# Application of the Interbull genomic reliability method for singlestep evaluations of test-day and conformation traits in German Holstein

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# Abstract

National single-step genomic evaluation required accurate genomic reliabilities, particularly for young, genotyped animals. The Interbull genomic reliability method was tested for single-step evaluation of four test-day traits as well as for 25 conformation traits in German Holstein. Genotypic, phenotypic and pedigree data were taken from the official genomic evaluation in April 2023. More than 1.3 million genotyped animals were considered jointly with non-genotyped animals, and the genomic reference population exceeded half a million animals for the test-day traits. Selecting fewer SNP markers in reliability calculation for direct genomic values (DGV) was proven to be an efficient way of decreasing computing time or memory usage while retaining a reasonable accuracy when at least 15,000 equidistant SNP markers were chosen. Due to the extremely large reference population, the level of DGV reliabilities was very high, also for the young, genotyped candidates. Adjusting the theoretical DGV reliabilities based on the Interbull reliability method seemed to be unavoidable, especially for the large reference population. Variation in the DGV reliabilities was shown to be small among animals born in the same year, especially among the young, genotyped animals without own phenotypic records. Therefore, a constant genomic effective daughter contribution could result in reasonably accurate genomic reliability values and at the same time may provide a computationally much less demanding way for routine genomic reliability calculation with several million genotyped animals included. The single-step genomic reliability values were compared to conventional reliabilities as well as genomic reliabilities from the current multi-step genomic model for diverse groups of animals of German Holstein. The single-step genomic reliabilities of the test-day and conformation traits seemed to be consistent with the variance of genomic breeding values.

Key words: Genomic reliability, genomic breeding values, single-step model, test-day traits

# Introduction

Single-step evaluation required accurate reliability values for estimated genomic breeding values (GEBV). The Interbull genomic reliability method (Liu et al., 2017) was developed for the current multi-step genomic model (MSM) as well as the singlestep genomic model (SSM). The main goal of the Interbull genomic reliability method was to make national genomic reliabilities comparable across countries by applying the same reliability method in all the countries. Ideally, genomic reliability values should be consistent with the variances of GEBV. The main features of the

Interbull genomic reliability method were 1) treating genotype data as an additional source of information contributing to animal's total reliability, 2) calculating exact, theoretical reliabilities of direct genomic values (DGV) for all genotyped animals under a SNP BLUP model, and 3) adjusting genomic reliabilities based on GEBV variance changes of validation bulls (VanRaden and O'Connell, 2018).

The step of calculating exact reliabilities of DGV in the Interbull genomic reliability method may take considerable computing time for countries with extremely large reference populations, even with the highly efficient software snp\_blup\_rel (Ben Zaabza et al. 2020).

Reducing the number of SNP markers can decrease the computing time for the calculation of DGV reliabilities. The impact of skipping this step of DGV reliability calculation in routine evaluation needed to be investigated.

The aims of this study were 1) to apply the Interbull genomic reliability method to genotypic, phenotypic and pedigree data of the German Holstein single-step evaluations for test-day and conformation traits, 2) to compare the accuracy of DGV reliabilities between scenarios using all and fewer SNP markers, and 3) to investigate the level and variation of the exact DGV reliabilities for young, genotyped candidates.

# **Materials and Methods**

# Data for single-step evaluation

Phenotypic, genotypic and pedigree data were obtained from the April 2023 routine evaluation of German dairy cattle breeds. Two groups of traits were chosen for this study: 25 conformation traits (Alkhoder et al. 2021) and four test-day traits (Alkhoder et al., 2023) including milk yield (MKG), fat yield (FKG), protein yield (PKG), and somatic cell scores (SCS). The conformation trait stature (STA) represented a linear type trait with a complete classification history, whereas the recording of locomotion (LOC) started several years later than STA. The national trait udder balance (EUB) was not included in Interbull MACE evaluation, and a new definition of angularity (ANG) was recently introduced in Germany in April 2023 with a much smaller phenotypic data set. Table 1 describes the data sets for the single-step evaluations of the test-day traits as well as the conformation traits for the German dairy breeds. The size of the bull and cow reference population for German Holstein breed is 524,187 for each of the four test-day traits or 386,062 for the conformation traits.

