environmental variance ratios or random herd-year-month effect ratios were twice higher than heritability. In the multi-trait models, udder type traits, such as fore udder attachment, rear udder attachment, rear udder width, front teat placement and udder depth, were employed. The highest genetic correlations of clinical mastitis occurred with rear udder width (0.41) and the lowest with front teat placement (–0.10). Both the multi-trait model and the genomic model provided breeding value estimates with higher reliability. In contrast, the model with random herd-year-season effects provided breeding values with lower accuracy.

Keywords: dairy cow; udder health; heritability; genomic breeding values; multi-trait model

Clinical mastitis (CM) in dairy cattle has significant negative impacts on animal welfare and the quality of produced milk and fundamentally affects the profitability of dairy cattle farms. The adjustment of environmental conditions can positively influence the prevalence of clinical mastitis in dairy herds. However, resistance to udder diseases in cattle breeds can be increased by selection.

CM traits generally show a lower heritability (up to 10%) (Martin et al. 2018). The positive results of genetic selection were demonstrated previously. Heringstad et al. (2001) affirmed the positive effect of genetic selection on the genetic trend of clinical mastitis in a Norwegian cattle population, where clinical mastitis was a part of the selection goal from 1978.

Currently, health data monitoring is applied for example in the USA, Canada, Austria, Germany, Australia and in the Czech Republic (Kasna et al. 2017).

Although CM is mostly binary, i.e. expressed as 0/1, a linear animal model is regularly used for routine estimates of CM breeding values. Even though a threshold model provides slightly higher estimates of heritability (Carlen et al. 2006), the linear approach is not as computationally demanding compared to the threshold analysis, and the resulting breeding values are not significantly different from those obtained by the threshold model.

As auxiliary traits for CM, in addition to somatic cell scores, udder-type traits are often used. As the best linear type predictor of udder health, udder...
depth is very often highlighted (Govignon-Gion et al. 2016).

In the Czech Republic, the first attempts to estimate genetic parameters and breeding values for Holstein cattle by Wolf et al. (2010) led to a heritability of 0.11, where CM was defined as the number of cases and was analysed by a linear model. Following this study, Zavadilova et al. (2015; 2016) and Kasna et al. (2018) made further estimates of both genetic parameters and breeding values on the available data source. The heritability found by Zavadilova et al. (2015) was approximately 0.10.

The objectives of this study were firstly to estimate genetic parameters for clinical mastitis and selected udder and teat type traits and secondly to analyse using a genomic multi-trait model with random herd-year-season effects in the genetic evaluation of clinical mastitis in Czech Holstein cattle.

MATERIAL AND METHODS

The dataset consisted of 14 329 cows with 28 626 lactations with 19 888 CM diagnoses recorded on 37 farms. Records of clinical mastitis were gathered by farmers and registered voluntarily in the national cattle health monitoring system, “The Diary of Diseases and Medication” (The Diary) (Kasna et al. 2017). In the Czech Republic, this recording system was implemented in August 2018 after a one-year trial period. The system consists of an on-line health recording form for farmers and a simplified key of diagnoses based on the recommendations of the International Committee for Animal Recording. The data on cow type traits were extracted from the official database of the Holstein Cattle Breeders Association of the Czech Republic provided by the Czech Moravian Breeding Corporation.

Records with missing sire identification, incorrect calving dates, or age at first calving outside the 550 to 1 200 days range were excluded. Only cows with 75% or higher proportions of the Holstein breed were included in the analysis. The minimal number of five daughters per sire and 50 cows per herd was required. Only cows with the first lactation were included in the analysis.

The following types of traits were used in the analysis after preliminary analysis: fore udder attachment, rear udder height, udder depth, rear udder width, and front teat placement. The type traits were scored on a nine-point scale. Descriptions of scores of analysed udder and teat linear type traits are provided in Table 1. The numbers of cows per linear type score are shown in Table 2. The average scores of the analysed cows were slightly over the score of 5 for all chosen type traits. Actually, the highest numbers of animals received scores 5 and 6 for all traits, as presented in Table 2.

The SAS® software package, v9.4 (SAS Institute Inc., Cary, USA), was used to perform data editing and calculation of elementary statistics. The incidence of clinical mastitis is reported in Table 3. The lactation incidence rate increased with parity.

Single- or multi-trait linear animal models were used for the estimation of heritability, genetic correlations and breeding value (BV) predictions. The multi-trait linear animal model system included six equations, one for CM and five for udder type traits.

