Introduction of single-step genomic evaluations in German Holsteins

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Abstract

In April 2025, we introduced single-step genomic evaluations for all traits that are subject to routine genetic evaluations in German Holstein. With all models, we estimate the same main effects as with the former conventional genetic evaluations of Holsteins. In addition, a fixed regression on the inbreeding coefficient was added to all the models. With the introduction of single-step models, reliabilities of GEBVs increased for all traits. This increase is especially pronounced in young animals with no own or offspring performance. As expected, the increase in reliability was greatest for many functional traits: longevity and direct calving index: +0.11, maternal calving index: +0.13, young stock survival and health index: +0.14, while it was lower for production traits: production index: +0.04. Additionally, validations with 2 and 4 years of right-truncated data confirm a substantial increase in the predictive ability of genomic GEBVs compared to the previous multi-step model: correlations of purely genomic GEBVs of young bulls with their later daughter-proven GEBVs are higher for all traits with the single-step model. Again, this increase in predictive ability is highest for the functional traits and lower for the production traits. With publication dates in April, August, and December, we conduct main runs with updated phenotypic information three times a year. In these main runs, we include MACE information from the respective current Interbull MACE run. In addition to these full runs, we conduct weekly genomic evaluations, for which we use the estimates of the SNP-effects and the residual polygenic effects from the main runs and apply them to the newly genotyped animals.

Key words: single-step model, genomic evaluation, genomic validation, Holstein cattle

Introduction

A multi-step SNP BLUP genomic model (MSM, Liu et al. 2011) was used for routine genomic evaluations of German Holsteins from August 2010 to December 2024. After the first publications of single-step genomic models (Aguilar et al. 2010; Christensen and Lund 2010), tremendous efforts were devoted to research and development in Germany with the goal of implementing a single-step SNP BLUP model (SSM, Liu et al. 2014) for routine genomic evaluations in German Holsteins.

There are a total of 10 trait groups evaluated routinely for German Holsteins. Logically, the SSM model must be implemented to all the trait groups simultaneously, to maintain the current weights of individual traits on total merit indices such as German RZG, RZ€ and RZOeko. To minimize the impact of genomic model change on genomic selection, all functionalities and features of the current MSM conventional and genomic evaluations must be retained as much as possible.

Although the conventional evaluations in MSM were multi-trait models for all trait groups, the genomic evaluations were single-trait models applied to deregressed conventional EBV (DRP) of reference bulls and cows (Liu et al. 2011). For instance, a multi-lactation random regression model was used to analyze test-day milk yields in the conventional evaluation of German Holsteins (Liu et al. 2004), but the evaluated trait in the genomic model of MSM was 305-day lactation milk yields combined over the first 3 lactations, calculated as a linear function of the genetic random regression coefficients from the conventional test-day model. In contrast to the MSM, SSM implicates direct modeling of the genomic information based on the national test-day milk yields. With the SSM, we therefore estimate for the first time SNP effects directly on the level of test-day random regression coefficients for production traits (Alkhoder et al. 2022; Alkhoder et al. 2024).

To enhance the reliability of genomic estimated breeding values (GEBV) and to represent foreign genetics in the German population as unbiased as possible, genotyped foreign bulls were included in the German Holstein genomic evaluation under the MSM model. The trait definitions of all traits evaluated in the Holstein MACE evaluation were used for the German genomic evaluation, e.g., the combined lactation 305-day milk yield under the MSM model as described above. With the introduction of the SSM model, phenotypic data of foreign cows is also included as in MSM model, via the MACE proofs of their sires, which increases the reliability of genomic prediction. In the SSM model, deregressed MACE EBVs of foreign bulls are treated as the same trait or a correlated trait as the national estimation traits, depending on the trait groups.

The objectives of this study were 1) to describe technical details of the genomic model upgrade from the MSM to the SSM model; and 2) to compare accuracy and prediction bias of the two genomic models via genomic validation.

Materials and Methods

Data sets for routine single-step evaluations

Phenotype data of cows and bulls

All national phenotypic data as used in the previous conventional evaluations are now used in SSM: phenotypic cow data recorded since 01.01.2000. As in the previous conventional and genomic MSM models, cows or calves with their own phenotypic records must have their sire and maternal grandsire known. The breeds of sire and maternal grandsire must be consistent with that of the animal. Other plausibility checks and edits on the data are also kept with SSM as they were for the conventional model in MSM. Because of the integration of foreign MACE data in the single-step evaluation, bulls with foreign daughter information in MACE are required to be born in 1995 or later to be consistent with the left truncation of national phenotype data.

