Discovering ancestors and connecting relatives in large genomic databases

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ABSTRACT

Genomic evaluation has improved both plant and animal breeding by allowing more accurate estimation of an individual’s genetic potential. Because often only a small proportion of the population to be evaluated has been genotyped, genomic estimations rely heavily on complete pedigree information. Confirmation, discovery, and correction of parentage and connected relatives allow the creation of more complete pedigrees, which in turn increase the number of usable phenotypic records and prediction accuracy. Previous methods accounted for parent-progeny conflicts using SNP. More recently haplotype methods allowed discovery of distant relationships such as maternal grandsire (MGS) and maternal great-grandsire (MGGS) with improved accuracy. However, discovered MGS and MGGS often were not used, because no dam information was available to link them to the calf. An automated procedure to discover and fill missing maternal identification information was developed, allowing discovered MGS and MGGS to be used in imputation as well as in calculating breeding values for animals in the US dairy cattle database. An MGS was discovered for 295,136 animals with unknown dam, and the MGGS was discovered for 153,909 of these animals. A virtual maternal identification was added for animals with missing information. The effect of pedigree completion on progeny inbreeding, breeding values, and reliabilities was examined. Mean inbreeding of animals with missing maternal pedigree information was 6.69% before and 6.87% after pedigree assignment; expected future inbreeding was 7.24% before and 7.20% after assignment. Reliabilities for traditional breeding values increased from 26.6 to 32.6% for milk yield, 25.9 to 32.0% for fat yield, and 26.9 to 32.9% for protein yield; genomic reliabilities also increased slightly from 76.2 to 77.1% for milk, 76.0 to 76.9% for fat, and 76.3 to 77.3% for protein. The procedure developed for pedigree completion is a useful tool for improving accuracy of national and international evaluations and aiding producers in making better mating decisions.

Key words: ancestry discovery, pedigree, genomics, genotype

INTRODUCTION

Discovery and correction of parentage has been one of the major benefits of genomic testing for plants, animals, and humans. In the case of humans, companies offer to identify ancestry, relatives, and ethnicity groups using chromosome segments shared by individuals (Ball et al., 2016). In cattle, parentage verification started by using blood group markers (Stormont et al., 1951), followed by the use of microsatellites in the 1990s (Heyen et al., 1997), and later was changed to a standard test using approximately 100 SNP markers (Heaton et al., 2002). Having more accurate pedigrees increased accuracy in estimating relationships and, therefore, reduced bias in estimates of inbreeding, genetic trend, sire variance, and across-country genetic evaluation (Banos et al., 2001; VanRaden et al., 2009). Complete pedigrees also allowed integration of genotyped and non-genotyped animals into a joint analysis using a pedigree-genomic relationship matrix (Legarra et al., 2009) and would reduce dependence on modeling unknown parents (Bradford et al., 2019).

The availability of affordable genotyping platforms allowed an increase in the number of genotyped animals and made pedigree discovery possible. Early studies (e.g., Weller et al., 2010) focused mainly on correcting or discovering sires, because most genotyped animals were males. However, introduction of the Illumina (San Diego, CA) Bovine3K BeadChip (3K) in 2010 resulted in more genotyped females that could be included in genomic evaluation and allowed for routine parentage checks (Wiggans et al., 2012). The increasing number of available SNP on commercial chips also has improved the accuracy of parentage discovery and correction. Because the information about relationships among individuals is more complete, effective confirmation and even discovery of more distant relatives has been possible using dense SNP chips and haplotypes.
The Council of Dairy Cattle Breeding (CDCB, Bowie, MD; https://www.uscdcb.com/) conducts the national genetic evaluation of dairy cattle in the United States and manages the largest cattle database in the world. For each calf record that enters the database, information about sire and dam is checked to ensure that a genotype is assigned to the correct animal and is consistent with pedigree information. The percentage of identification (ID) information that is validated is much higher for sires than for dams. For the 2.2 million animals genotyped as of January 2018, 97% of sires but only 39% of dams were validated (Wiggans et al., 2018). As the number of genotypes in the CDCB database grows daily, the number of animals that do not have dam ID reported also increases. Those animals cannot be linked properly to the rest of the population, and consequently their genetic predictions are less accurate.

Methods developed at USDA’s Animal Genomics and Improvement Laboratory (Beltsville, MD) use haplotypes to accurately discover distant relationships, such as maternal grandsires (MGS) and maternal great-granddams (MGGS; VanRaden et al., 2013). Suggestions and corrections for MGS have been reported by CDCB to breed associations and nominators of animals for genomic evaluation. However, in many cases, discovered MGS and MGGS were never used in genomic evaluation because dam ID and sometimes even maternal granddam (MGD) ID were missing, which made linking MGS and MGGS information to the calf impossible.

