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Genomic predictions based on a joint reference population for the Nordic Red cattle breeds

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ABSTRACT

The main aim of this study was to compare accuracies of imputation and genomic predictions based on single and joint reference populations for Norwegian Red (NRF) and a composite breed (DFS) consisting of Danish Red, Finnish Ayrshire, and Swedish Red. The single nucleotide polymorphism (SNP) data for NRF consisted of 2 data sets: one including 25,000 markers (NRF25K) and the other including 50,000 markers (NRF50K). The NRF25K data set had 2,572 bulls, and the NRF50K data set had 1,128 bulls. Four hundred forty-two bulls were genotyped in both data sets (double-genotyped bulls). The DFS data set (DSF50K) included 50,000 markers of 13,472 individuals, of which around 4,700 were progeny-tested bulls. The NRF25K data set was imputed to 50,000 density using the software Beagle. The average error rate for the imputation of NRF25K decreased slightly from 0.023 to 0.021, and the correlation between observed and imputed genotypes changed from 0.935 to 0.936 when comparing the NRF50K reference and the NRF50K–DFS50K joint reference imputations. A genomic BLUP (GBLUP) model and a Bayesian 4-component mixture model were used to predict genomic breeding values for the NRF and DFS bulls based on the single and joint NRF and DFS reference populations. In the multiple population predictions, accuracies of genomic breeding values increased for the 3 production traits (milk, fat, and protein yields) for both NRF and DFS. Accuracies increased by 6 and 1.3 percentage points, on average, for the NRF and DFS bulls, respectively, using the GBLUP model, and by 9.3 and 1.3 percentage points, on average, using the Bayesian 4-component mixture model. However, accuracies for health or reproduction traits did not increase from the multiple population predictions. Among the 3 DFS populations, Swedish Red gained most in accuracies from the multiple population predictions, presumably because Swedish Red has a closer genetic relationship with NRF than Danish Red and Finnish Ayrshire. The Bayesian 4-component mixture model performed better than the GBLUP model for most production traits for both NRF and DFS, whereas no advantage was found for health or reproduction traits. In general, combining NRF and DFS reference populations was useful in genomic predictions for both the NRF and DFS bulls.

Key words: imputation, genomic BLUP, Bayesian 4-component mixture model, multiple population genomic prediction

INTRODUCTION

Simulation studies (e.g., de Roos et al., 2009) as well as analyses of real data (Brøndum et al., 2011; Lund et al., 2011; VanRaden et al., 2012) have shown that genomic predictions can work across different populations. By combining different populations of the same breed or related breeds in the reference population, more information is available for the estimation of marker effects. Hence, more accurate predicted breeding values will be obtained in genomic predictions. Accuracies increased when 3 related dairy cattle populations—Danish Red, Swedish Red, and Finnish Ayrshire—were combined into one reference population (Brøndum et al., 2011). Reliabilities increased by 10 percentage points, on average, when 4 European Holstein populations were combined in the reference population (Lund et al., 2011). Increases in reliabilities from 6 to 45 percentage points were achieved by combining 6 Brown Swiss populations (Jorjani et al., 2011). However, most of the predictive accuracy, at least with density of 50,000 SNP, most likely comes from predicting the effect of large segments of chromosome or relationships, rather than individual QTL effects (Daetwyler et al., 2012; Wientjes et al., 2013).

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Norwegian Red (**NRF**), with approximately 230,000 cows, has good performance in health, fertility, and milk production. It is the main dairy breed in Norway (95% of dairy cows). A previous study (Luan et al., 2009) reported that correlations of genomic EBV (**GEBV**) and daughter yield deviations for NRF varied widely between 0.12 and 0.62 for different traits and were low for health and reproduction traits. Danish Red (38,000 cows), Finnish Ayrshire (143,000 cows), and Swedish Red (116,000 cows) are important red dairy cattle populations in these Nordic countries. These 3 red cattle populations were merged into one composite breed (**DFS**), also named VikingRed, which currently has a joint genetic evaluation. The reliabilities of genomic predictions of DFS averaged 0.28 for 17 traits (Brøndum et al., 2011). The NRF breed is related to Swedish Red and Finnish Ayrshire (Olsen et al., 2011). Danish Red, Finnish Ayrshire, and NRF were also used in the Swedish Red breeding program (Bett et al., 2010). Because sires have been exchanged and used between these populations, some genetic links exist between NRF and DFS.

Combining these 2 related breeds provides an interesting approach to improve accuracies in their genomic predictions. Bayesian variable selection models have been shown to give a better persistence of genomic predictions (Gao et al., 2013). Because this joint data set includes many distant relationships across the breeds, we expect that these models would result in higher accuracies in genomic predictions than traditional genomic BLUP (**GBLUP**) models. In this study, our first objective was to investigate accuracies in imputation from 25,000 (**25K**) to 50,000 (**50K**) SNP for NRF bulls, using only the NRF data set or the NRF and DFS data sets together as the reference. The second objective was to investigate accuracies in genomic predictions for NRF and DFS using the single or joint NRF DFS reference population, and to compare the GBLUP model and a Bayesian 4-component mixture model in genomic predictions of different traits.

MATERIALS AND METHODS

Genotypic and Phenotypic Data

The SNP data for NRF consisted of 2 data sets of progeny-tested bulls: a data set with 2,572 bulls genotyped with 25,000-SNP chips (**NRF25K**; Affymetrix, Santa Clara, CA; Affymetrix, 2007) and a data set with 1,128 bulls genotyped with the 54,001 SNP of the BovineSNP50 chip (**NRF50K**; Illumina Inc., San Diego, CA; Matukumalli et al., 2009). A total of 442 NRF bulls were genotyped using both the 25K and 50K chips (double-genotyped bulls). The DFS data (**DFS50K**) included genotypes of BovineSNP50 chips on Danish Red, Finnish Ayrshire, and Swedish Red. A total of 13,427 genotyped animals were included in the DFS50K data set, of which around 4,700 were progeny-tested bulls, 3,440 were cows, and the rest were young bulls without progeny test results. The genetic correlations and number of common sires between DFS and NRF from Interbull international genetic evaluations are shown in Table 1 (http://interbull2.slu.se). According to the pedigree, 291 DFS bulls (242 Swedish Red, 44 Finnish Ayrshire, and 5 Danish Red) have been used in the NRF population, and 58 NRF bulls have been used in the DFS population. In our data, 18% (864/4,741) of the progeny-tested bulls in the DFS50K data set have common sires with the NRF bulls, and 14% (366/2,572) of the NRF bulls have common sires with the DFS bulls. The 864 DFS bulls that had common sires with NRF bulls represent 6% (58/911), 17% (394/2,344), and 28% (412/1,486) of the Danish Red, Finnish Ayrshire, and Swedish Red bulls, respectively.