To validate the calculated genomic reliabilities of the test-day traits, the same truncated phenotypic data for the GEBV validation were used as in Alkhoder et al. (2023). Test-day records in last 4 years from the evaluation April 2021 were truncated for simulating an early prediction back in April 2017. In contrast to the data truncation of 4 years for the test-day traits, conformation records in last two years were removed from the full evaluation of April 2023 for simulating an early evaluation in April 2021.

**Table 1.** Description of the data sets for the single-stepevaluationsoffourtest-dayand25conformation traits in April 2023

Frequency	Test-day	Conformation	
	traits	25 traits	
Genotyped	1,318,780 Holstein animals		
animals	(1,138,039 females and		
	180,741 males)		
Phenotyped	13,528,444	3,144,366	
animals			
Phenotypic	263,673,267	3,144,366	
records	test-day	type records	
	yields		
Genotyped or	14,402,662	4,131,336	
phenotyped			
animals			
Animals in	21,850,276	10,048,593	
pedigree			
Reference	524,187	386,062	
animals			

Table 2 shows the data sets used for validating genomic reliabilities, including both the full and truncated evaluations. For each testday trait, the number of reference animals decreased more than a half in the truncated evaluation, due to the rather short history of female animal genotyping in Germany. To make the genomic validation reflect more realistically a future prediction, only two years of phenotypic data were therefore deleted for the conformation traits. The number of reference animals for the conformation traits was reduced from 386,062 in the full evaluation in April 2023 to 263,252 in the truncated evaluation in April 2021. The genomic validation for the test-day traits was conducted using data from an older evaluation than the conformation traits.

and

FrequencyTest-dayType4 traits25 traitsFull evaluationApril 2021April 2023Truncated runApril 2017April 2021Genotyped949,6361,318,780Holstein animals12,571,7103,144,366animals (full &11,032,3952,862,770truncated runs)20,461,40010,048,593pedigree86,062156,970263,252evaluations)156,970263,252	conformation traits		
4 traits25 traitsFull evaluationApril 2021April 2023Truncated runApril 2017April 2021Genotyped949,6361,318,780Holstein animals12,571,7103,144,366animals (full &11,032,3952,862,770truncated runs)20,461,40010,048,593pedigree156,970263,252evaluations)10,048,252	Frequency	Test-day	Туре
Full evaluationApril 2021April 2023Truncated runApril 2017April 2021Genotyped949,6361,318,780Holstein animals12,571,7103,144,366animals (full & truncated runs)11,032,3952,862,770Animals in pedigree20,461,40010,048,593Reference animals353,347386,062(full & truncated156,970263,252evaluations)10,048,593		4 traits	25 traits
Truncated run         April 2017         April 2021           Genotyped         949,636         1,318,780           Holstein animals         12,571,710         3,144,366           animals (full & 11,032,395         2,862,770           truncated runs)         20,461,400         10,048,593           pedigree         353,347         386,062           (full & truncated         156,970         263,252	Full evaluation	April 2021	April 2023
Genotyped Holstein animals         949,636         1,318,780           Phenotyped animals (full & truncated runs)         12,571,710         3,144,366           Animals (full & truncated runs)         11,032,395         2,862,770           Animals in pedigree         20,461,400         10,048,593           Reference animals         353,347         386,062           (full & truncated evaluations)         156,970         263,252	Truncated run	April 2017	April 2021
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Phenotyped         12,571,710         3,144,366           animals (full &         11,032,395         2,862,770           truncated runs)         20,461,400         10,048,593           pedigree         20,461,400         10,048,593           Reference animals         353,347         386,062           (full & truncated         156,970         263,252           evaluations)         20         20	Holstein animals		
animals (full &       11,032,395       2,862,770         truncated runs)       20,461,400       10,048,593         pedigree       20,461,400       10,048,593         Reference animals       353,347       386,062         (full & truncated       156,970       263,252         evaluations)       20,461,400       20,048,593	Phenotyped	12,571,710	3,144,366
truncated runs)Animals in pedigree20,461,40010,048,593Reference animals353,347386,062(full & truncated156,970263,252evaluations)	animals (full &	11,032,395	2,862,770
Animals in pedigree         20,461,400         10,048,593           Reference animals (full & truncated evaluations)         353,347         386,062	truncated runs)		
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(full & truncated 156,970 263,252 evaluations)	Reference animals	353,347	386,062
evaluations)	(full & truncated	156,970	263,252
	evaluations)		