The first objective of this study was to develop an automated procedure to discover and fill missing maternal ID numbers in the CDCB database, thus creating a virtual ID for these animals and allowing discovered MGS and MGGS to be linked for completion of pedigrees and for national and international evaluation. The second objective was to evaluate the effect of the added pedigree information on progeny inbreeding, breeding values, and reliabilities.

MATERIALS AND METHODS

As of April 2019, the CDCB database included genotypes for almost 2.9 million animals, pedigree information for more than 80 million animals, and yield phenotypes for more than 38 million cows. Mendelian inconsistencies were detected and resolved before genotypes for all loci were imputed, by combining population and pedigree haplotyping using Fortran program findhap.f90, version 3 (VanRaden, 2017). Imputation was based on 79,294 SNP and breed-specific haplotype libraries and allele frequencies for 5 breeds (Ayrshire, Brown Swiss, Guernsey, Holstein, and Jersey), as described by VanRaden et al. (2011).

After imputation, the HAP method of VanRaden et al. (2013) was used to detect incorrect ancestors and discover missing ancestors by comparing and counting the number of haplotypes that the animal shares with a proposed relative. Haplotypes in 1 individual were evaluated by dividing each chromosome into 150-SNP segments. To find the maternal contribution after a genotyped sire was confirmed or discovered, the paternal haplotype was removed from the calf’s genotype, and a match was declared if the maternal haplotype matched either of the 2 MGS haplotypes. This process was repeated for the next generation. If an MGS was confirmed or discovered, its contributing haplotype was removed from the animal’s maternal haplotype to determine the MGD contribution, which was then checked against MGGS haplotypes (Figure 1).

The discovered ancestors were used to create a new and unique virtual ID for missing dams and to connect young animals with their ancestors, thus replacing the unknown-parent group value for that animal. In the same way, virtual dams with discovered sires and unknown dams also received a virtual dam if the MGGS was discovered, thus creating a virtual MGD. Before creating a virtual dam ID, known cow ID in the same herd as the calf were examined. If only 1 cow had a fresh date that matched the calf’s birth date and a pedigree that matched the calf’s discovered pedigree, that cow was suggested as the calf’s dam. If no actual dam was discovered, a virtual dam ID was assigned sequentially.

After pedigrees were updated with the new virtual ID, inbreeding and expected future inbreeding (EFI) (Vanraden and Smith, 1999), traditional and genomic breeding values, and their reliabilities were estimated for milk, fat, and protein yields (Vanraden et al., 2014), to evaluate the effects of more complete pedigrees.

RESULTS AND DISCUSSION

Initial Parent Status

Of the 2,868,531 genotypes in the CDCB dairy cattle database used for genomic evaluation in April 2019, 88% were from females. Before the Bovine3K BeadChip became available in September 2010, only 39% of genotypes used in genomic evaluation were from females (Council on Dairy Cattle Breeding, 2019). North America (United States, Canada, and Mexico) accounted for 90% of all genotyped animals and 72% of genotyped bulls in April 2019; the other genotypes were from more than 50 other countries. Using the 2019 CDCB parent discovery and validation method of counting opposite
homozygous SNP (Wiggans et al., 2009), the percentage of confirmed sires was 78% in April 2019, which was lower than the 88% reported for a subset of animals with 3K genotypes in September 2011 (Wiggans et al., 2012). This difference reflects the exponential growth in the number of genotypes received in recent years from many different countries, sometimes without a sire genotype in the database. The number of genotyped bulls has increased over 6-fold, from 53,788 in September 2011 to 340,906 in April 2019 (Council on Dairy Cattle Breeding, 2019). The percentage of confirmed dams was 33% in 2019, compared with 30% for the 2011 subset of animals with 3K genotypes (Wiggans et al., 2012). The number of genotyped cows was more than 32 times greater in April 2019 (2,520,514) than in September 2011 (77,753; Council on Dairy Cattle Breeding, 2019). The percentages of conflicts for sires and dams were higher in 2019 (8.6% and 1.4%, respectively) than for the 2011 3K subset (6.9% and 0.7%, respectively; Wiggans et al., 2012). The percentage of non-genotyped sires was lower in 2019 (1.3%) than for the 2011 3K subset (2.5%; Wiggans et al., 2012), as was percentage of non-genotyped dams (43.9% in 2019 compared with 60.2% in 2011).