Table 2. Numbers of cows per score

<table>
<thead>
<tr>
<th>Score</th>
<th>udder depth</th>
<th>rear udder height</th>
<th>fore udder attachment*</th>
<th>rear udder width</th>
<th>front teat placement*</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>80</td>
<td>117</td>
<td>265</td>
<td>57</td>
<td>164</td>
</tr>
<tr>
<td>3</td>
<td>357</td>
<td>469</td>
<td>1 336</td>
<td>335</td>
<td>914</td>
</tr>
<tr>
<td>4</td>
<td>1 568</td>
<td>1 791</td>
<td>3 103</td>
<td>1 690</td>
<td>3 376</td>
</tr>
<tr>
<td>5</td>
<td>3 495</td>
<td>4 505</td>
<td>4 048</td>
<td>3 726</td>
<td>3 888</td>
</tr>
<tr>
<td>6</td>
<td>4 049</td>
<td>3 879</td>
<td>3 586</td>
<td>4 465</td>
<td>4 410</td>
</tr>
<tr>
<td>7</td>
<td>3 618</td>
<td>2 686</td>
<td>1 637</td>
<td>3 050</td>
<td>1 327</td>
</tr>
<tr>
<td>8</td>
<td>1 101</td>
<td>809</td>
<td>354</td>
<td>903</td>
<td>250</td>
</tr>
<tr>
<td>9</td>
<td>61</td>
<td>73</td>
<td>103</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Scores 8 and 9 were combined into score 8

For each linear type trait score, one and two were analysed together.
and front teat placement; YEAR-SEASON\_i – the fixed effect of the year and season of classification (26 levels); HERD\_j – the fixed effect of herd \_j (37 levels); CLASSIFIER\_j – the fixed effect of classifier \_j (8 levels); SEASON\_k – the fixed effect of the season of calving (4 levels); The model included linear and quadratic regressions on age at calving \(\beta_1\) \text{AGE}\_i \text{AGE}\_i; \(\beta_2\) \text{AGE}\_i \text{AGE}\_i^2 \text{DIM} \text{DIM}\_m; \(\gamma_1\) \text{DIM}\_m; \(\gamma_2\) \text{DIM}\_m^2; \(\alpha_n\) – the random additive genetic effect of animal \_n (50 784 levels); \(e_{ijklmn}\) – the random residual effect.

For the prediction of genomic breeding values, a single-step procedure was applied (Aguilar et al. 2010; Christensen and Lund 2010). A genomic relationship matrix (G) was calculated according to deviations from the averages of observed and standardised allele frequencies using division by the average value of the diagonal of G. Average of diagonal elements = 1 (Forni et al. 2011). The elements of an additive pedigree relationship matrix for genotyped animals \(A_{22}\) and elements of G have the same average (Vitezica et al. 2011). The total number of effective SNPs used in the calculation of the G matrix was 41 197 with 2 585 effective animals.

The relative breeding values in % (RBV) were calculated using a reference level derived from sires born in 2010 (mean = 100%) and SD = 12. Higher relative breeding values mean a favourable value for CM, i.e., higher resistance to clinical mastitis.

The reliability of CM breeding value was calculated using the standard error of prediction and additive variance estimated by the single- or multi-trait model with or without random effect of herd-year-two calving month.

The DMU program (Madsen and Jensen 2013) was employed for the estimation of genetic parameters and breeding value predictions.

**RESULTS AND DISCUSSION**

**Variances and heritability**

Variances and heritability estimates for CM from the single- and multi-trait models are shown in Table 4. The results for models with the random herd-year-two calving month effects (hym) showed
that including the random \textit{hym} effect in the model results in a slight decrease in heritability together with a distinct increase in permanent environmental variance. The heritability estimates for CM ranged from 0.021 (multi-trait model with random \textit{hym}) to 0.027 (single-trait model). The \textit{hym} effect variation has reached almost the variation of the effect of permanent environment. Therefore, we cannot consider the \textit{hym} effect variation negligible.

An increase in the variance of both effects \textit{hym} and \textit{pe} is associated with a decrease in residual variance and a slight increase in the total sum of variances. According to Schaeffer (2018), the random herd-season effect should describe the time trend and represent contemporary groups. In the genetic evaluation of CM, the random effect of herd-year was used by Jamrozik et al. (2013) when the linear multi-trait animal model was employed. We assume that the time span of our dataset is too short to use the random effect of contemporary group. Jamrozik et al. (2013) analysed a dataset ten times larger, with CM monitored for seven years.