The 3 data sets (NRF25K, NRF50K, and DFS50K) were edited by removing SNP with minor allele frequencies <0.001 and call rate (per locus) <0.1. After SNP editing, SNP common to both the NRF50K and DFS50K data sets were kept for further analyses. To impute the NRF from 25K to 50K, SNP in NRF25K that were not present in the 50K data set were excluded

Table 1. Genetic correlations between a composite breed (Danish Red, Finnish Ayrshire, and Swedish Red; DFS) and Norwegian Red (NRF), and number of bulls in common, for some production and fertility traits according to Interbull international genetic evaluation December 2013

Trait	Genetic correlation	No. of common bulls
Milk yield	0.91	66
Fat yield	0.90	67
Protein yield	0.89	67
56-d nonreturn rate of heifers	0.79	66
Interval from calving to first insemination	0.88	58
56-d nonreturn rate of cows	0.73	61
Cows' ability to conceive[1]	0.71	53
Calving interval	0.86	53

[1]Calving interval (NRF) and interval from first to last insemination (DFS).

from the NRF25K data set. In this procedure, around 15,000 SNP in the 25K chip were removed. Finally, 45,475 SNP were selected from the 50K chip. The number of SNP used from the 25K chip was 7,611. Among these 7,611 SNP, half of them had switched genotype by replacement of A ↔ T and C ↔ G, because of the inverse genotyping calling strategies of Illumina and Affymetrix companies. To validate the imputation accuracy, 500 SNP were randomly selected from the 7,611 SNP of the NRF25K data set as a validation set. The NRF25K data set, with 2,130 (2,572 − 442) bulls, was imputed to 50K by (1) using only the NRF50K data set as the reference and (2) using the NRF50K and DFS50K data sets together as the reference. The Beagle software program (version 3.3.2; Browning and Browning, 2009) was used for imputation. To ensure that all the available information was used, all the genotyped individuals, including DFS cows, were used in the joint imputation.

Deregressed proofs (**DRP**) were used as the response variables in genomic predictions. For genomic predictions of NRF bulls, DRP of all NRF and DFS bulls in the Norwegian scale were calculated from Interbull EBV in the Norwegian scale, and vice versa for genomic prediction of DFS bulls. The Mix99 program (Lidauer and Strandén, 1999; Strandén and Mäntysaari, 2010) was used to calculate DRP. Traits analyzed for NRF were milk yield, fat yield, protein yield, 56-d nonreturn rate of heifers (**NR56H**), interval from calving to first service (**CFI**), and 56-d nonreturn rate of cows (**NR56C**); traits analyzed for DFS were milk yield, fat yield, protein yield, and fertility index and mastitis index as composite traits. The fertility index combined CFI, interval from first to last insemination, and number of inseminations, whereas the mastitis index was calculated from clinical mastitis with SCC and udder conformation.

Only progeny-tested bulls with DRP were included in genomic predictions. Bulls were split into reference and validation populations by birth date: January 1, 2000, for the NRF data sets and October 1, 2001, for the DFS50K data set. To ensure enough bulls in the reference population, the cut-off date was set later for the DFS50K data set. All NRF bulls in the NRF25K and NRF50K data sets were used in the NRF single population predictions, and all DFS bulls in the DFS50K data set were used in the DFS single population predictions. All NRF and DFS bulls with DRP were used in the multiple population predictions. Table 2 shows the number of bulls in the reference and validation populations used for genomic predictions for each trait and breed.

GBLUP Model

The GBLUP model (VanRaden, 2008) used in this study was

$$\mathbf{y} = \mathbf{1}\mu + \mathbf{Zg} + \mathbf{e},$$

where $\mathbf{y}$ was the vector of DRP, $\mathbf{1}$ was a vector of ones, μ was the population mean, $\mathbf{g}$ was the vector of genomic breeding values (GEBV), $\mathbf{e}$ was the vector of residuals, and $\mathbf{Z}$ was a design matrix allocating $\mathbf{g}$ to $\mathbf{y}$.

Table 2. Number of bulls in the reference and validation populations for each trait in the Norwegian Red (NRF) and a composite breed (DFS; Danish Red, Finnish Ayrshire, and Swedish Red), and multiple population genomic predictions

		Single population predictions[2]		Multiple population predictions	
Breed	Trait[1]	Reference	Validation	Reference	Validation
NRF	Milk yield	2,076	508	2,076 NRF + 3,357[3] DFS	508
	Fat yield	2,076	508	2,076 NRF + 3,357 DFS	508
	Protein yield	2,076	508	2,076 NRF + 3,357 DFS	508
	NR56H	2,076	508	2,076 NRF + 3,065 DFS	508
	CFI	2,076	508	2,076 NRF + 3,325 DFS	508
	NR56C	2,076	508	2,076 NRF + 3,324 DFS	508
DFS	Milk yield	3,367	1,349	3,364 DFS + 2,353 NRF	1,349
	Fat yield	3,367	1,349	3,364 DFS + 2,353 NRF	1,349
	Protein yield	3,367	1,349	3,364 DFS + 2,353 NRF	1,349
	Fertility	3,376	1,312	3,322 DFS + 2,353 NRF	1,312
	Mastitis	3,367	1,341	3,363 DFS + 2,353 NRF	1,341

[1]NR56H = 56-d nonreturn rate of heifers, CFI = interval from calving to first service, and NR56C = 56-d nonreturn rate of cows. Fertility was a composite index that combined CFI, interval from first to last insemination, and number of inseminations; mastitis index was calculated from clinical mastitis with SCC and udder conformation.

