

## Genomic prediction of health traits using a mixed reference bull and cow reference population for German Holsteins

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### Abstract

German breeding organizations started a long-term project of whole-herd genotyping, called KuhVision, in 2016. The main goal of the project KuhVision was to establish a cow reference population for more accurate genomic prediction. In addition to recording of currently evaluated traits, more than half of the participating herds recorded also direct health traits. A conventional genetic evaluation for the health traits has been set up and running for German Holsteins for several years. The health traits included clinical mastitis, six claw traits, three reproduction and three metabolic health traits. To increase the size of genomic reference population for the novel health traits, genotyped cows were added to the current reference population composing only genotyped bulls. For April 2019 genomic evaluation, 100,319 or 67,994 cows were included in the German Holstein reference population for clinical mastitis or digital dermatitis, respectively. In contrast to the newly recorded health traits, calf fitness, defined as survival of female calves from day 3 to 458, had a much larger reference population containing 298,499 female calves and 10,424 bulls. Genomic prediction of the health traits was optimized and validated via Interbull GEBV test. Reasonable increase in  $R^2$  values was observed for the health traits from the conventional EBV to the genomic model, despite a short history of whole-herd cow genotyping. Phenotypes of youngest genotyped cows that were not included in the mixed reference population were compared in four groups that were defined based on their candidate GEBV. A strong association was observed between their early candidate GEBV and later own phenotypes. Since April 2019, the mixed bull and cow reference population has been used in routine genomic evaluation of the health traits for German Holsteins.

**Key words:** genomic prediction, health traits, reference cows, mixed reference population

### Introduction

Since 2010 a genomic evaluation system for German Holstein breeds has been set up based on an across country bull reference population, with reference bulls originated mainly from EuroGenomics countries (Liu et al. 2013) and approximately 1000 bulls from North America from birth year 2014 onwards. Due to genomic selection, Holstein reference bulls born in 2010 or later were pre-selected with a high intensity, for instance only 1 out of 40 genotyped male candidates selected for breeding in German Holsteins, evaluation bias by the strong pre-selection was expected to be present in the current multi-step genomic model. More over, the number of reference bulls decreased

significantly from c.a. 1000 progeny-tested bulls in pre-genomic era to about 300 Holstein bulls born in 2017 with daughters in Germany. In fact, male calves that were intended to be genotyped were chosen firstly based on their genomic parental averages, only those exceeding a certain minimum level of genetic merit were genotyped for genomic selection. In summary, use of the bull reference population became more and more prone to bias in genomic prediction.

In 2016, German breeding organizations started a long-term industry project, termed as KuhVision, to establish a reference cow population and maintain or even enhance the accuracy of genomic prediction. By early June 2019, 1421 herds from Germany, Austria and

Luxembourg participated in KuhVision project, representing about 13% of all herdbook cows, approximately 50 herds were added each month. A total of 664 herds recorded also direct health traits. Until June 3<sup>rd</sup>, 2019, more than 379,000 female calves were genotyped.

The objectives of this study were to develop and validate genomic prediction for direct health traits and to compare accuracy of genomic predictions between the mixed and bull reference populations for German Holsteins.

### Materials and Methods

Table 1 shows a list of direct health traits, heritability values, relative weights for sub-indices and total health index. There are currently six claw health traits included in routine evaluation, a new trait interdigital dermatitis that is recorded routinely in Germany will be added to the evaluated traits for the sake of trait harmonization across EuroGenomics countries. A linear multi-trait animal model was used for estimating genetic parameters of those health traits. Heritability values are low for individual health traits. Based on the current weights on individual component traits, heritability values of sub-indices of the four trait complexes were 0.80 for udder health (clinical mastitis), 0.112 for claw health sub-index, 0.066 for reproduction sub-index and 0.042 for metabolic stability sub-index.

Calf fitness (RKF) is defined as survival of female calves from day 3 to 458 in Germany. The whole time period was divided into five intervals that are treated as genetically distinct traits: day 3-14, 15-60, 61-120, 121-200, and 201-458. A linear multiple trait animal model was used to estimate genetic parameters of these five traits. Heritability values were low, ranging from 0.5% to 0.9%. Heritability of the total survival was 1.4%. For conventional evaluation in April 2019, there were 9.6 million Holstein female calves with records and about 17 million animals included in pedigree.