Genotype data

Unlike the previous MSM model, genotype records of animals born before 2005 are no longer used in the SSM model to avoid possible negative impact of selective genotyping in the early years of genomic selection. Genotype imputing and routine checks on the genotype data are kept with SSM.

Pedigree data

In contrast to the trait-dependent pedigree processing procedures in the previous MSM evaluations, we now apply the same procedures to the sub-pedigrees for all trait groups: starting from youngest genotyped animals (including embryos) or cows / calves with phenotype records, a maximum of 20 generations of ancestors is traced back in the main pedigree. Additionally, at least 3 generations of ancestors are included for the oldest animals with phenotypic data, e.g., bulls with foreign daughters. Pedigree-based inbreeding coefficients are computed once, using all animals present in Germany's pedigree database for dairy breeds and the resulting inbreeding coefficients are then used to build the diagonals of the inverse relationship matrix and to define the fixed effect on the inbreeding coefficient in all SSM evaluations.

Data sets for two genomic validation studies

For a comparison between SSM and MSM, two comprehensive genomic validation studies were performed. For a 2-year validation, phenotypic data from the most recent two years were removed from the full evaluation 2304 to simulate an earlier evaluation in April 2021 (2104). The phenotypic, genotypic and pedigree data for the two evaluations 2304 and 2104 were already described in Tables 1 and 2 in the paper (Liu et al. 2023). Corresponding to the two different national cow data sets in evaluations 2304 and 2104, bull MACE data were obtained from the MACE evaluations in April 2023 and 2021.

In addition to this 2-year validation, a 4-year validation was conducted: with the results of a truncated conventional evaluation based on data as of the August 2020 evaluation (2008), Germany had also participated in Truncated MACE (TMACE) in October 2024. These TMACE EBVs were then considered in a subsequent, reduced SSM evaluation 2008, simulating an earlier SSM evaluation as of August 2020. Table 1 describes the data used for the full SSM evaluation 2408 and the reduced SSM evaluation 2008 for four test-day traits.

A genomic validation was conducted for all evaluated traits of German Holsteins, including MACE traits as well as national-only traits with both the 2-year and the 4-year validation data sets. For the earlier validation with 2304 and 2104 data, a linear regression test (LR, Legarra and Reverter, 2018) was applied. Using Interbull's GEBV Test software made available in September 2024, we conducted genomic validation with the second validation data set 2408 versus 2008.

Trait-wise development of SSM

Until today, a total of 10 trait groups are considered in routine evaluations of German Holsteins using: a multi-lactation random regression test-day model for milk, fat and protein yields and somatic cell scores (TDMS), a multi-trait model for 25 conformation traits (CONF), a multi-trait linear animal model for direct functional longevity (LONG), a multi-trait model for six female fertility traits (FERT), a multi-parity maternal-effect model for calving ease and stillbirth (CALV), a multi-trait model for workability traits (WORK), a multi-trait model for 16 direct health traits (HEAL), a multi-trait linear model for calf fitness (CFIT), a multi-trait random regression model for feed efficiency (FEFS), and a multi-trait model for four disposal reasons (DPRS) that serve as indicator traits for the health traits only.

Table 1. Description of the data sets for the single-step full and truncated evaluations of test-day milk, fat and protein yields and somatic cell scores

Frequency	Single-step evaluation 2408	 2008
Genotyped animals	1,631,844 Holstein animals (1,433,599 females and 198,245 males)	
Phenotyped cows or bulls	14,189,574	12,195,546
Test-day records	277,884,084	235,578,132
Genotyped or phenotyped animals	15,165,965	13,565,673
Animals in pedigree SSM	22,743,486	20,983,007
Reference animals MSM	604,587	246,910

Starting with the simplest evaluation model for the conformation trait group CONF with only 1 record per cow, we tested the SSM for the genomic evaluation of German Holsteins (Alkhoder et al. 2021). After a positive experience with the simplest statistical model, we extended the SSM test implementation to the two most complex statistical models: the multi-lactation random regression test-day model for milk production traits and somatic cell scores TDMS (Alkhoder et al. 2022) and the multi-parity maternal-effect model for calving ease and stillbirth CALV (Alkhoder et al. 2022a). Having successfully tested the SSM model for the three trait groups CONF, TDMS and CALV, we finally moved on to include all the remaining 7 trait groups and completed the SSM testing processes in 2023.