[2]In single population predictions, the reference and validation populations contained individuals only from the NRF or the DFS data set. In multiple population predictions, the reference populations included individuals from both the NRF and DFS data sets.

[3]Number of bulls in the reference populations of single and multiple population predictions were different because some DFS bulls lacked EBV on the NRF scale.

It was assumed that $\mathbf{g} \sim N\left(0, \mathbf{G}\sigma_g^2\right)$ and $\mathbf{e} \sim N\left(0, \mathbf{D}\sigma_e^2\right)$, where $\mathbf{G}$ was the genomic relationship matrix (G-matrix), σ_g^2 was the additive genetic variance, $\mathbf{D}$ was a diagonal matrix with weights on the residual variance, and σ_e^2 was the residual variance. Diagonal elements of $\mathbf{D}$ were calculated as $d_{ii} = \left(1 - r_i^2\right)\Big/ r_i^2$, where r_i was the accuracy of DRP for animal i (Su et al., 2012). The G-matrix was constructed by method 1 of VanRaden (2008), where the genomic relationship (g_{ij}) of individual i and j was calculated as

$$g_{ij} = \sum_{k=1}^{n} m_{i,k} m_{j,k} \,/\, 2\sum 2p_k(1-p_k),$$

where $m_{i,k}$ and $m_{j,k}$ were the marker genotypes for individuals i and j at locus k with values $0 - 2p_k$, $1 - 2p_k$, and $2 - 2p_k$ for genotypes A_1A_1, A_1A_2, and A_2A_2, respectively; p_k was the allele frequency of A_2 at locus k, and n was the total number of markers. Allele frequencies (p) were calculated individually for the NRF and DFS50K data sets in the single population predictions but were recalculated after merging the NRF and DFS50K data sets for the multiple population predictions. When using the DFS50K data set, only progeny-tested bulls were included in the calculation of p. The G-matrices and their inverses were calculated using the Fortran program Gmatrix (Su and Madsen, 2010). Genomic predictions using the GBLUP model and estimation of variance components were conducted by using the DMU package (Madsen and Jensen, 2010).

Bayesian Four-Component Mixture Model

We assumed that most SNP individually only explained very little variance and very few SNP effects explained large variance. Therefore, GEBV were also predicted using a Bayesian 4-component mixture model (Gao et al., 2013). The model was

$$\mathbf{y} = \mathbf{1}\mu + \mathbf{Mq} + \mathbf{e},$$

where $\mathbf{y}$ was the vector of DRP, $\mathbf{1}$ was a vector of ones, μ was the overall mean, $\mathbf{M}$ was the marker genotype matrix with 1, 2, 3, and 4 representing A, C, G, and T alleles, $\mathbf{q}$ was the vector of SNP effects, and $\mathbf{e}$ was the vector of residuals. The SNP effects $\mathbf{q}$ were assumed to be a mixture of 4 normal distributions:

$$q_i \sim \pi_1 N\left(0, \delta_{\pi_1}^2\right) + \pi_2 N\left(0, \delta_{\pi_2}^2\right) + \pi_3 N\left(0, \delta_{\pi_3}^2\right) + \pi_4 N\left(0, \delta_{\pi_4}^2\right),$$

where $\delta_{\pi_1}^2$, $\delta_{\pi_2}^2$, $\delta_{\pi_3}^2$, and $\delta_{\pi_4}^2$ were 4 different variances of SNP effects. Proportions of SNP (π_i) in different classes of the normal mixture distribution were assumed known and set to $\pi_1 = 0.889$, $\pi_2 = 0.1$, $\pi_3 = 0.01$, and $\pi_4 = 0.001$ with extremely small, small, medium, and large effects variance (Gao et al., 2013). Residuals were assumed normally distributed with $\mathbf{e} \sim N\left(0, \mathbf{D}\delta_e^2\right)$, where $\mathbf{D}$ was the same weight matrix as in the GBLUP model. The prior distributions of SNP effects variance $\left(\delta_{\pi_i}^2\right)$ and residual variance $\left(\delta_e^2\right)$ were uniform $(0, +\infty)$, where i indicated the 4 classes of the normal mixture distribution. Each of the Bayesian analyses was run as a single chain with a total length of 50,000 Markov chain samples by Gibbs sampling, with the first 20,000 cycles discarded as burn-in. The Bayesian 4-component mixture model analyses were performed using the BayZ package (http://www.bayz.biz/).

Imputation Error Rate

Imputed genotypes of 500 randomly selected SNP from the NRF25K data set were compared with the observed genotypes, and the imputation error rate for each SNP was measured as the proportion of individuals with incorrectly imputed genotypes among the 2,130 individuals in the validation data set. Correlations between observed and imputed genotypes were also calculated for each SNP. In addition, to determine the relationship between imputation error rates and genotyping errors, we compared the 442 double-genotyped NRF bulls for the 500 validation SNP. Due the observed genotype not being the real genotype of individuals, the real genotyping error could not be measured. Therefore, the genotype disagreement rate between the genotypes of 25K and 50K chips of the 442 double-genotyped NRF bulls was used here as a measure of the genotyping error rate.

Validations of Genomic Predictions

The GEBV of validation individuals were calculated from single and multiple population predictions using both the GBLUP and the Bayesian 4-component mixture models. Accuracies of genomic predictions were calculated as correlations between GEBV and DRP, which were a proxy of the actual accuracies of GEBV. Regression coefficients of DRP on GEBV were calculated and their deviations from 1 were used as a measure of prediction biases. To further investigate reasons for increased accuracies of the multiple population predictions for the DFS bulls, accuracies of the 3 DFS populations—Danish Red, Finnish Ayrshire, and Swedish Red—were calculated individually.