Table 2. Description of the data sets for validating

genomic reliabilities for the test-day

For computing the exact, theoretical reliability values of DGV for all genotyped animals, a genomic reference population comprising genotyped cows or bulls with own phenotypic data needed to be set up. Table 3 describes the composition of genomic reference population for 5 selected traits: PKG representing the test-day traits. four conformation traits STA, LOC, ANG and EUB. In Table 3 it can be seen that the test-day milk production trait PKG has more than half a million reference animals as a result of the large-scale female animal genotyping in Germany. The 4 conformation traits have a smaller reference population than the test-day trait PKG, because not all cows in milk recording program were classified for conformation. The national trait EUB has only a little lower number of reference animals than the regular type traits STA and LOC. Due to the trait definition change that was introduced in April 2023, the conformation trait ANG has the lowest number of genotyped cows with classification record according to the new definition.

Between the data sets for April 2023 and April 2021 there was a difference in genotype editing for bulls. Due to un-intentional selective genotyping of bulls in early years of genomic selection, we decided to remove genotype records of bulls born before 2005 in the singlestep evaluations with the data set from April 2023. However, this genotype data editing was not implemented in the single-step evaluations with the data set from April 2021.

**Table 3.** Genomic reference populations of selectedtraits in April 2023 evaluation

Troit	Reference animals		
ITali	Cows	Bulls	Total
Protein yield	478,588	45,591	524,179
Stature	357,365	28,635	386,000
Locomotion	349,083	27,696	376,779
Angularity	198,170	27,748	225,918
Udder balance	305,122	27,205	332,327

# Scenarios of reducing SNP markers for faster calculation of DGV reliabilities

As a core component of the Interbull genomic reliability method (Liu et al. 2017), the calculation of DGV reliability values may be computationally demanding for extremely large reference populations like those in Table 3. Therefore, the impact of reducing SNP markers on the DGV reliabilities was investigated in a similar way by selecting equidistant SNP markers as by Sargolzaei et al. (2014) and Strandén and Mäntysaari (2015). Table 4 describes the test scenarios of selecting the SNP markers for faster DGV reliability calculation. The base scenario of using all SNP markers, RELall, has 45,613 SNP markers included in the DGV reliability calculation as in the routine genomic evaluation for German Holstein. Five additional scenarios were simulated by selecting every 2 (RELevery2), every 3 (RELevery3), every 4 (RELevery4), every 5 (RELevery5) and every 10 (RELevery10) equidistant SNP markers. When every 10 SNP markers were selected in scenario RELevery10, the number of markers was reduced to 4,562. For this specific investigation, genotypic and phenotypic data from April 2021 were used (see Table 2) and the selected trait was PKG.

	No.
Scenario	markers
All SNP markers (RELall)	45,613
Every 2 markers (RELevery2)	22,807
Every 3 markers (RELevery3)	15,205
Every 4 markers (RELevery4)	11,404
Every 5 markers (RELevery5)	9,123
Every 10 markers (RELevery10)	4,562

 Table 4.
 Scenarios of selecting equidistant SNP

 markers for faster calculation of DGV reliabilities

# **Results & Discussion**

All computations were done on a Linux server equipped with 42 cores and 512Gb RAM.

#### Impact of fewer markers on DGV reliabilities

Reducing the number of SNP markers for the DGV reliability calculation leads to significant decreases in computing time and memory usage, which can be seen clearly in Table 5.