In April 2019, an alternative sire was suggested for 95% of animals with sire conflicts, 23% of animals with a non-genotyped sire, and 80% of animals without sire pedigree information; the overall mean was 83%. An alternative dam was suggested for 73% of dam conflicts, 3% of non-genotyped animals, and 18% of animals without dam pedigree information, with an overall mean of 9%.

**Ancestor Discovery and Predictions**

Several programming modifications to reduce the number of comparisons made with the HAP method resulted in a substantial gain in processing speed for the discovery of MGS and MGGS compared with the previous method of Wiggans et al. (2018). Haplotype detection that included but was not limited to MGS and MGGS required 3.5 h with 20 processors for the entire population with 2.9 million genotypes. Wiggans et al. (2018) reported that processing took 8 h with 20 processors for 1.3 million Holsteins. Maternal grandsires were discovered for 295,136 animals with unknown dam, and MGGS were discovered for 153,909 of those animals having also unknown MGD. Of the animals with discovered MGS and MGGS, 98% were females and 2% were males, from 5 different breeds (Holstein, 90.9%; Jersey, 9.01%; Brown Swiss, 0.07%; Ayrshire, 0.01%; and Guernsey, 0.01%). Those animals were connected to their MGS and MGGS through virtual dams and virtual MGD. Because the modified HAP-method programming is faster and possibly more accurate than the method of Wiggans et al. (2018), new features for discovering additional relationships can be added to check all relatives in a single round of processing with little or no extra time cost. Relationships to sires, dams, paternal grandsires, full sibs, and clones can be

![Figure 1](image-url). Schematic representation of haplotype inheritance for a chromosome. Haplotypes from 1 individual (calf) are evaluated by dividing each chromosome into 150-SNP segments (dashed vertical lines). Original haplotypes (represented by different colors) are traced back to the maternal grandsire (MGS) and maternal great-grand sire (MGGS) by removing paternal haplotype contributions. Dam and maternal granddams (MGD) contributions can then be discovered, to allow known or virtual identification information to link the calf to its ancestors.
added to the discovery of MGS and MGGS procedures. The program can also verify or discover ancestors of only new individuals very quickly on a daily or weekly basis after editing and imputing their genotypes.

**Pedigree Inbreeding.** As expected, inbreeding and EFI changed for most (284,010 and 253,171, respectively) of the animals with a virtual dam (Table 1). Before pedigree completion, their inbreeding ranged from 0 to 16.1%, with a mean of 6.69% (SD of 0.45 percentage points); EFI mean was 7.24% (SD of 0.56 percentage points). After parentage assignment, their mean inbreeding increased by 0.18 percentage points to 6.87% (SD of 1.59 percentage points), with a range of 1.1 to 32.4%, and their EFI decreased slightly to 7.20% (SD of 0.62 percentage points). Inbreeding estimates before and after parentage assignment were statistically different \((P < 2.2 \times 10^{-16})\) and were correlated (Pearson’s product-moment) by only 0.28 for an animal’s own inbreeding (Spearman’s rank correlation of 0.42) and by 0.90 for EFI (rank correlation of 0.88). Large changes in inbreeding were expected for these animals, because they were now connected to the rest of the animals in the pedigree. Their mean inbreeding increased because lack of information on animal ancestry often resulted in mating of highly related animals. Extreme changes in an animal’s inbreeding occurred for animals with missing dam and MGD information. For example, one cow’s inbreeding increased from 7.9 to 32.4%. After this cow received a virtual dam and a virtual MGD, the new pedigree revealed that her sire was also her MGS. Although EFI also changed, the effect of virtual dams and MGD was less, because EFI measures average relationship to the whole population.

**Genomic Inbreeding.** Mean genomic inbreeding for animals with a virtual dam or MGD based on the diagonal of the genomic relationship matrix was 5.1% (SD of 2.9 percentage points), and inbreeding ranged from −7.8% (negative inbreeding indicated a crossbred animal) to 44.3%. The mean for genomic EFI based on the off-diagonals of the genomic relationship matrix was 4.1% (SD of 0.5 percentage points). The correlation between genomic and pedigree inbreeding was 0.19 before pedigree completion and 0.57 afterward. The correlation between EFI and genomic EFI was 0.71 before pedigree completion and 0.66 afterward.