RESULTS

The average error rate for imputation of NRF25K from 25K to 50K was 0.023 when using the NRF50K

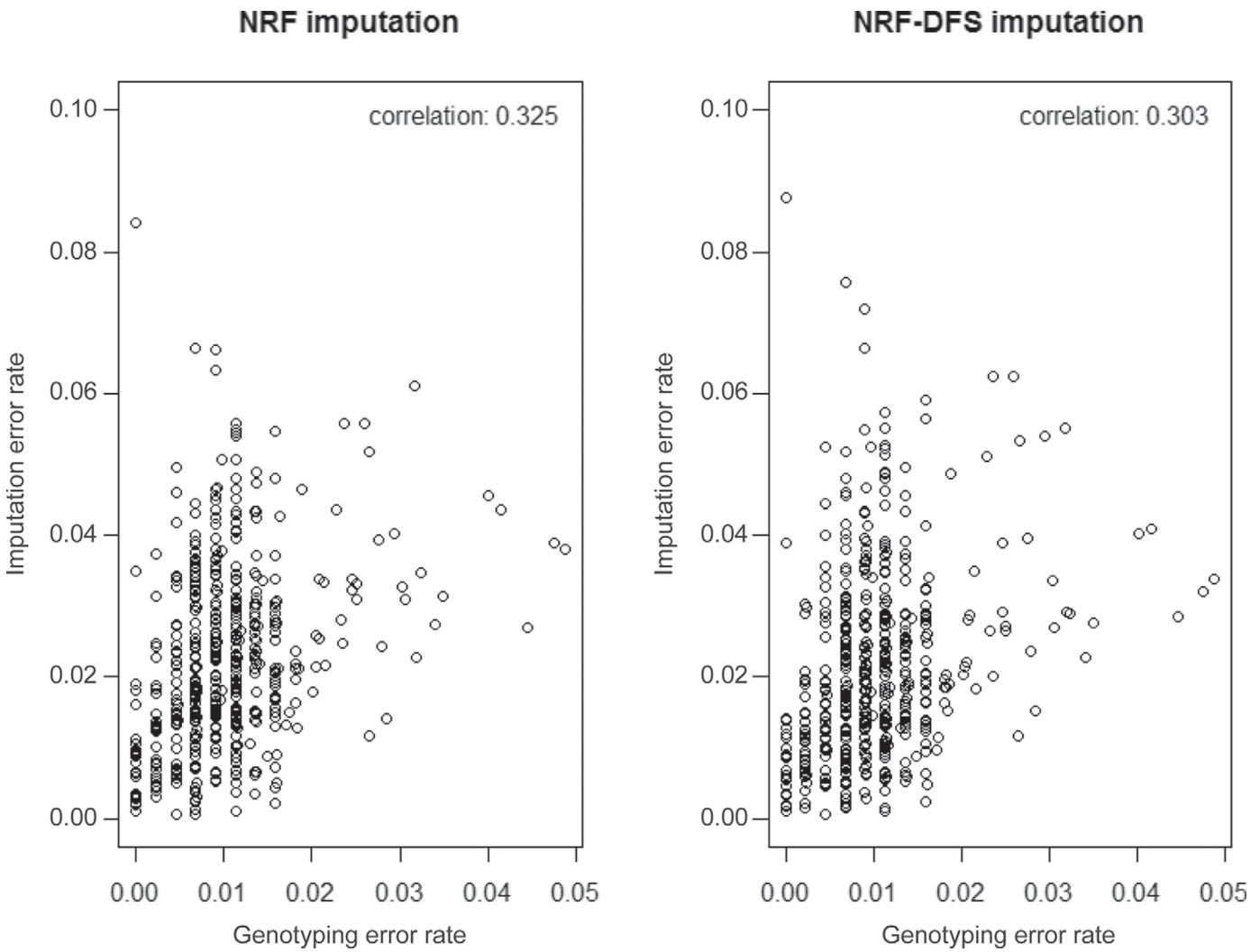

Figure 1. Comparison of imputation error rates of imputations from ~25,000 (25K) to ~50,000 (50K) for Norwegian Red (NRF) using the NRF50K reference data set or the joint NRF50K and DFS50K reference data set (where DFS = composite breed including Danish Red, Finnish Ayrshire, and Swedish Red) and genotyping error rates for the 500 validation SNP. Imputation error rates were measured as the proportions of incorrectly imputed genotypes to total number of imputed genotypes in the validation data set. Genotyping error rates were measured as the genotypes disagreement rates in the 442 bulls genotyped with both 25K and 50K chips.

reference data set and 0.021 when using the NRF50K and DFS50K joint reference data set. The imputation error rates for the 500 SNP from the 2 imputations were highly correlated (r = 0.94). Correlations between observed and imputed genotypes were, on average, 0.935 and 0.936 for the NRF50K reference imputation and the joint reference imputation. The imputation error rates were related to genotyping error rates (disagreement rate of genotypes in the double-genotyped bulls), as shown in Figure 1. The correlations between imputation error rates and genotyping error rates were 0.325 and 0.303 for the NRF imputation and the joint reference imputation.

The genomic relationship coefficients between NRF and DFS bulls from the genomic relationship matrix are shown in Figure 2. Genomic relationship coefficients were higher within breeds or populations than across breeds or populations. Figure 2 also indicates a higher genomic relationship of Swedish Red and Danish Red with NRF than that between Finnish Ayrshire and NRF. The distribution of genomic relationship coefficients of the 3 DFS populations with NRF, shown in Figure 3, illustrates that most of DFS bulls have close to zero genomic relationship with NRF.