Presented estimates of CM heritability are within the limits indicated in the literature, although they are close to the minimum of published values. For binary CM, the heritability published by Perez-Cabal et al. (2009) for Spanish Holstein cattle was 0.07, and for the number of CM cases it was 0.10. Wolf et al. (2010) and Zavadilova et al. (2015) found very similar values for Czech Holstein. Notably, both studies (Wolf et al. 2010 or Zavadilova et al. 2015) analysed different datasets of Czech Holstein cows from those analysed in the present study. Negussie et al. (2010) reported heritability from 0.03 to 0.06 for binary CM traits estimated by the linear model. Jamrozik et al. (2013) estimated heritability for CM 0.03 (first lactation) and 0.05 (later lactation); CM was recorded from calving to 150 days of lactation and expressed as a binary trait 0/1.

The heritability of type traits (see Table 5) was very similar to values estimated by Nemcova et al. (2011). Except for udder depth, the heritability found by us was approximately 0.09 higher than that found by Nemcova et al. (2011). All chosen udder type traits have been confirmed to have a lower to moderate heritability but at least six times higher than CM, justifying their use in the multi-trait model (Negussie et al. 2006).

### Table 4. Estimated variances, heritability and variance ratios estimated for clinical mastitis by different models with or without random herd-year-two-months (\textit{hym}) effects

<table>
<thead>
<tr>
<th></th>
<th>Single-trait model (estimate ± SE)</th>
<th>Single-trait model with random \textit{hym} (estimate ± SE)</th>
<th>Multi-trait model (estimate ± SE)</th>
<th>Multi-trait model with random \textit{hym} (estimate ± SE)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Additive</td>
<td>0.003 87 ± 0.000 93**</td>
<td>0.003 22 ± 0.000 85**</td>
<td>0.003 72 ± 0.000 85**</td>
<td>0.003 02 ± 0.000 76**</td>
</tr>
<tr>
<td>Permanent environmental</td>
<td>0.006 46 ± 0.001 28**</td>
<td>0.008 04 ± 0.001 24**</td>
<td>0.006 60 ± 0.001 24**</td>
<td>0.008 18 ± 0.001 19**</td>
</tr>
<tr>
<td>Herd-year-two month</td>
<td>0.132 16 ± 0.001 46**</td>
<td>0.125 80 ± 0.001 41**</td>
<td>0.132 17 ± 0.001 46**</td>
<td>0.125 85 ± 0.001 41**</td>
</tr>
<tr>
<td>Total</td>
<td>0.142 49</td>
<td>0.143 75</td>
<td>0.142 49</td>
<td>0.143 77</td>
</tr>
<tr>
<td>Heritability (%)</td>
<td>2.72*</td>
<td>2.24*</td>
<td>2.61*</td>
<td>2.10*</td>
</tr>
<tr>
<td>Permanent environmental variance ratio (%)</td>
<td>4.53**</td>
<td>5.59**</td>
<td>4.63**</td>
<td>5.69**</td>
</tr>
<tr>
<td>Herd-year-season variance ratio (%)</td>
<td>4.65**</td>
<td>4.65**</td>
<td>4.67**</td>
<td>4.67**</td>
</tr>
</tbody>
</table>

Herd-year-season variance ratio = the ratio of herd-year-two month variance from total variance; permanent environmental variance ratio = the ratio of permanent environmental variance from total variance

*P < 0.05; **P < 0.01

### Table 5. Heritability of linear type traits and phenotypic correlations between clinical mastitis and linear type traits

<table>
<thead>
<tr>
<th></th>
<th>Udder depth</th>
<th>Rear udder height</th>
<th>Fore udder attachment</th>
<th>Rear udder width</th>
<th>Front teat placement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heritability</td>
<td>0.41**</td>
<td>0.24**</td>
<td>0.23**</td>
<td>0.19**</td>
<td>0.26**</td>
</tr>
<tr>
<td>Phenotypic correlations</td>
<td>−0.08**</td>
<td>−0.05**</td>
<td>−0.06**</td>
<td>−0.01</td>
<td>−0.02</td>
</tr>
</tbody>
</table>

**P < 0.01
Table 6. Genetic correlations (± SE) between clinical mastitis and linear type traits, the model with random effect hym (above diagonal) and the model without random effect hym (below diagonal)