Integration of bull MACE data

Due to the simplicity of this approach, the previous conventional evaluations from MSM are still run in parallel to SSM at every main run (3x per year) to provide genomic-free EBVs to Interbull as input for MACE.

The current Interbull MACE evaluation uses a single-trait multi-country model. For instance, the MACE trait of milk yield for German Holstein is 305-day lactation milk yield combined over the first three lactations, which corresponds to an aggregated linear function of the 9 random regression coefficients used in the German random regression test-day model (Liu et al. 2004; Alkhoder et al. 2024). Unlike the MSM genomic evaluation that was previously applied to DRP from the aggregated, combined traits on lactation basis, the SSM model analyzes original cow test-day milk yields also on the genomic level. Another example is the calving trait model: a multi-parity maternal-effect single-step model for calving ease and stillbirth phenotypic records of calving cows in the first three parities. In comparison, DRP of first parity calving ease or stillbirth, defined as official breeding values for publication, were pseudo-phenotypic records for the single-trait MSM evaluation.

Across all the trait groups, the trait definition for the single-trait MSM model was the German official breeding value for publication as well as the MACE trait, some of which were aggregated from the original national traits / estimation variables.

Deregressed MACE EBVs are included in the SSM for the different trait groups as correlated traits, using the national genetic covariance matrices and the weights used to combine national traits to MACE traits to derive covariances between national and MACE traits. In most cases, the genetic correlation between the combination of national traits and the MACE trait is assumed to be 1. Only in some cases, this correlation was pruned to 0.97 to enhance convergence.

Calculation of indices from the SSM

After completion of the test implementations for all the 10 trait groups, we upgraded our calculation procedures for various sub-indices, and the German total merit indices *RZG* and *RZ€* for the SSM system slightly.

The previously used non-linear index for longevity (RZN, Heise 2017; Heise et al. 2016) was replaced with its linear approximation and we adapted the genetic standard deviation that is used to express the production index RZM on its relative scale, resulting in a reduction of variance of GEBVs for RZM. The procedures to calculate all other index breeding values are retained from MSM.

Modelling inbreeding depression in the SSM

Pedigree-based inbreeding was considered in the previous MSM evaluations only with its effect on the pedigree-based relationship matrix for German Holstein evaluations. And the effects of inbreeding depression were ignored. With SSM, we now include a fixed effect in form of a linear regression on the pedigree-based inbreeding coefficient for all traits.

Using sire genotypes for calf fitness

An unexpectedly high genetic trend was observed in the initial SSM developments for the early-measured calf fitness CFIT (young stock survival). Causes for the overestimated SSM GEBVs were traced back to the delayed genotyping of female calves and the therefore extremely limited genotyping of dead or sick calves for the early survival traits (Alkhoder et al. 2025). As a validated solution to the inflated genomic prediction, we use only genotypes of the sires of the female calves. This contrasts with all other trait groups, where we use genotype data of all animals born 2005 or later.

Weekly evaluations with the SSM

In addition to the full SSM evaluations, a weekly genomic evaluation system was developed (Alkhoder et al. 2004). From the SSM evaluation that includes the current MACE data, SNP effect estimates and residual polygenic effect estimates for all the genotyped animals are obtained and then applied to the newly genotyped animals weekly. Direct genomic values of the new animals are computed using the SNP effect estimates and allele frequencies of SNP markers. Residual polygenic effects and GEBVs of the newly genotyped animals are indirectly predicted based on their pedigree relationship to the genotyped animals that are included in the main evaluation.