Correlations between GEBV and DRP for the NRF and DFS validation bulls, respectively, are shown in Tables 3 and 4. In general, when the NRF and DFS bulls were combined in the joint reference population, accuracies increased for all 3 production traits by both models. For milk, fat, and protein yields, accuracies

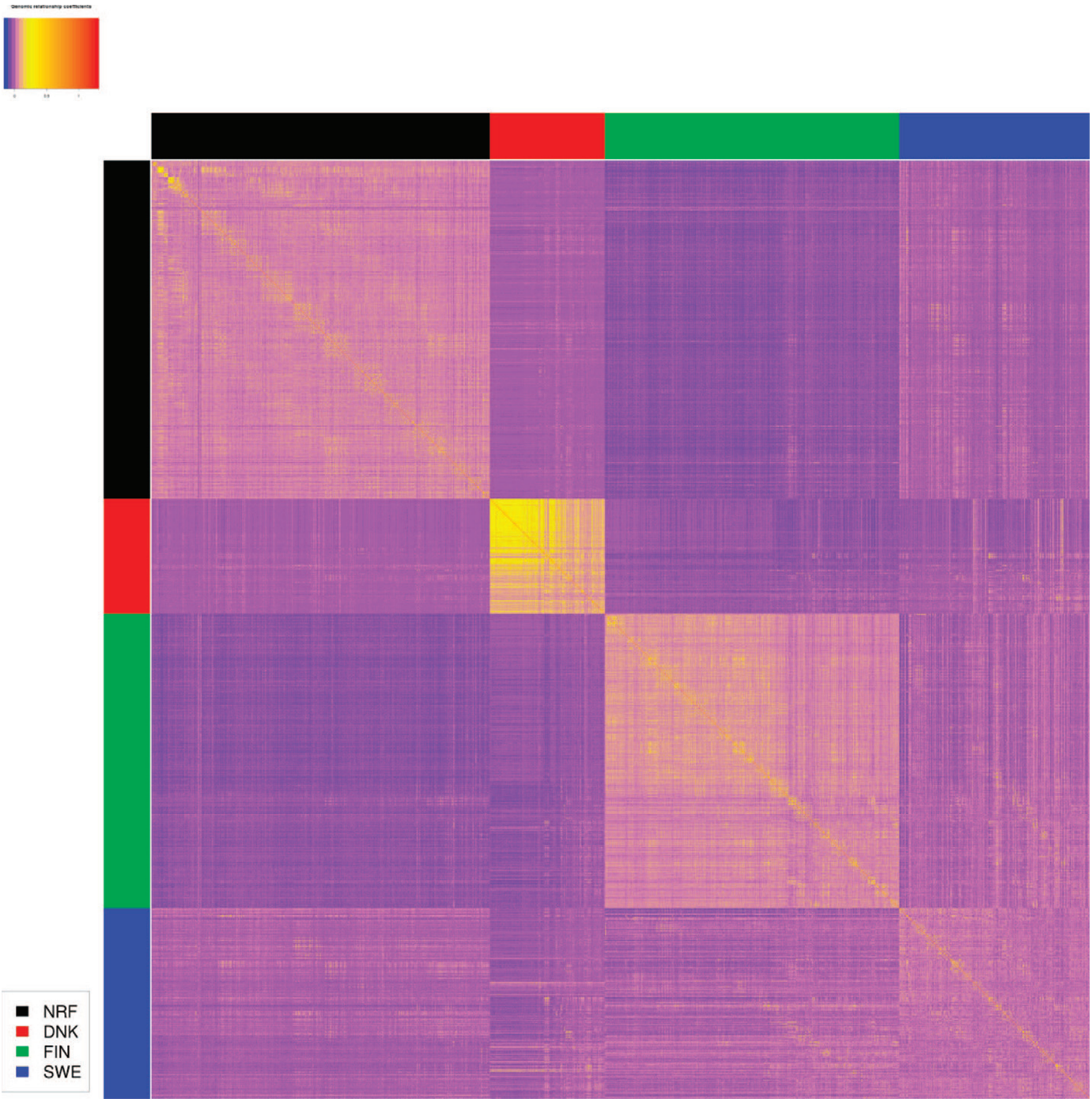

Figure 2. Genomic relationship coefficients among Norwegian Red (NRF) and composite breed (DFS) bulls, where DFS includes Danish Red (DNK), Finnish Ayrshire (FIN), and Swedish Red (SWE). The plot included 2,717 NRF, 923 Danish Red, 2,363 Finnish Ayrshire, and 1,535 Swedish Red bulls.

increased on average by 6 and 1 percentage points for the NRF and DFS bulls, respectively, using the GBLUP model, and by 9 and 1 percentage points when using the Bayesian 4-component mixture model. For NRF, regression coefficients of DRP on GEBV (Table 3) were similar for the single and multiple population predictions for the production traits, but the regression coefficients decreased slightly for the health and reproduction traits in the multiple population predictions. For DFS, there were no obvious difference in regression