<table>
<thead>
<tr>
<th></th>
<th>CM</th>
<th>UD</th>
<th>RUH</th>
<th>FUA</th>
<th>RUW</th>
<th>FTP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clinical mastitis (CM)</td>
<td>-0.41 ± 0.084**</td>
<td>0.13 ± 0.093</td>
<td>-0.28 ± 0.093**</td>
<td>0.53 ± 0.103**</td>
<td>-0.10 ± 0.092</td>
<td></td>
</tr>
<tr>
<td>Udder depth (UD)</td>
<td>-0.39 ± 0.077**</td>
<td>0.53 ± 0.046**</td>
<td>0.82 ± 0.030**</td>
<td>-0.14 ± 0.064*</td>
<td>0.23 ± 0.057</td>
<td></td>
</tr>
<tr>
<td>Rear udder height (RUH)</td>
<td>0.13 ± 0.085</td>
<td>0.53 ± 0.046**</td>
<td>0.45 ± 0.057**</td>
<td>0.51 ± 0.058**</td>
<td>0.08 ± 0.067</td>
<td></td>
</tr>
<tr>
<td>Fore udder attachment (FUA)</td>
<td>-0.28 ± 0.086**</td>
<td>0.80 ± 0.030**</td>
<td>0.43 ± 0.057**</td>
<td>0.08 ± 0.073</td>
<td>0.33 ± 0.063**</td>
<td></td>
</tr>
<tr>
<td>Rear udder width (RUW)</td>
<td>0.49 ± 0.093**</td>
<td>-0.13 ± 0.062*</td>
<td>0.52 ± 0.055**</td>
<td>0.08 ± 0.071</td>
<td>0.21 ± 0.070**</td>
<td></td>
</tr>
<tr>
<td>Front teat placement (FTP)</td>
<td>-0.10 ± 0.085</td>
<td>0.23 ± 0.057**</td>
<td>0.06 ± 0.067</td>
<td>0.33 ± 0.062**</td>
<td>0.19 ± 0.069*</td>
<td></td>
</tr>
</tbody>
</table>

*P < 0.05; **P < 0.01

**Genetic and phenotypic correlations**

Phenotypic correlations between linear type traits and CM (see Table 5) were lower than the genetic correlations, consistently with commonly published results (e.g., Lund et al. 1994; Rupp and Boichard 1999). All phenotypic correlations were negative. Except the correlations between CM and rear udder width or fore teat placement, they were significantly different from zero. This relationship means that cows with strong fore udder attachment, shallow udder and high rear udder showed low CM incidence.

Genetic correlations between linear type traits (see Table 6) mostly correspond to the values found by Nemcova et al. (2011). The highest correlations 0.80 and 0.82 (model with random effects hym and model without random effect hym, respectively) were between udder depth and fore udder attachment; 0.53 and 0.53 between rear udder height and udder depth; 0.50 and 0.51 between rear udder height and rear udder width. Almost zero genetic relationships occurred between fore udder attachment and rear udder width (0.08; 0.08) or between front teat placement and rear udder height (0.06; 0.08). Slightly stronger genetic correlations for the former (0.20) as well as for the latter traits (0.22) were presented by Nemcova et al. (2011). Schaeffer (1984) emphasised the importance of using correct genetic correlations in a multi-trait model because variances of prediction error increase when incor-
rect estimates of the correlations between traits are used. Comparisons with previous estimates for Czech Holstein cows confirmed that we also used correct correlation values between CM and selected auxiliary traits.

In Figure 1, the values of the highest least-squares means, i.e., the highest incidence of CM, occurred for extreme scores of 1–2 for each linear type trait, meaning that phenotypically, the highest CM incidence occurred for cows with deep but narrow udder, poorly attached with broadly parted teats. However, especially for rear udder height and udder width and front teat placement, the phenotypic relationship between the incidence of clinical mastitis and the type trait score is not linear. The lowest values of CM for those type traits of clinical mastitis were found for the scores 6 and 7. We can assume that for those traits, the optimum score due to CM incidence is not the opposite extreme score. It should also be emphasised that the small number of cows influences the accuracy of the estimates for extreme levels 1 and 9. The nonlinear relationship between CM and type trait score also diminished the resulting phenotypic correlation, primarily the linear indicator.

Udder depth is often cited as one of the traits associated with clinical mastitis (Seykora and McDaniel 1985; Sorensen et al. 2000; Rupp and Boichard 2003). Genotypic correlations with CM were negative; thus, cows with a high genetic value for the shallow udder will have a high breeding value for resistance to clinical mastitis. Sorensen et al. (2000) came to the same conclusion. Rupp and Boichard (1999) analysed the incidence of clinical mastitis as binary traits in Holstein cows until the 180th day of lactation. They found negative genetic correlations between CM and udder depth (−0.40) and fore udder attachment (−0.36).