In German milk recording program, disposal reasons for cows have been recorded routinely for a long time. Four disposal reasons were shown to be genetically correlated with direct health traits: disposal reason for infertility (DR4), disposal reason for udder health (DR6), disposal reason for claw problems (DR8), and disposal reason for metabolic diseases (DRX). A linear multi-trait animal model similar to the German longevity model was used to analyze each of the four disposal reasons (Heise et al. 2016). Heritability value estimates were 0.041 for DR4, 0.049 for DR6, 0.053 for DR6 and 0.024 for DRX, respectively. Genetic correlations of the indicator traits disposal reasons with direct health sub-indices were used for blending correlated disposal reasons to the health sub-indices: 0.55 of DR4 with reproduction sub-index, 0.85 of DR6 with udder health / clinical mastitis sub-index, 0.60 of DR8 with claw health sub-index, and 0.80 of DRX with metabolic sub-index. The four blended sub-indices were combined using the weights in Table 1 to obtain the total health index (RZgesund).

Conventional evaluations for German dairy breeds include seven groups of regular traits: milk production traits, somatic cell scores, female fertility, calving, conformation and workability traits, as well as the new trait groups: calf fitness, four disposal reasons, claw health, clinical mastitis, metabolic stability and reproduction diseases. Table 2 summarizes data volumes in April 2019 national evaluation.

Genotype data of 607,503 Holstein animals were used after all selection and editing steps for SNP effect estimation and genomic evaluation in April 2019. Allele frequencies of 45,613 SNP markers were calculated from all the genotyped animals.

### Statistical Analysis

A mixed reference population of bulls and cows was set up for estimating SNP effects using a SNP BLUP model (Liu et al. 2011). There were

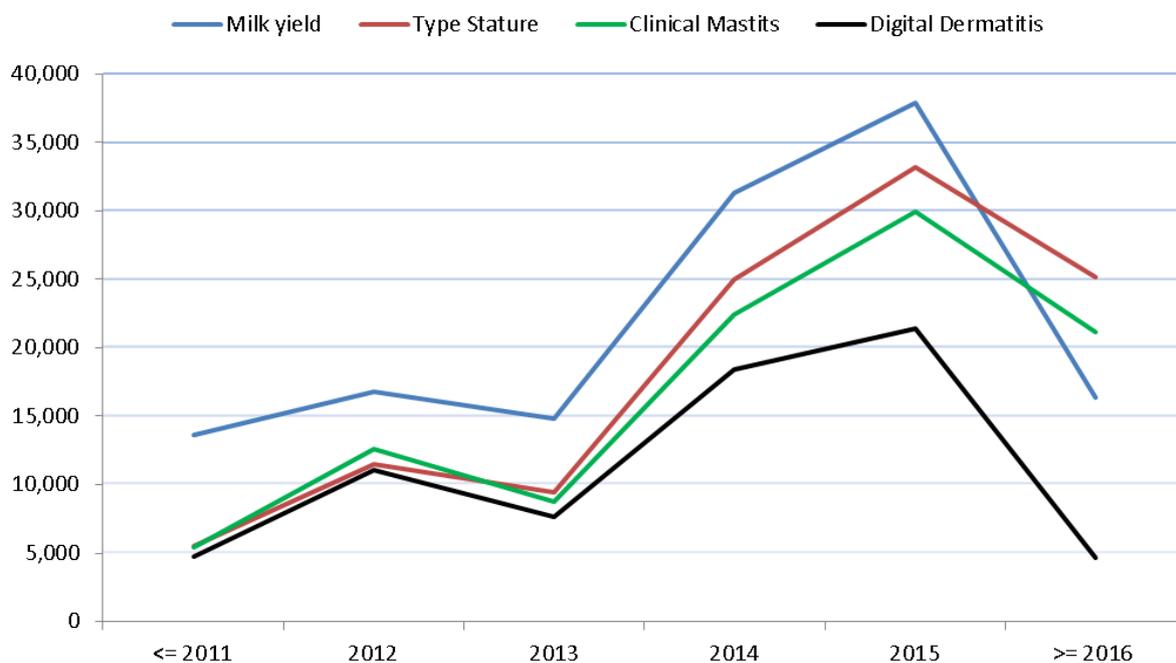
a total of 38,772 Holstein reference bulls for milk yield in April 2019, mainly originating from EuroGenomics countries and increasingly also from USA or Canada. Table 3 shows increases in number of reference cows for selected traits in last four conventional evaluations. It can be seen that the mixed reference population enlarged rapidly in the last two years. Figure 1 shows increasing number of reference cows by birth year for four selected traits. Please note that last birth year 2016 was not completed yet.