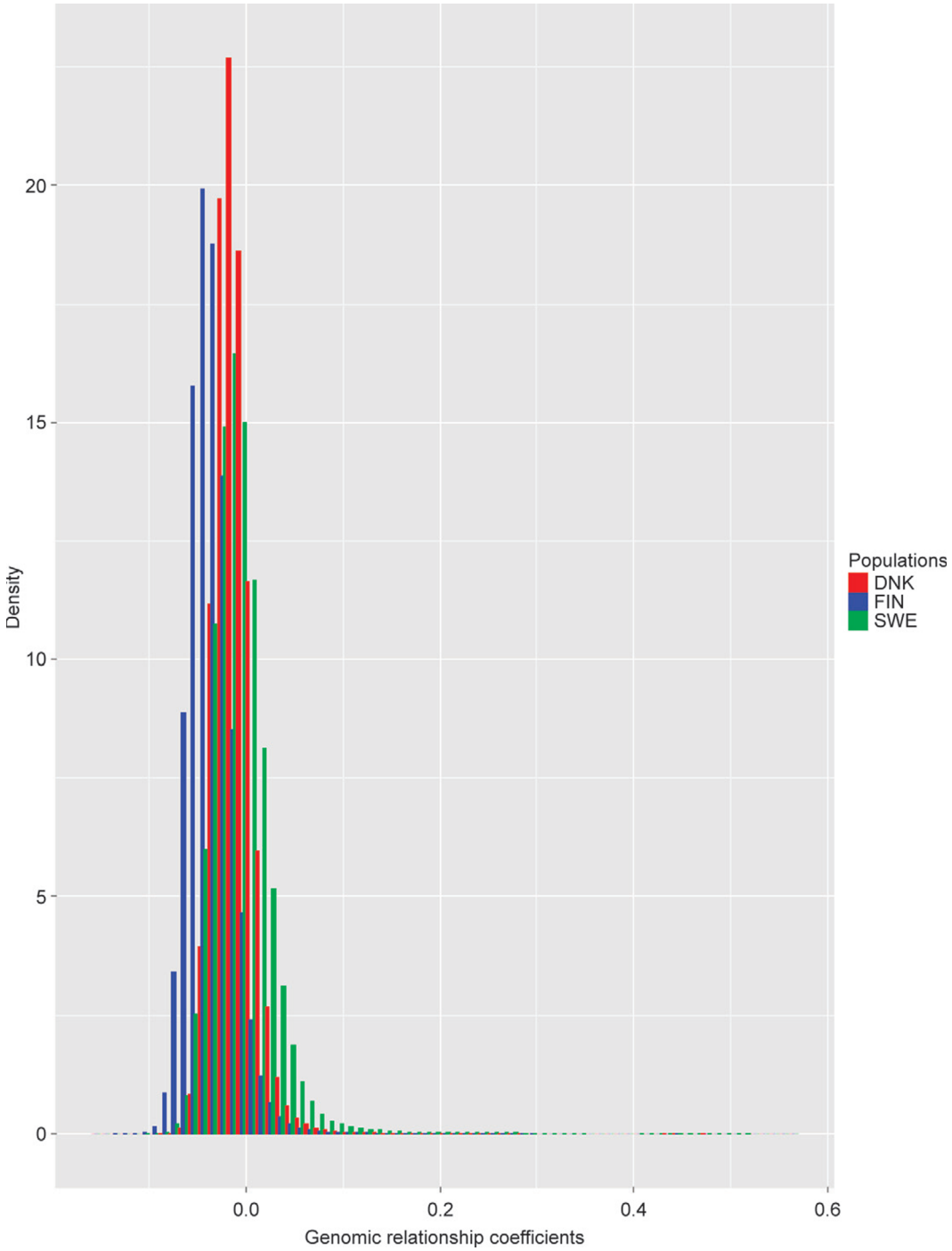

Figure 3. Distribution of genomic relationship coefficients of 3 composite breed (DFS) populations with Norwegian Red (NRF), where DFS includes Danish Red (DNK), Finnish Ayrshire (FIN), and Swedish Red (SWE). The plot included 2,717 NRF, 923 Danish Red, 2,363 Finnish Ayrshire, and 1,535 Swedish Red bulls.

coefficients of DRP on GEBV for the single and multiple population predictions (Table 4).

Accuracies of predictions by the GBLUP model for the 3 DFS populations are shown in Table 5. Danish Red had lower accuracies compared with Finnish Ayrshire and Swedish Red. Only Swedish Red showed increased accuracies for all 3 production traits in the multiple population predictions, with an increase of 3 percentage points for milk yield, 2 percentage points for fat yield, and 2 percentage points for protein yield. For

Table 3. Correlations (r; SE in parentheses) of genomic EBV (GEBV) and deregressed proofs (DRP) and regression coefficients (b) of DRP on GEBV for the Norwegian Red (NRF) validation bulls from genomic predictions with the genomic BLUP (GBLUP) and the Bayesian 4-component mixture models using either the single or multiple reference populations

	GBLUP model				Bayesian 4-component mixture model			
	Single-pop[2]		Multi-pop[3]		Single-pop		Multi-pop	
Trait[1]	r (SE[4])	b	r (SE)	b	r (SE)	b	r (SE)	b
Milk yield	0.53 (0.038)	0.87	0.58 (0.036)	0.86	0.53 (0.038)	0.88	0.62 (0.035)	0.92
Fat yield	0.58 (0.036)	0.91	0.63 (0.035)	0.86	0.59 (0.036)	0.91	0.65 (0.034)	0.88
Protein yield	0.49 (0.039)	0.81	0.57 (0.037)	0.85	0.49 (0.039)	0.80	0.62 (0.035)	0.91
NR56H	0.35 (0.042)	0.83	0.37 (0.041)	0.73	0.35 (0.042)	0.81	0.38 (0.041)	0.76
CFI	0.36 (0.041)	1.08	0.35 (0.042)	0.90	0.36 (0.041)	1.03	0.34 (0.042)	0.84
NR56C	0.35 (0.042)	1.01	0.39 (0.041)	0.84	0.34 (0.042)	0.92	0.39 (0.041)	0.84

[1]NR56H = 56-d nonreturn rate for heifers, CFI = interval from calving to first service, and NR56C = 56-d nonreturn rate for cows.

[2]Single-pop predictions: only NRF bulls were included in the reference population.

[3]Multi-pop predictions: both NRF and composite breed (Danish Red, Finnish Ayrshire, and Swedish Red; DFS) bulls were included in the reference population.

[4]Standard errors (SE) of correlations were calculated as $\sqrt{\left(1-r^2\right)/\left(n-2\right)}$, according to Snedecor and Cochran (1980), where r was the correlation of GEBV and DRP, and n was the number of individuals in the validation population.

Danish Red, accuracies increased by 2 and 1 percentage points for fat and protein yields, whereas the accuracy increased only for fat yield in Finnish Ayrshire (by 2 percentage points).

Overall, the Bayesian 4-component mixture model obtained higher accuracies than the GBLUP model for production traits. It gave slightly higher accuracies than the GBLUP model for milk and protein yields in the multiple population predictions for both NRF and DFS bulls (Tables 3 and 4). The Bayesian 4-component mixture model performed better for multiple population predictions of fat yield in the predictions of NRF but not for DFS. For fertility and health traits, the Bayesian 4-component mixture model and the GBLUP model gave similar accuracies in both the single and multiple population predictions.