Parallel SSM genomic evaluations in the testing phase

The introduction of SSM represented a significant improvement over the previous MSM genomic evaluation and caused greater changes to GEBVs of many animals, especially the youngest genotyped animals. This imposed a challenge to breeding organizations and farmers for adopting to SSM. To help the users of genomic evaluations adapt to the new SSM model smoothly, we conducted parallel SSM evaluations in the testing phase. Starting with the August 2024 main evaluation, SSM test evaluation results were provided to the breeding organizations for the purpose of receiving valuable feedback from the breeders' perspective to further optimize all SSM systems and models. After the development and internal testing of the SSM weekly evaluation system (Alkhoder et al. 2024), we also provided the breeding organizations with test results from weekly evaluations starting in October 2024. In multiple workshops with various delegates of the breeding organizations, we gathered the feedback and used it to 1) answer yet open questions from the breeders, 2) to further improve our communication strategies regarding the introduction of SSM, and 3) to further improve our SSM models and procedures.

Results and Discussion

SSM evaluations for all the 10 trait groups are conducted using the software MiX99 (Strandén and Lidauer,1999). Approximate genomic reliabilities for GEBVs from the SSM are computed using either APAX99 from the MiX99 software suite or using own Fortran programs (Liu et al. 2023), following Interbull's Guidelines for approximating genomic reliabilities for SSM (Liu et al. 2024). Similarly, effective daughter contributions (EDC) of bulls and effective record contributions (ERC) of cows or calves are calculated using either APAX99 or own programs. The sub-indices of individual trait groups as well as the total merit indices RZG and RZ€ are computed with a central, configurable software, developed in Python. All workflows are managed with Snakemake as a workflow management software (https://snakemake.readthedocs.io/en/stable/project_info/citations.html).

Differences in technical steps between the SSM and MSM models are schematically described in Figure 1. The SSM model simplifies the genomic evaluation process with a joint analysis of genotyped and non-genotyped animals having or having no phenotypic data in one single step. In contrast, genotype data was used at a later stage of the evaluation process for MSM than phenotype data.

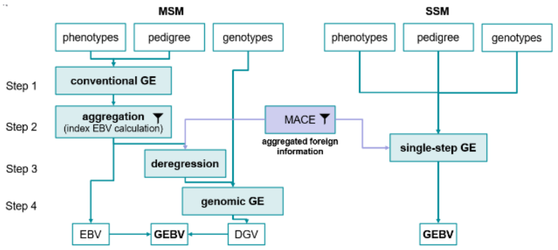

Figure 1. Comparison of technical steps between the single-step and multi-step genomic models

Figure 2 shows the numbers of validation bulls used in both validation studies: 2408 vs 2008 and 2304 vs 2104. The selection of validation bulls was conducted, following the Interbull GEBV test rules (Mäntysaari et al. 2010) for the validation study 2408 vs 2008. However, an additional selection criterion was imposed on the validation bulls for the earlier validation 2304 vs 2104: the validation bull had to be owned by a German AI organization. Unlike the 4-year validation 2408 vs 2008, the 2-year validation 2304 vs 2103 did not include data from TMACE. Instead, the actual MACE results from 2104 were used, and thus the results of the validation may likely also be impacted by model changes between 2104 and 2304 in Germany or in the other countries participating in MACE. Overall, the numbers of validation bulls are reasonably high to deduce reliable and accurate validation results.

Figure 3 shows, based on the 2-year validation 2304 vs 2104, GEBV correlations of the validation bulls between the early evaluation 2104 as young candidates without daughters and the later evaluation 2304 when these bulls were evaluated as daughter-proven bulls. The SSM model has evidently higher GEBV correlation, i.e., prediction accuracy, than the MSM model for all traits, RZG and RZ€. Similar levels of prediction accuracy are also observed for the validation study 2408 vs 2008.

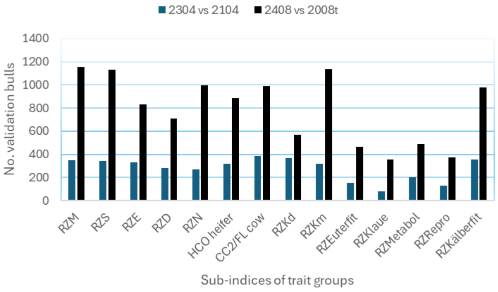

Figure 2. Number of validation bulls for all trait groups in the two validation studies