Table 4 displays detailed numbers of reference bulls and cows separately for all the novel traits, including the disposal reasons. Number of reference cows for direct health traits ranged from 54,037 for digital phlegmon to 100,319 clinical mastitis. There were c.a. 4000 to 5000 reference bulls with national phenotypes for those traits. Calf fitness had the largest reference population, due to many female calves newly genotyped within the framework of the project KuhVision. A total of 298,499 female calves were included in the mixed reference population and the total number of reference animals reached 308,923

for calf fitness. Genomic reference populations for the four disposal reasons resembled reference population for longevity, due to their similar data structures. Across all the four indicator traits, approximately 12,000 reference bulls and 92,000 to 155,000 reference cows with national phenotypes were present in the reference populations.

*Use of phenotype data of cows and bulls*

Phenotype data of a bull from German national and MACE evaluations were qualified for being used in SNP effect estimation or for calculating conventional parental average (PA) / pedigree index (PI) of genotyped candidates, if his number of herds was 10 or more. In general, a genotyped cow was considered to be qualified for the reference population, if she had her own phenotype records. For test-day data of milk production traits or somatic cell scores, at least six test-day records in first lactation were required for reference cows. To avoid proportionally too many culled cows entering the reference population earlier than non-culled cows, a waiting time period of 250 days from first calving was imposed on direct longevity



**Figure 1.** Number of reference cows by birth year in April 2019 evaluation

trait. Phenotypic data of reference cows were used for computing PA / PI of genotyped candidates, in contrast phenotype data non-reference cows were ignored in this process for avoiding too many cows. When a bull and his daughters were both present in the mixed reference population or in a phenotype data set for PA/PI calculation, EDC of the bull were adjusted for the contribution by those daughters. Additionally, DRP of the bull was corrected for DRP of the daughters and EBV of their dams. The adjustments were made to avoid double counting the contributions by those daughters, because the reference daughters might be genetically superior to non-reference daughters of the bull.

*A new genomic evaluation system*

The mixed reference population of bulls and cows was much bigger than the bull reference population, thus estimation of SNP effects was more time consuming. As for the bull reference population, the SNP effect estimation for the mixed reference population took advantage of multiple threads. Via UC4 more than 60 traits were optimally distributed to six Linux servers for reducing total clock time. Routine genomic evaluation based on the mixed reference

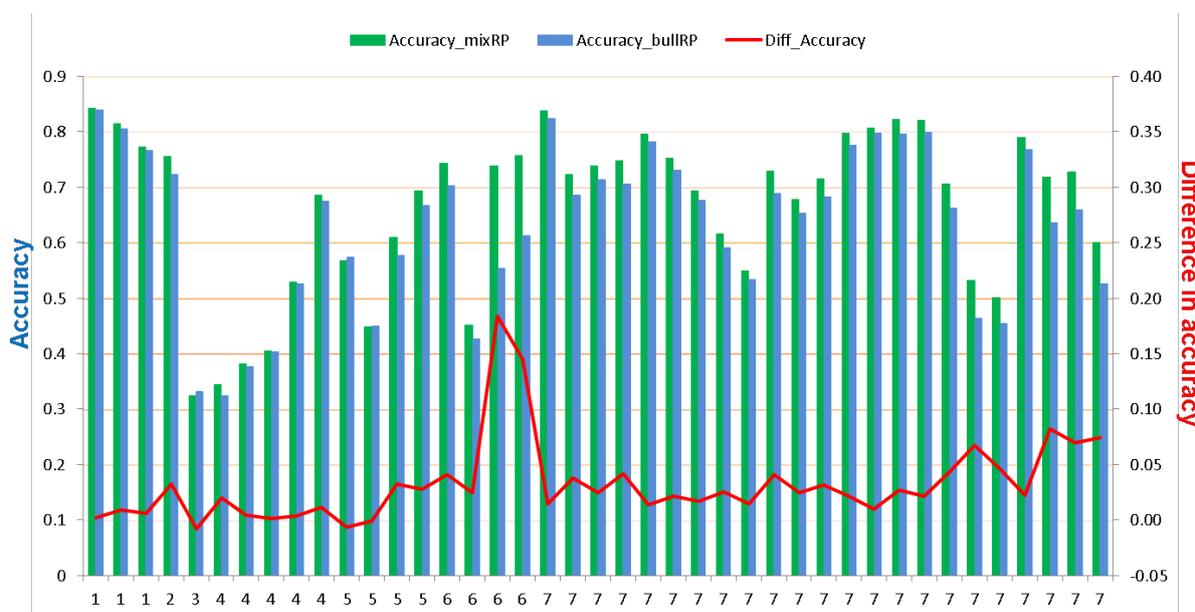
population was conducted on a weekly basis, same as for the bull reference population.