For low heritability traits, such as health and fertility, accuracies were lower and less affected by the reference populations (single or multiple) or by the prediction models. For predictions of the NRF bulls, accuracies tended to increase slightly (Table 3) for NR56H and NR56C in the multiple population predictions by both the GBLUP and Bayesian 4-component mixture models. However, accuracies decreased for CFI in the multiple population predictions by both the models.

For predictions of DFS bulls (Table 4), the 2 models gave similar accuracies for fertility and mastitis, except that the accuracy for mastitis from the Bayesian 4-component mixture model decreased by 9 percentage points compared with that from the GBLUP model in the multiple population prediction. Generally, the mul-

Table 4. Correlations (r; SE in parentheses) of genomic EBV (GEBV) and deregressed proofs (DRP) and regression coefficients (b) of DRP on GEBV for the composite breed (Danish Red, Finnish Ayrshire, and Swedish Red; DFS) validation bulls from genomic predictions with the genomic BLUP (GBLUP) and the Bayesian 4-component mixture models using either the single or multiple reference populations

	GBLUP model				Bayesian 4-component mixture model			
	Single-pop[1]		Multi-pop[2]		Single-pop		Multi-pop	
Traits	r (SE[3])	b	r (SE)	b	r (SE)	b	r (SE)	b
Milk yield	0.56 (0.023)	0.78	0.57 (0.022)	0.77	0.58 (0.022)	0.82	0.60 (0.022)	0.79
Fat yield	0.60 (0.022)	0.79	0.62 (0.021)	0.79	0.61 (0.022)	0.80	0.61 (0.022)	0.84
Protein yield	0.56 (0.023)	0.75	0.57 (0.022)	0.74	0.56 (0.023)	0.73	0.58 (0.022)	0.71
Fertility	0.44 (0.025)	1.02	0.44 (0.025)	1.02	0.44 (0.025)	0.99	0.45 (0.025)	1.03
Mastitis	0.46 (0.024)	0.87	0.48 (0.024)	0.88	0.46 (0.024)	0.94	0.39 (0.025)	1.11

[1]Single-pop predictions: only DFS bulls were included in the reference population.

[2]Multi-pop predictions: both DFS and Norwegian Red (NRF) bulls were included in the reference population.

[3]Standard errors (SE) of correlations were calculated as $\sqrt{\left(1-r^2\right)/\left(n-2\right)}$, where r was the correlation of GEBV and DRP, and n was the number of individuals in the validation population.

Table 5. Correlations (r) between genomic EBV (GEBV) and deregressed proofs (DRP) and regression coefficients (b) of DRP on GEBV for Danish Red (DNK), Finnish Ayrshire (FIN), and Swedish Red (SWE), from genomic predictions with the genomic BLUP model using either the single or multiple reference populations

			Trait					
	Validation population		Milk yield		Fat yield		Protein yield	
Reference population	Population	No. of bulls	r	b	r	b	r	b
Single-pop[1]	DNK	267	0.39	0.60	0.45	0.68	0.41	0.64
	FIN	670	0.56	0.73	0.63	0.79	0.57	0.72
	SWE	412	0.62	0.88	0.64	0.87	0.62	0.86
	DFS all[2]	1,349	0.56	0.78	0.60	0.79	0.56	0.75
Multi-pop[3]	DNK	267	0.39	0.60	0.47	0.70	0.42	0.63
	FIN	670	0.56	0.71	0.65	0.79	0.57	0.71
	SWE	412	0.65	0.88	0.66	0.86	0.64	0.85
	DFS all	1,349	0.57	0.77	0.62	0.79	0.57	0.74

[1]Single-pop predictions: Only composite breed (DFS; DNK, FIN, and SWE) bulls were included in the reference population.

[2]Accuracies calculated with DNK, FIN, and SWE combined as one breed.

[3]Multi-pop predictions: Both DFS and Norwegian Red (NRF) bulls were included in the reference population.

tiple population predictions did not improve accuracies in fertility and mastitis for the DFS bulls.

DISCUSSION

Accuracies of imputation for the NRF25K from the NRF50K reference and the joint NRF50K-DFS50K reference imputations were very similar. Although many animals (13,427) from a related population or breed were included in the reference data set, imputation accuracies did not change much. The imputation error rates were similar to those reported in other studies. A previous study (Ma et al., 2013) reported that allele correct rates of imputation varied from 93.5 to 97.1% in the imputation from 3K to 50K using Swedish Red and Finnish Ayrshire data. The correlations between observed and imputed genotypes were >97.5% in the imputation from ~50,000 to ~777,000 for Fleckvieh cattle (Pausch et al., 2013). In other studies, genetic relationship has been found to be the key factor in improving imputation accuracies (Pausch et al., 2013). In our data, the 3 DFS populations were not very closely related with NRF. Among the 4,741 progeny-tested DFS bulls, 28% (412/1,486), 17% (394/2,344), and 6% (58/911) Swedish Red, Finnish Ayrshire, and Danish Red bulls, respectively, had common sires with the NRF bulls. This level of relationship among the DFS and NRF bulls may explain the small increase in imputation accuracy when DFS animals were added.

Correlations between observed and imputed genotypes for each of the 2,130 NRF validation bulls varied from 0.64 to 1.00, and around 150 individuals had correlations <0.9 from both the NRF50K reference and the joint reference imputation. Genotyping errors, which were measured as genotype disagreement rates in the 442 double-genotyped bulls in this study, are one possible reason for the lack of obvious increase in accuracies from the joint imputation. Low marker density around the SNP and low minor allele frequency of some particular SNP are other possible reasons for higher imputation error rates of these SNP. In general, the DFS data contributed very little information in the imputation of the NRF25K data.