**Breeding Values.** Pedigree EBV for milk, fat, and protein yields (Table 2) increased by 6% on average after pedigree completion, with increases of 116 kg for milk, 4.5 kg for fat, and 3.6 kg for protein. Reliabilities for pedigree EBV increased by 6 percentage points for milk yield (from 26.6 to 32.6%), 7 percentage points for fat yield (from 25.0 to 32.0%), and 6 percentage points for protein yield (from 26.9 to 32.9%) after adding virtual ID for dam and MGD and connecting to discovered ancestors. On average, reliabilities for pedigree EBV increased by 24% after pedigree completion.

Genomic EBV for milk, fat, and protein yields (Table 2) increased 3.4% on average after pedigree completion, with increases of 72 kg for milk, 2.7 kg for fat, and 2.1 kg for protein. Genomic reliabilities increased by 1.2% for the same traits after pedigree completion.

### Table 1. Inbreeding and expected future inbreeding based on pedigree for 295,136 animals with newly found ancestors, before and after pedigree completion

<table>
<thead>
<tr>
<th>Statistic</th>
<th>Incomplete pedigree</th>
<th>Complete pedigree</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Inbreeding (%)</td>
<td>Expected future inbreeding (%)</td>
</tr>
<tr>
<td>Minimum</td>
<td>0.00</td>
<td>2.80</td>
</tr>
<tr>
<td>Mean</td>
<td>6.69</td>
<td>7.24</td>
</tr>
<tr>
<td>Maximum</td>
<td>16.10</td>
<td>9.40</td>
</tr>
</tbody>
</table>

### Table 2. Traditional and genomic EBV value means, SD, and reliabilities for yield traits of 295,136 animals with newly found ancestors, before and after pedigree completion

<table>
<thead>
<tr>
<th>EBV</th>
<th>Trait</th>
<th>Incomplete pedigree</th>
<th>Complete pedigree</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean (kg)</td>
<td>SD (kg)</td>
<td>Reliability (%)</td>
</tr>
<tr>
<td>Traditional</td>
<td>Milk</td>
<td>1.948</td>
<td>720</td>
</tr>
<tr>
<td></td>
<td>Fat</td>
<td>72.2</td>
<td>15.1</td>
</tr>
<tr>
<td></td>
<td>Protein</td>
<td>59.4</td>
<td>14.6</td>
</tr>
<tr>
<td>Genomic</td>
<td>Milk</td>
<td>2,186</td>
<td>492</td>
</tr>
<tr>
<td></td>
<td>Fat</td>
<td>74.9</td>
<td>19.7</td>
</tr>
<tr>
<td></td>
<td>Protein</td>
<td>63.6</td>
<td>13.4</td>
</tr>
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</table>
on average after pedigree completion, with increases of 0.9 percentage points for milk and fat yields (from 76.2 to 77.1% and from 76.0 to 76.9%, respectively) and 1.0 percentage points for protein yield (from 76.3 to 77.3%). Trait evaluation models remove the effect of the cow’s own inbreeding but include EFI in PTA to predict inbreeding depression when mated to the current population (VanRaden, 2005). Correlations of genomic EBV with EFI included for milk, fat, and protein yields before and after pedigree completion were greater than 0.99 for all 3 traits. Mean EBV increased because parent averages for genotyped animals were higher than parent averages for the unknown-parent groups to which the genotyped animals were assigned before pedigree completion.

As numbers of genotyped animals increase, the need for optimized tools to effectively detect and fill in missing pedigree information is imperative. Because information from different sources is not always complete, genetic estimations are suboptimal. In June 2019, more than 11% of genotyped animals in the CDCB database had no dam pedigree information, but a likely candidate dam could be discovered in the pedigree database for more than 10% of the unknown dams and a unique virtual dam assigned for the remaining 90%. Virtual dam ID could be replaced later by true dam ID if it became known, such as from foreign pedigrees that were not provided before genotyping.

To increase efficiency, calf pedigrees should be transferred from on-farm software to dairy record processing centers, and from there to CDCB before calf genotypes arrive, so that CDCB can confirm ancestry of calves quickly and reduce the need for discovery. Growing use of embryo transfer also increases the need for better tracking of embryos in DHI records so that recipient ID are not sent as dam ID. Timely access to complete pedigrees can improve accuracy and consistency of genomic and pedigree data.

CONCLUSIONS

Discovering MGS and MGGS enabled completion of pedigrees with missing dam information, through the discovery of genotyped dams and the use of virtual ID for dams and MGD. Linking these disconnected animals with the rest of the pedigree improved genetic and genomic predictions by correcting pedigree errors and using extra information. Identifying pedigree errors early can aid in making better pedigree- and genome-guided mating decisions. Implementing parentage discovery could reduce bias in genetic predictions for animals in unknown-parent groups and improve across-country predictions.

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