**Results & Discussion**

***Comparing prediction accuracies of the mixed and the bull reference populations***

To derive prediction accuracy of the mixed reference population, phenotypes of last generation reference animals needed to be removed for simulating a forward prediction, like what was done routinely for the bull reference population. However, due to the short history of cow genotyping, the majority of the reference cows were born in last three years 2014 to 2016, four years of youngest reference cows could not be deleted for the validation purpose. Because we were primarily interested in whether the mixed reference population resulted in higher prediction accuracy than the current bull reference population, identical data sets were applied to the two types of reference population for genomic validation. Phenotype and genotype data were taken from August 2018 national and MACE evaluations.

As validation bulls, 606 Holstein bulls with daughters in Germany, which were born in the last two birth years of 2012 and 2013, were selected for the mixed as well as bull reference



**Figure 2.** Observed correlation (accuracy of prediction) between candidate GEBV and later deregressed EBV of validation bulls for the mixed bull reference populations

populations. Additionally, all reference daughters of the validation bulls, 8415 reference cows in total, were removed from the truncated reference population as well. For the two truncated reference populations, SNP effects were estimated for all regular traits and GEBV test (Mäntysaari et al. 2010) was performed. For the accuracy comparison study, the new health traits were not included for convenience, because the mixed reference population was used from the beginning and the bull reference population was never set up.

Figure 2 shows observed correlation, representing accuracy of genomic prediction which was not adjusted for reliability values of the validation bulls, between candidate GEBV and later deregressed EBV of the validation bulls for all the regular traits. The secondary Y axis shows difference in the correlations between the mixed and bull reference populations. Overall, the two types of reference populations resulted in high and very similar accuracies of prediction. The accuracy differences were above zero for all the regular traits, indicating that the mixed reference population had always a higher prediction accuracy than the bull reference population, even for traits with low heritability values. Two traits from group 6, two workability traits, showed a much higher accuracy for the mixed reference population because of a change in trait definition over time. Majority of young (reference) cows were recorded increasingly with electronic milk flow rather than subjectively scored for milking speed. Because the young validation bulls had daughters with the new trait recorded, the mixed reference population was favored in the accuracy comparison for the two workability traits.

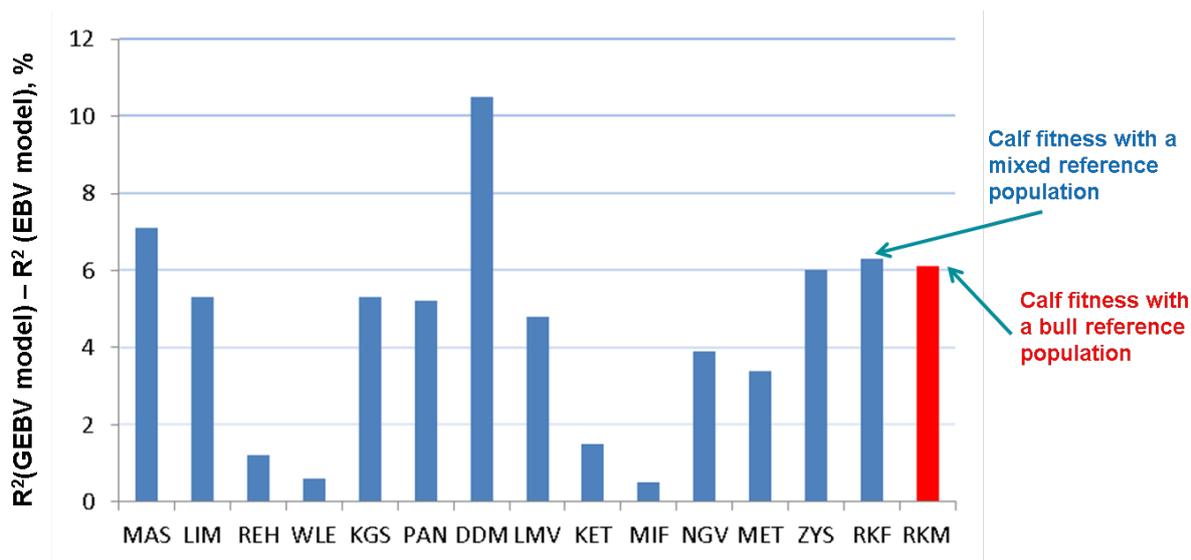
#### ***A special genomic validation for calf fitness***

Due to logistic constraints, female calves from some herds were not genotyped within one or two weeks after birth, before they were culled or had left the farms for some reasons. Because of the early measured trait calf survival evaluated the survival from day 3 up to day 458,

there may be proportionally too few reference female calves not survived in first or two weeks. On the other hand, EBVs of sires of calves, reference bulls, contain information from both the culled and survived calves for the time period. A question arose if the bull reference population should be kept for the early measured trait. Therefore, a special genomic validation was conducted to investigate impact of this kind of selective phenotyping on genomic prediction using both the bull and the mixed reference populations.