The genomic relationship coefficients of NRF and DFS bulls describe the relationships within and between breeds and populations (Figures 2 and 3). The Swedish Red breed has a closer genomic relationship with NRF because Swedish Red has been used more in the NRF breeding program in recent years (Olsen et al., 2011). We detected some negative genomic relationship coefficients, mainly between Finnish Ayrshire and NRF (Figure 3), even though Finnish Ayrshire has been used in the breeding history of NRF (Olsen et al., 2011). This was most likely caused by our strategy of choosing the base population in building the genomic relationship matrix. We simply chose all the progeny-tested genotyped bulls from NRF and DFS as the base population. However, SNP allele frequencies differed among these 4 populations (results not shown). Therefore, the multiple population allele frequencies were influenced more by the population with more individuals, which was NRF in our study. Negative genomic relationships were also reported for French Holsteins and Montbéliarde (Karoui et al., 2012). How to set the base population in the genomic relationship matrix is an important aspect for research in genomic predictions across breeds or populations.

Accuracies of GEBV for production traits improved (5–13 percentage points) for the NRF bulls in the multiple population prediction and slightly improved (1–2

percentage points) for DFS bulls. This may be because NRF has a relatively smaller reference data set, and the DFS data set therefore contributes more useful information to the predictions for the NRF bulls. Around 3,300 DFS bulls were included in the reference data set in the multiple population predictions, and the number was 2,353 for the NRF bulls. Increased accuracies are due to the genetic relationship between NRF and DFS (Figure 2). In our data, 18% (864/4,741) of the DFS progeny-tested bulls had common sires with the NRF bulls, and 14% (366/2,572) of the NRF bulls had common sires with the DFS bulls. Generally, increased accuracies in the multiple population predictions demonstrated that related populations or breeds are useful in genomic predictions of another population or breed. A previous report (Lund et al., 2011) noted that a large reference population increased reliabilities of European Holstein. Increased accuracies of genomic predictions by merging reference populations were also reported for other cattle breeds (Jorjani et al., 2011; VanRaden et al., 2012).

Danish Red showed relatively lower accuracies of GEBV compared with Finnish Ayrshire and Swedish Red in both the DFS single population predictions and the multiple population predictions (Table 5). This is probably because the Danish Red breed has weaker genetic links to Finnish Ayrshire and Swedish Red (Brøndum et al., 2011). This was also clear from the genomic relationship coefficients of Danish Red with Finnish Ayrshire and Swedish Red (Figure 2). Among the 3 DFS populations, accuracies increased most for Swedish Red. This is because Swedish Red has the closest genetic links with NRF. Thus, a tendency exists that the closer the relationship between the populations or breeds, the greater the increase in accuracies when going from the single population to the multiple population genomic predictions. The genomic prediction methods are better to predict the effects of relatively large chunks of chromosome from key ancestors, and these large chunks of chromosome are more likely to be shared between closely related breeds or populations that have common ancestors. These results also confirm the recent arguments that close or family relationships strongly contribute to accuracies in single-breed genomic predictions (Legarra et al., 2008; Habier et al., 2010; Daetwyler et al., 2012; Wientjes et al., 2013).

Little or no increased accuracy was observed for health and fertility traits in the multiple population predictions, which agree with results of a previous study (Heringstad et al., 2011). In addition, the regression coefficients of DRP on GEBV deviated slightly further from 1 in the multiple population predictions for health and reproduction traits of NRF, indicating more prediction bias in the joint predictions. One possible reason is the differences in definitions and genetic evaluation of health and reproduction traits for NRF and DFS. For NRF, fertility was evaluated as separate traits, including NR56H, CFI, and NR56C. However, these traits were combined as an index for DFS. Genes or QTL may have different roles in each of the abovementioned traits, and their effects may become weak or diffuse in the combined index trait. Other possible reasons are that heritability of health and reproduction traits are low, and the genetic correlations between NRF and DFS of these traits are lower than for production traits (Table 1). Reliabilities of the DRP of DFS on the Norwegian scale and reliabilities of the DRP of NRF on the DFS scale for these traits are lower than for production traits. The less accurate information (DRP) from another population or breed is less useful in the multiple population predictions. It is also possible that more QTL have small effects affecting health and reproduction traits, and our current SNP density (50K) and models are not efficient in capturing these QTL.

It is assumed that most markers have very small effects and very few markers have large effects for complicated traits such as health and fertility. Bayesian models, which are consistent with this assumption, could therefore be better in genomic predictions of these traits. Many studies reported that Bayesian models showed higher accuracies than GBLUP models. A previous study (Hayes et al., 2010) reported that the BayesA model was better than the GBLUP model for fat percentage of Holstein. A Bayesian mixture model (BayesR) that had higher accuracies than the GBLUP model for milk, fat, and protein yields of Australian Holstein and Jersey (Erbe et al., 2012). Another study (Gao et al., 2013) reported that a Bayesian mixture model performed better than the GBLUP model, especially when there were fewer genetic links between the reference and validation populations.

In the present study, the Bayesian 4-component mixture model performed slightly better than the GBLUP model for the production traits but similar to the GBLUP model for the health and reproduction traits. Higher accuracy of the Bayesian 4-component mixture model in production traits could be explained by the fact that some QTL have large effects for production traits, and their effects are more accurately estimated by the Bayesian 4-component mixture model than the GBLUP model. For health and reproduction traits, the Bayesian 4-component mixture model had accuracies similar to that of the GBLUP model in both single and multiple population predictions. No advantage of the Bayesian 4-component mixture model in the single population predictions could be explained by low heritability of these traits. More QTL with smaller effects

may relate to these traits, and dominance and epistasis effects may also explain the larger amount of genetic variance in these traits than production traits. Therefore, more research is needed for genomic predictions in low heritability traits.

CONCLUSIONS

The average error rates of NRF25K decreased slightly from 0.023 to 0.021, and the correlation between observed and imputed genotype changed from 0.935 to 0.936, when comparing imputation using the NRF50K reference and the joint NRF50K-DFS50K reference populations. For production traits, accuracies of GEBV increased in the multiple population predictions for the NRF and DFS bulls by both the GBLUP and Bayesian 4-component mixture models. For health and reproduction traits, we found no obvious advantages of the multiple population predictions. The multiple population genomic predictions were beneficial for production traits in both NRF and DFS. Swedish Red, which has closer relationship with NRF, had more gain in accuracies from single population to multiple population predictions.

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