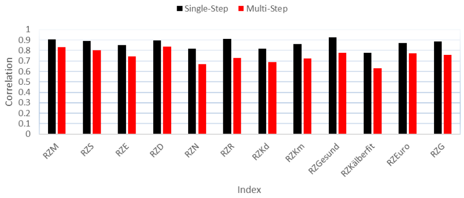

Figure 3. GEBV correlations of the validation bulls between two evaluations 2304 and 2104 (2-year validation) for the indices

For a total of 258 German AI bulls born in 2021, we compared their genomic reliabilities between SSM and MSM in the evaluation as of April 2023. Figure 4 shows the increase of genomic reliabilities from MSM to SSM for all the trait group indices, RZG and RZ€. The functional traits, like direct health traits (RZGesund) and calf fitness (RZKälberfit), gain more in reliabilities than the production trait index RZM. The traits which already had high reliabilities under the MSM tend to have a smaller increase in reliabilities when upgraded to the SSM model, such as somatic cell scores RZS.

Prediction bias, measured as the regression slope from the two validation studies, is given in Table 2. As mentioned above, Interbull's official GEBV test software was used for the validation 2408 vs 2008, with deregressed GEBVs as the dependent target variable. However, for the 2-year validation study 2304 vs 2104, our own software for a weighted linear regression (Legarra and Reverter 2018) was applied to the two SSM and MSM models.

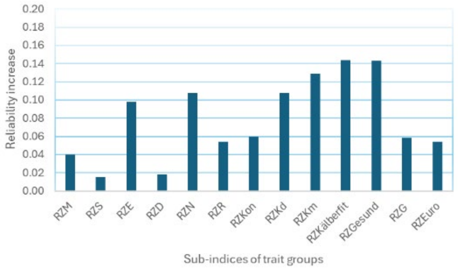

Figure 4. Increase of the genomic reliability from the multi-step to single-step model for German AI bulls in the 2-year validation

Across all the trait groups and both validation studies, we can see that both SSM and MSM give nearly unbiased prediction for almost all the trait groups, but the SSM has somewhat less over- or under-estimation, i.e., b_1 is closer to 1, than the MSM model. Using the deregressed GEBVs as dependent variable of the GEBV test results in b_1 values deviating more from 1 than using direct GEBVs as dependent variable, verified for the new calf fitness SSM model (Alkhoder et al. 2025). Over all traits of the 10 trait groups evaluated for German Holstein, we can see that neither overestimation nor underestimation seem to be an issue for the genomic models in German Holsteins. Relatively poor b_1 estimates were obtained for the calving traits, which may possibly be explained by the fact that very few stillborn/dead calves are usually genotyped. Additionally, male calves that have more problems with calving ease or stillbirth than female calves are rarely genotyped, in contrast to female calves that are all genotyped under the German whole-herd genotyping scheme if the herd participates in genotyping. Because of the relatively short history of routine recording of direct health traits and some new conformation traits in German Holsteins, the removal of phenotypic data from the last four years for the validation 2408 vs 2008t reduces the representativeness of the validation results for these traits, i.e., caution needs to be taken when interpreting the b_1 estimates for the direct health traits in the validation study 2408 vs 2008. No genomic validation was conducted for feed efficiency due to the small number of genotyped cows with dry matter intake records and a lack of cow sires that qualified for being selected as validation bulls.

Figure 5 shows GEBV correlations for the RZG between SSM and MSM for 8,661 genotyped German Holstein Black-and-White AI bulls born between 2005 and 2023 using the evaluation results as of December 2024. The GEBV correlation for AI bulls with daughters is about 0.95 and drops to 0.85 for the youngest purely genomic AI bulls born in 2023, although GEBV correlations for the youngest AI bulls are about 0.95 for individual traits like milk production and conformation traits (data not shown here). The lower RZG correlation is caused, besides the model changes, also by the changes introduced in the milk production index RZM and the longevity index RZN.