As a consequence of the short history of female genotyping, only last two years of data were removed for the genomic validation. Youngest reference bulls born in 2014 and 2015 were chosen as validation bulls. In total, 530 youngest of all 10,352 reference bulls from the full evaluation were defined as validation bulls. Additionally, 102,470 youngest female reference animals born in 2017 and 2018 were removed from the complete mixed reference population for the validation study. A truncated bull reference population included 9577 reference bulls, all born before 2014. In comparison, the truncated set of the mixed reference population included additionally 143,710 female calves that were born before 2017. The two reference populations were compared in both prediction accuracy and bias.

GEBV test (Mäntysaari et al. 2010) was applied to the two types of reference population. Because of the identical validation bulls for the two types of reference population, reliability values of the GEBV model and EBV model can be directly compared. Difference in observed  $R^2$  value between the EBV and GEBV model represented the increase in prediction accuracy due to genomic information. For the mixed reference population, an increase of 6.3% was observed from the EBV to GEBV model, whereas the increase was slightly lower, 6.1%, for the bull reference population. Regression coefficient of the GEBV model showed no difference between the two types of reference



**Figure 3.** Observed  $R^2$  increase from the EBV to GEBV model for the mixed reference population of bulls and cows for the new health traits

population either. Based on the validation  $R^2$  and regression coefficient values, we can conclude that the missing female calves in first two weeks did not lead to different genomic prediction for the two types of reference populations. The mixed reference population will thus be used for the calf fitness trait as for all the other traits. Reasons for the negligible impact of the missing female calves in the mixed reference population may be that the first two weeks represented only a small part of the time period for the calf fitness and secondly DRP of reference bulls in the mixed reference population contained information from the culled or missing female calves in the first two weeks after birth.

#### ***Prediction accuracy increase due to genomic information for the new health traits***

According to the GEBV test (Mäntysaari et al. 2010), model  $R^2$  values of the EBV model and GEBV model indicate the contribution of genomic data to prediction. Figure 3 shows model  $R^2$  value increases observed in the genomic validation from the EBV to GEBV model for all the new health traits using the mixed reference population. The model  $R^2$  values were not adjusted for reliabilities of the validation bulls. It can be seen that the GEBV

model has higher  $R^2$  values than the EBV model for all the traits, due to the contribution of genomic information. Some traits like WLE or MIF have lower  $R^2$  increase than the others, this may be caused by lower heritability, lower reliability values of the validation bulls or different incidence levels in the reference and validation populations. As stated previously, two types of reference population were compared for the early measured trait calf fitness, the  $R^2$  value increases are similar for the bull and the mixed reference population.

#### ***Comparison of phenotypes of validation cows***

In contrast to validation bulls, validation cows have significantly lower reliability, especially for low heritable traits. Application of the GEBV test to validation cows may produce illogical results for some traits. Therefore, we used a different validation procedure for the cows by comparing their own phenotypes in groups that were defined based on their early candidate GEBV. A total of 8415 validation cows were selected which were daughters of the national validation bulls, neither the bulls nor the cows were present in the truncated reference population. GEBV of the validation cows as candidate without own phenotypes were used to equally divide them into four quarters. We

compared phenotype values of the validation cows for the four GEBV quarters. First lactation 305-day milk yields of 6992 validation cows with first lactation completed were analyzed with:

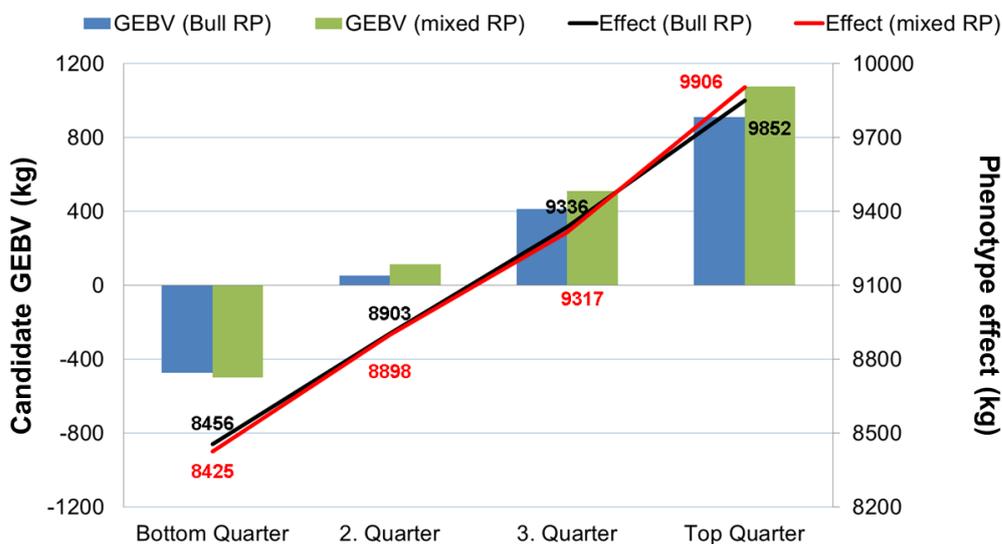
$$y_{ij} = \mu + herd_i + GEBVgroup_j + e$$

to account for effects of herds. Figure 4 shows averages of later phenotypes of the four quarters of the validation cows. Phenotype effects of the validation cows shown in Figure 4 were calculated as sum of the general mean  $\mu$  and effect of GEBV group.

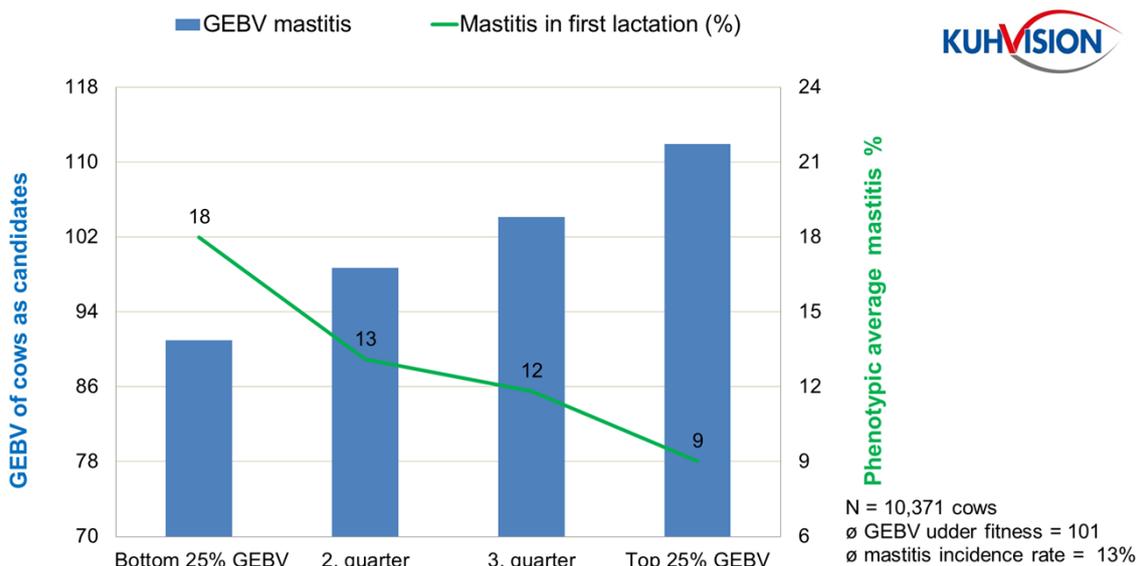
GEBV of the validation cows as candidates without own phenotypes showed more differences between the top and bottom quarters for the mixed reference population than the bull reference population, reflecting the fact there was more variation in SNP effect estimates of the mixed than the bull reference population. The two solid lines show phenotype effects of the four quarters of the validation cows defined using their candidate GEBV. It can be seen that the higher the early candidate GEBV of the validation cows, the higher their later phenotype effects. This clearly demonstrates that the early candidate GEBV can predict well later phenotypes, at least for the average of the four quarters. The two solid lines for phenotypes of the validation cows, red for the mixed and black for the bull

reference population, crossed with each other, indicating that the top GEBV quarter of the validation cows produced more milk, whereas the bottom GEBV quarter gave less milk for the mixed than the bull reference population. Difference in phenotype values between the top and bottom quarters is 1481 kg or 1396 kg milk for the mixed than the bull reference population, respectively. The two values have a difference of 85 kg milk, which corresponds to 15% genetic standard deviations. These results demonstrate that the mixed reference population can better differentiate good from bad cows based on its early candidate GEBV than the bull reference population.