For the base scenario of using all SNP markers, RELall, the computing time of the DGV reliability values depended mostly on the number of all genotyped animals and the number of animals in reference population. For weekly genomic evaluation by adding up to 20,000 newly genotyped animals, the DGV reliability calculation required less than 4 minutes.

**Table 5.** Computational requirements for thescenarios of the calculation of DGV reliabilities

	Total	Peak
	time	RAM
Scenario	(min.)	(Gb)
All SNP markers (RELall)	215	88
Every 2 markers (RELevery2)	96	42
Every 3 markers (RELevery3)	71	28
Every 4 markers (RELevery4)	60	21
Every 5 markers (RELevery5)	55	18
Every 10 markers	47	10
(RELevery10)		

Figure 1 shows average DGV reliabilities of all 949,636 genotyped Holstein animals in the April 2021 evaluation for trait PKG. The

number of genotyped animals (in blue bar) increased drastically in recent years, due to the routine herd genotyping of female animals in Germany. Thanks to the higher number of reference animals, 353,347 (Table 2), DGV reliabilities for the genotyped animals have a rather high average, above 0.94 for candidates younger than 1 year old.



**Figure 1.** Average DGV reliabilities of protein yield for genotyped Holstein in April 2021 evaluation

Figure 2 shows correlation of DGV reliabilities between a scenario and the base scenario for protein yield of all the genotyped animals. Across all the birth years, the within-year correlation has an average of 0.997, 0.990, 0.980, 0.968, and 0.903 for the scenarios RELevery2, RELevery3, RELevery4, RELevery5, and RELevery10, respectively.



**Figure 2.** Correlation of DGV reliabilities between different scenarios for protein yield of all genotyped Holstein animals

Figure 3 shows average difference in DGV reliabilities of protein yield for all genotyped animals between a scenario and the base scenario. With fewer SNP markers selected, DGV reliabilities tend to be over-estimated by comparing to the base scenario using all the SNP markers. The difference in DGV reliabilities seems to higher for the youngest or oldest genotyped animals than animals in between. It can be clearly seen that using fewer SNP markers leads to higher DGV reliability values.



**Figure 3.** Average differences of DGV reliabilities of protein yield of the scenarios with the base scenario using all markers for all genotyped animals

For the youngest candidates born in 2020 and later in the April 2021 evaluation, their DGV reliabilities of the base scenario were regressed on those from each of the scenarios. Figure 4 shows that selecting every 3 equidistant markers of scenario RELevery3 gives a reasonably high correlation of DGV reliabilities with the base scenario of using all SNP markers and at the same time requires only c.a. 1/3 RAM usage and computing time (Table 5).



**Figure 4.** Regression of DGV reliabilities of youngest candidates born after 2020 from the base scenario on the other scenario for trait protein yield

#### Average and variance of DGV reliabilities

For 8,123 Holstein AI bulls owned by German AI studs, Figure 5 and Figure 6 show average DGV reliabilities by birth year for the test-day traits and for the four chosen conformation

traits, respectively. Because of the extremely large reference populations (Table 3), the average of DGV reliabilities is very high for any of the 8 selected traits, particularly for the young genomic AI bulls born in 2020 to 2022. Trait ANG has the lowest DGV reliabilities, due to its smallest reference population. Another reason for the extremely high level of DGV reliabilities is that no residual polygenic effect be assumed in the SNP BLUP model for the DGV reliability calculation.



**Figure 5.** Average DGV reliabilities of German Holstein AI bulls for test-day traits



**Figure 6.** Average DGV reliabilities of German Holstein AI bulls for four conformation traits

Standard deviation of DGV reliabilities of the AI bulls is shown in Figure 7 for the test-day traits and in Figure 8 for the four conformation traits, respectively. It can be seen in both figures that traits with larger or more informative reference population have lower variation in DGV reliabilities. Test-day trait MKG, having the highest heritability value and thus the highest reliability among the four test-day traits and all the 8 traits, has shown to be least variable in DGV reliabilities. In contrast, conformation trait ANG has the largest variance in DGV reliabilities due to its smallest reference population. Across all the traits, the DGV reliabilities have rather small variation, especially for young genomic AI bulls born in 2020 and later.