Table 2: Regression slope estimates of the GEBV test of selected traits for the genomic models in the two validation studies

Trait	2408 vs 2008	2304 vs 2104	
	SSM	SSM	MSM
Milk yield	1.02	1.02	1.01
Fat yield	1.06	1.03	1.15
Protein yield	1.05	1.03	1.10
Somatic cell scores	1.11	1.05	1.07
Functional longevity	0.97	1.00	0.95
Heifer fertility HCO	1.17	1.05	1.11
Cow fertility CC2	0.98	0.99	0.88
Stillbirth direct	0.84	0.88	0.97
Calving ease maternal	0.78	0.89	0.96
Milking speed	1.01	1.06	1.13
Milking temperament	0.97	0.96	0.97
Stature	0.99	1.03	1.07
Udder support	0.98	1.01	1.23
Body condition score	1.07	1.10	1.09
Locomotion	1.01	1.00	1.14
Digital dermatitis	0.88	0.92	0.92
Clinical mastitis	0.58	0.86	0.67
Calf fitness	0.95	1.04	0.92

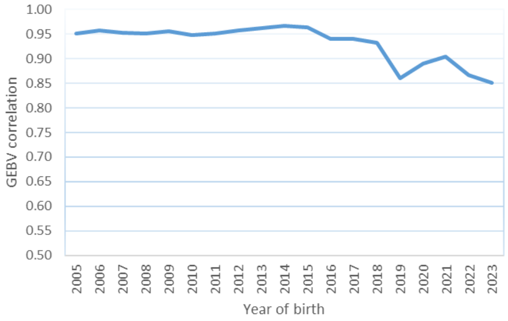

Figure 5. GEBV correlations of the total merit index RZG between SSM and MSM for German Holstein AI bulls

Like Figure 5 for AI bulls, Figure 6 shows GEBV correlations of RZG for 1,478,613 genotyped Black-and-White Holstein female animals. For all the female animals born from 2016 on and genotyped under the German whole-herd genotyping scheme, SSM and MSM have a GEBV correlation of about 0.95 for the total merit index RZG, despite the above-mentioned additional changes impacting

RZG. The youngest female animals have much higher RZG GEBV correlation than the youngest AI bulls, 0.95 vs 0.85, which may be attributed to the different selection intensities between the two groups of genotyped animals.

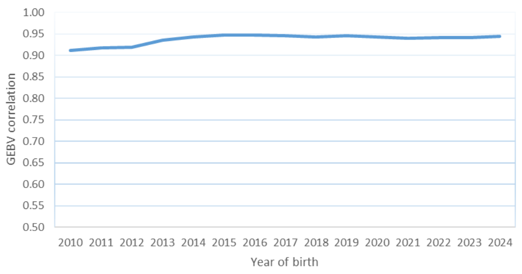

Figure 6. GEBV correlations of the total merit index RZG between SSM and MSM for genotyped Holstein female animals

With respect to expectation management during the preparations prior to the introduction of SSM, we emphasized clearly from the beginning that the change to SSM was a major upgrade of the genomic model and would thus have a profound impact on the breeding organizations' selection programs. In addition to the early research projects on the theory of the SSM model, we invested huge efforts and resources to develop the new evaluation systems as well as to help the breeding organizations and farmers adjust their programs for the new SSM model. Besides the numerous parallel full and weekly SSM test evaluations, we provided our new results at several meetings with our customers and improved our SSM systems based on their feedback. These iterations of providing additional analyses and explanations to our breeding organizations and including customer feedback in the improvement of our models led to a strong support of the introduction of SSM by the German breeding organizations.

As the MSM genomic model uses deregressed proofs as the dependent variable / pseudo-records calculated from the conventional evaluation which ignores the rapid genetic progress by genomic selection, the differences between the two genomic models SSM and MSM are likely to increase with time. A delayed update from MSM to SSM would make it increasingly difficult for farmers and breeders to adapt to the one-time model change.

Conclusions

SSM represents a major improvement of our genomic evaluation compared to the previous MSM. All genotypic, phenotypic and pedigree data are jointly evaluated by the single-step model, resulting in a higher accuracy of the genomic prediction. With two genomic validation studies, we have shown higher prediction accuracy, i.e., correlation between the early candidate GEBVs and later daughter-proven GEBVs for AI bulls and higher genomic reliabilities for all traits from SSM compared to MSM. GEBVs from SSM have been proven to be more stable across evaluation runs and to have substantially increased reliabilities compared to GEBVs from the previous MSM for all the trait groups. Because of these major improvements, the introduction of SSM received strong support from the breeders and their breeding organizations, despite the relatively large one-time changes. Intensive collaboration with the German breeding organizations prior to and after the introduction of SSM led to strong commitment from the breeders' community and helped the introduction of SSM in German Holsteins become a great success.

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