For the new health traits, no comparison of the two types of reference population was made as for the regular traits. However, at the April 2019 routine genomic evaluation, there were young genotyped cows not qualified yet to be included in the reference population, their GEBV were estimated without considering their own phenotypes, although their sires may be reference bulls. Phenotypes of those young genotyped, non-reference cows can be compared against their candidate GEBV.



**Figure 4.** Comparison of phenotypes of validation cows between the bull and the mixed reference population for milk yield



**Figure 5.** Comparison of mastitis incidence levels in first lactations between four quarters of clinical mastitis GEBV of 10,371 young genotyped, non-reference cows in April 2019 genomic evaluation

Figure 5 shows a routine verification of candidate GEBV of young genotyped, non-reference cows by comparing their phenotypes. It can be seen that the top GEBV quarter of 10,371 young genotyped, non-reference cows, whose GEBV were estimated without own phenotypes, had only 9% clinical mastitis; in contrast, the bottom quarter of the cows with worst candidate GEBV in clinical mastitis had 18% clinical mastitis. In summary, this graph shows a strong association between early candidate GEBV and later own phenotype for cows for the new health trait. Like Figure 5 for clinical mastitis, we found also similar results for all the other health traits.

For early measured traits, like direct effects of stillbirth (SBd) or calving ease (CEd) and calf fitness (RKF), reference cows did not represent dead calves for SBd or CEd or female calves that left farm before being genotyped. Using phenotypes of only the reference cows may result in biased genomic prediction. However, DRP of bulls contain information of dead calves for SBd or CEd or non-genotyped female calves for RKF, thus for the mixed reference population the impact of phenotypes

of reference cows or female calves is limited for the early measured traits.

With youngest cows included in a genomic reference population, the distance of selection candidate, like a genotyped male calf, to the mixed reference population is shorter than to the bull reference population, when the male calf's sire or male ancestors have daughters only in foreign countries. A mixed reference population contains genomic information on all four selection paths, whereas only the sire to son selection path is present in the bull reference population.

### Conclusions

Genomic selection in German Holsteins since 2010 has doubled genetic progress on a yearly basis, due to shorter generation interval and higher selection intensity. At the national level more than 80% of semen usage is from young genomic bulls in Germany. The EuroGenomics bull reference population has already led to a high accuracy of genomic prediction. To maintain or even further enhance the prediction accuracy, German breeding organizations started the long-term whole-herd genotyping

project KuhVision in 2016. By June 2019 the number of genotyped Holstein female calves exceeded 300,000. Size of the mixed reference population of bulls and cows has increased significantly.

The same phenotype and genotype data sets were used to compare prediction accuracies of the mixed reference population to the current bull reference population. Slightly higher accuracy, observed correlation between DRP and GEBV for validation bulls, was found for the mixed reference population than the bull reference population for all traits, even for the low heritable traits like fertility. No significant differences were found in regression coefficients between the two reference populations. Validation cows showed more variation in (later) own phenotypes based on their (early) GEBV from the mixed reference population than the bull reference population. By applying the GEBV test to the new health traits, notable increase in model  $R^2$  value from the EBV to GEBV model was observed for all the traits, despite of using only national phenotype data. For the early measured calf survival trait, no differences in prediction accuracy and bias were found between the mixed and the bull reference populations, although some female calves were culled or left farms without being genotyped.

A series of test evaluations were conducted using the mixed reference population in parallel

to the bull reference population. Genomic evaluation results were distributed to breeding organizations and farmers prior to the official introduction of the mixed reference population in April 2019 for German Holsteins.

## References

- Heise, J., Liu, Z., Stock, K.F., Rensing, S., Reinhardt, F., and Simianer, H. 2016. The genetic structure of longevity in dairy cows. *J. Dairy Sci.* 99:1253-1265.
- Liu, Z., Aamand, G.P., Fritz, S., and Schrooten, Ch. 2013. Comparison of national genomic predictions of EuroGenomics exchanged bulls. *Interbull Buletin.* 47, 38-42.
- Liu, Z., Seefried, F., Reinhardt, F., Rensing, S., Thaller, G., and Reents, R. 2011 Impacts of both reference population size and inclusion of a residual polygenic effect on the accuracy of genomic prediction. *Genet. Sel. Evol.* 43:19.
- Mäntysaari, E., Liu, Z., and VanRaden P.M. 2010 Interbull validation test for genomic evaluations. *Interbull Bulletin* 41.