**Figure 7.** Standard deviations of DGV reliabilities of the German Holstein AI bulls for test-day traits



**Figure 8.** Standard deviations of DGV reliabilities of the German Holstein AI bulls for the four conformation traits



**Figure 9.** Genomic and conventional reliabilities of the German Holstein AI bulls for trait protein yield

#### Genomic and conventional reliabilities

For trait PKG, Figure 9 shows genomic and conventional reliability values of Holstein AI bulls owned by German AI studs. For bulls with complete daughter information born between 1998 and 2015, genomic and conventional reliabilities are essentially equal. However, for bulls born in 2016 and later with incomplete or no daughter information yet, genomic reliabilities are a little or significantly higher than the conventional reliabilities, respectively.

Figure 10 shows genomic and conventional reliabilities of trait ANG. Due to much less national cow data for this newly changed trait, bulls with or without daughters have always higher genomic reliabilities than conventional reliabilities.

Like trait PKG, genomic and conventional reliabilities are nearly equal for bulls with daughters and higher for young AI bulls without daughters for all the other test-day or the conformation traits, except ANG.



**Figure 10.** Genomic and conventional reliabilities of the German Holstein AI bulls for trait angularity

# Single-step and multi-step genomic reliability values

For trait PKG, both single-step and multi-step genomic reliabilities are shown in Figure 11 for the German Holstein AI bulls. As a result of the removal of genotype data of bulls born before 2005, single-step reliabilities are a little bit lower than the multi-step ones for the daughterproven bulls born between 1998 and 2004. Overall, the two sets of genomic reliabilities are nearly equal for all the bulls with daughters. The single-step genomic reliabilities are evidently higher than the multi-step ones for the young AI bulls born in 2020 and later, because the SSM uses more phenotypic and genotypic information than the MSM.



Figure 11. Single-step and multi-step genomic reliabilities of the German Holstein AI bulls for protein yield

For trait ANG with its definition changed recently, only two years of domestic cows had phenotypic records, besides the MACE data of foreign bulls. The SSM reliabilities are significantly higher than reliabilities of the MSM, as shown in Figure 12. The much lower SSM reliabilities for the AI bulls born before 2005 can be explained by the truncation of genotype data of the bulls born in 2004 and earlier. For new traits like ANG with limited phenotypic information, SSM is shown to have clearly higher genomic reliabilities than the current MSM.



Figure 12. Single-step and multi-step genomic reliabilities of the German Holstein AI bulls for angularity

### Conclusions

Interbull genomic reliability method was tested for the single-step genomic evaluation using phenotypic, genotypic and pedigree data of German dairy cattle from the April 2023 official evaluation. Calculation of exact, theoretical DGV reliability values for all genotyped animals was shown to be the most timeconsuming step of the Interbull genomic reliability method. Five scenarios of reducing the number of SNP markers were conducted to investigate the computational efficiency and DGV reliability accuracy. For the extremely large reference population of German Holstein, at least 15,000 equidistant SNP markers must be chosen to achieve a reasonably high accuracy of the DGV reliabilities while significantly reducing the computing time and memory usage. Based on the genotypic and phenotypic data of four test-day traits and 25 linear conformation traits, average and variances of the DGV reliabilities for various groups of animals were calculated. The average of the DGV reliabilities for young, genotyped animals was found to be rather high, possibly caused by the size of the extremely large reference population. The very high level of DGV reliabilities suggested that an adjustment of the theoretical DGV reliabilities be necessary to guarantee the proper level of genomic reliabilities for young candidates. Meanwhile it was shown that variation of the DGV reliabilities within birth year was small, which indicated that calculating individual DGV reliabilities be less crucial for a large reference population like German Holstein. By comparing to conventional reliabilities and the current MSM genomic reliabilities, the final genomic reliabilities of the single-step model were shown to be higher for young, genotyped candidates without own phenotypic data. Based on the application of the Interbull genomic reliability method to the German dairy cattle data, guidelines for a routine implementation in national single-step evaluation will be developed.

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