Similarly, Lund et al. (1994) emphasised the strong genetic relationship between a higher incidence of clinical mastitis and deep udder in cows. For the Holstein breed, Govignon-Gion et al. (2016) found negative genetic correlations of binary CM with udder depth (−0.30) and fore udder attachment (−0.13); thus, a genetic predisposition to the lower incidence of clinical mastitis will be inherited together with a genetic predisposition to shallower udder and strong fore udder attachment. Jamrozik et al. (2013) found genetic correlations between CM and udder depth (−0.52) and fore udder attachment (−0.46). In the multi-trait model for genetic evaluation of mastitis resistance, they analysed, in addition to udder depth and fore udder attachment, body condition score with genetic correlation with CM (−0.34).

**Genetic trends**

The genetic trends for males or females are presented in Figures 2 and 3. A graphic comparison between models is possible. There was also no substantial change in breeding values, at least in trends, when using a multi-trait model, i.e., a change of up to two units of RBV (+2 percentage points). There is a shift between trends based on different models but not an essential change in the curve shape. Genetic trends for the same models but genomic BV...
Figure 3. Genetic trends for clinical mastitis resistance for sires

vs non-genomic BV follow the same direction and, especially in sires, are very close. Genomic models provided higher average BV than non-genomic models. The average values of trends based on multi-trait models are lower than those based on single-trait models. Additionally, the inclusion of the random hym effects in the model led to a reduction in the BV. All three facts apply to both sexes, but the increase in average BV due to genomic prediction was higher in cows than in sires.

Figure 4. Average reliability of breeding values for cows

Figure 5. Average reliability of breeding values for sires
Reliability

The advantage of using auxiliary traits together with CM records is demonstrated by the increase in reliability when a multi-trait prediction is used. Figures 4 and 5 present the average reliability for the analysed models. For both sexes, the reliabilities were substantially higher for multi- than for single-trait models. The genomic prediction of reliability was more elevated than for non-genomic prediction. The inclusion of the random hym effects in the model caused a low decrease in the average reliability of BVs.

The increase in reliability is approximately 1 unit of reliability, i.e. % (+1 percentage point) for cows with records and nearly 3 percentage points for sires with ten or more daughters on average when the non-genomic and genomic models are compared. As Table 7 shows, the highest reliability occurred in the genomic multi-trait model (81%) for sires, on average 47.85%. The lowest reliability in sires occurred for the single-trait model with random hym (9%), on average 32.60%. For cows with records, the maximum reliability was 43%, but BV with zero reliability was also found for the genomic multi-trait model, single-trait model with random hym and genomic single-trait model with random hym. However, on average, for cows with records, lower reliability was found regularly for every single-trait model compared with the multi-trait models.

The increase in the reliability of the estimate is proportional to a decrease in the error of the estimate. For multi-trait analysis, the decline of the standard errors was confirmed by Gengler and Groen (1997). Based on their results, Negussie et al. (2006) recommended the use of the multi-trait model for traits with low heritability and auxiliary traits with high heritability. Their suggestions are in line with our analysis because CM has much lower heritability than the chosen udder type traits.

Rank correlations

Rank correlations between the breeding values obtained by the single-trait model and BVs predicted by other models are presented in Table 8. It is evident that with the change in the statistical model also comes the change in the ranking of animals according to the breeding values.
ing to predicted BV. This alteration in BV ranking results in the lowering of rank correlations between animal BVs. The rank correlations were calculated for different groups of animals. The lowest rank correlation occurred when the multi-trait model replaced the single-trait model, especially in the cow and all animal groups. In sires or genotyped animals, the substantial drop in correlation values was caused by genomic prediction. The inclusion of the random hym effect in the model resulted in slight decreases in rank correlations in every analysed group.

CONCLUSIONS

This report describes the very positive impact of the multi-trait model on BV reliability. Increasing the amount of information due to the employment of udder and teat type traits in multi-trait model analysis led to a higher increase in reliability compared with the use of genomic prediction. The positive effect of inclusion of the random hym effect in the model could not be confirmed. However, this approach can be useful in CM genetic evaluation in the future when the dataset covers a longer time span. These findings are significant, especially for the genetic evaluation of CM and have a practical impact in genetic selection for increasing clinical mastitis resistance in Czech Holstein cattle.

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Conflict of interest

The authors declare no conflict of interest.

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