**Table 1.** Direct health traits evaluated for German Holstein breeds

| Trait complex | Name of individual trait | Weight for heritability | Weight for sub-index (%) | Sub-index heritability | Weight for total health index (%) |
|---------------|--------------------------|-------------------------|--------------------------|------------------------|-----------------------------------|
| Udder health  | Clinical mastitis        | 0.080                   | 100                      | 0.080                  | 40                                |
|               | Digital dermatitis       | 0.117                   | 30                       |                        |                                   |
| Claw health   | Laminitis                | 0.030                   | 15                       | 0.112                  | 30                                |
|               | White line disease       | 0.060                   | 15                       |                        |                                   |
|               | Claw ulcers              | 0.110                   | 15                       |                        |                                   |
|               | Digital phlegmon         | 0.085                   | 15                       |                        |                                   |
|               | Interdigital hyperplasia | 0.113                   | 10                       |                        |                                   |
| Reproduction  | Ovarian cycle disorders  | 0.058                   | 50                       | 0.055                  | 20                                |
|               | Retained placenta        | 0.033                   | 25                       |                        |                                   |
|               | Endometritis / Metritis  | 0.032                   | 25                       |                        |                                   |
| Metabolic     | Displaced abomasum left  | 0.029                   | 50                       | 0.042                  | 10                                |
| Stability     | Milk fever               | 0.041                   | 25                       |                        |                                   |
|               | Ketosis                  | 0.027                   | 25                       |                        |                                   |

**Table 2.** Data used in April 2019 national evaluation for German dairy breeds

| Regular traits                          | Number of cows with data (number of records) | New traits          | Number of cows with data |
|---|--|---------------------|--------------------------|
| Milk production and somatic cell scores | 21,984,517 (420,238,055 test-day records)    | Disposal reasons    | 12,083,208               |
| Conformation                            | 2,856,592                                    | Claw health         | 424,240                  |
| Workability                             | 8,591,548                                    | Clinical mastitis   | 659,870                  |
| Longevity                               | 14,735,985                                   | Metabolic stability | 526,432                  |
| Female fertility                        | 19,825,324 heifers or cows                   | Reproduction        | 611,492                  |
| Calving                                 | 30,424,609 calves or cows                    | Calf fitness        | 9,558,532 female calves  |

**Table 3.** Numbers of reference cows in past evaluations for German Holsteins

| Evaluation    | Cows (milk production traits) | Cows (conformation) | Female animals (calf fitness) | Cows (clinical mastitis) | Cows (digital dermatitis) |
|---------------|-------------------------------|---------------------|-------------------------------|--------------------------|---------------------------|
| April 2018    | 90,737                        | 73,614              |                               | 61,550                   | 40,915                    |
| August 2018   | 105,390                       | 87,150              |                               | 75,789                   | 46,276                    |
| December 2018 | 117,644                       | 97,266              | 262,818                       | 87,809                   | 57,392                    |
| April 2019    | 130,960                       | 109,746             | 298,499                       | 100,319                  | 67,994                    |

**Table 4.** Mixed genomic reference populations for the new health traits in April 2019

| Trait                                 | Abbreviation | No. of bulls | No. of cows | Total   |
|---------------------------------------|--------------|--------------|-------------|---------|
| Clinical mastitis                     | MAS          | 4998         | 100,319     | 105,317 |
| Interdigital hyperplasia              | LIM          | 4214         | 67,691      | 71,905  |
| Laminitis                             | REH          | 4229         | 67,734      | 71,963  |
| White line disease                    | WLE          | 4219         | 67,740      | 71,959  |
| Sole ulcers                           | KGS          | 4045         | 61,118      | 65,163  |
| Digital phlegmon                      | PAN          | 3974         | 54,037      | 58,011  |
| Digital dermatitis                    | DDM          | 4140         | 67,994      | 72,134  |
| Displaced abomasum left               | LMV          | 4510         | 78,621      | 83,131  |
| Ketosis                               | KET          | 4709         | 90,211      | 94,920  |
| Milk fever                            | MIF          | 4632         | 86,959      | 91,591  |
| Retained placenta                     | NGV          | 4873         | 87,585      | 92,458  |
| Endometritis                          | MET          | 4413         | 74,197      | 78,610  |
| Ovarian cycle disorders               | ZYS          | 4355         | 60,625      | 64,980  |
| Calf fitness                          | RKF          | 10,424       | 298,499     | 308,923 |
| Indicator traits: disposal reason for |              |              |             |         |
| infertility                           | DR4          | 11,901       | 92,204      | 104,105 |
| udder health                          | DR6          | 12,431       | 129,949     | 142,380 |
| claw health                           | DR8          | 12,431       | 124,671     | 137,102 |
| metabolic disorders                   | DRX          | 12,399       | 155,025     | 167,424 |