# GBLUP ANALYSIS PREDICTED FERTILITY PHENOTYPES OF CROSSBRED BULLS USING DATA FROM BRAHMAN AND TROPICAL COMPOSITE

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# SUMMARY

Bull fertility directly impacts the outcome of cow herds under a natural mating system. Blood concentration of the hormone inhibin (INH), measurements of the scrotal circumference (SC18 and SC24) and the percentage of normal sperm (PNS) in an ejaculate are heritable indicators of bull fertility. We analyzed bulls from the CRC for Beef Genetic Technologies consisting of three breed types (Brahman, Tropical Composite, and Crossbreds) to which those four fertility-related traits were observed. We used 9,012 SNP markers to generate a genomic relationship matrix and to run a GBLUP analysis. We adjusted the model for the population substructure using the first two principal components derived from all genotypes. The GBLUP analyses were run twice, one with the whole dataset and another setting the phenotypes of the Crossbred animals to missing. The accuracy and bias of genomic estimated breeding values (GEBV) was estimated using the Method LR. Heritability estimates ranged from 0.17 (PNS) to 0.43 (SC24), and GEBV accuracies from 0.54 (PNS) to 0.81 (SC24). No bias was observed for any trait. Also, there is no evidence of over- or under-dispersion for INH. However, the GEBVs for PNS seems to be over-dispersed, and the ones of SCs (both SC18 and SC24) seem to be under-dispersed. The use of large enough multi-breed reference populations can lead to accurate GEBV for bull fertility traits.

#### **INTRODUCTION**

The vast majority of Australian beef cows are bull mated, especially in the north where artificial insemination is virtually inexistent. Therefore, the bull's ability to reach puberty, produce good quality sperm and effectiveness in serving cows are of fundamental importance with a direct impact on herd productivity. There are several indicators of bull fertility that are polygenic and heritable traits (Corbet *et al.* 2013). Serum levels of Inhibin (INH) measured at approx. four months of age is an early indicator of puberty (Burns *et al.* 2013). Scrotal circumference (SC) is related to bull fertility and correlated to heifer puberty (Fortes *et al.* 2012, 2013). Percentage of normal sperm (PNS) is an indicator of calf-output (Holroyd *et al.* 2002).

The application of genomic selection approaches for fertility-related traits is of interest. However, the collection of fertility-related phenotypes is expensive and the number of available animals with phenotypes and genotypes of any particular breed is too small to generate accurate estimates of breeding value. Therefore, the use of a multi-breed reference population is a valid alternative approach. The use of multi-breed genomic selection is a current hot topic of research, with some promissing results in hard to measure traits, as female fertility (Hayes *et al.* 2019).

Here we analyzed data on four traits related to bull fertility, and built a multi-breed reference population that included Brahman and Tropical Composite, to estimate GEBVs of crossbred animals. It should be noted that the results presented here are part of a work in progress towards a multi-breed evaluation, and are not final.

# MATERIALS AND METHODS

Animals and phenotypes. There were 2,979 bulls of three breed types: Brahman, Tropical

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Composites and crossbreds. They were the progeny of cows from the Beef CRC Lifetime Performance Population previously described (Barwick *et al.* 2009; Johnston *et al.* 2009). The crossbred bulls were the product of Brahman crosses with Tropical Composites. Four indicators of bull fertility (INH, SC18, SC24 and PNS) were considered; Descriptive statistics in Table 1.

**SNP genotypes.** Two SNP genotyping arrays were used, the BovineSNP50 (Illumina Inc., San Diego, CA) and the Indicus 74K array (Neogen). Initial quality control (QC) for genotypes were performed within breed and specifically to each SNP chip. After initial QC, SNP were remapped to the new bovine reference genome ARS-UCD1.2. Only SNP that were genotyped in both platforms and had a call rate greater than 95% were kept for analyses (n = 9,012 SNP). This SNP set was distributed across the genome, including the X chromosome.

**Statistical Analyses.** Principal components analysis on SNP genotypes was conducted using PLINK 1.09 (Chang et al, 2015; www.cog-genomics.org/plink/1.9/). Following recent approaches of multibreed datasets (Hayes *et al.* 2019), our GBLUP was performed using the software Golden Helix, fitting a mixed linear model with cohort (year and contemporary group) as fixed effect, and the covariates of age at measurement and PC1 and PC2 that accounts for the different breed composition. Two GBLUP runs were performed for each trait, one using the full dataset and a second setting the phenotypes of the Crossbred animals as missing. The accuracy, dispersion and bias were calculated using the Method LR (Legarra and Reverter 2018). In brief, bias was computed from the difference between the GEBV using the full data minus the GEBV setting the crossbred data as missing. Dispersion was computed from the slope of the regression of the GEBV using the full data on the GEBV with the crossbred data as missing. Finally, accuracy was computed from the covariance between the two GEBV divided by the genetic variance weighted by the average inbreeding coefficient and the average relationship between individuals.

		PNS			Inhibin			SC18			SC24	
Breed	n	Mean	SD	n	Mean	SD	n	Mean	SD	n	Mean	SD
BRM	1023	0.70	0.22	806	7.41	1.89	1098	26.70	2.71	1098	29.89	2.86
Cross	159	0.60	0.24	161	8.34	2.05	161	30.18	2.96	161	33.07	3.00
TCO	1648	0.72	0.19	1329	7.76	1.88	1719	29.82	2.82	1719	31.43	2.80

## Table 1. Descriptive statistics of samples\* and phenotypes\*\* used for analysis

\*BRM - Brahman, Cross - Crossbred, TCO - Tropical Composite.

\*\* PNS – Percent of normal sperm at 24 month of age, Inhibin – Blood level of inhibin at around 4 months of age, SC18 and SC24 scrotal circumference at 18 and 24 months of age. In table, n is the number of animals, and SD is the standard deviation.

#### **RESULTS AND DISCUSSION**

Using the principal components analysis, we captured the expected sub-structure of our population. Three main clusters were observed corresponding to each of the breed types included in the study (Figure 1). Also as expected, the Tropical Composite designation showed the highest variation within each of the breed types. While PC1 captured the differences between the three main populations (Brahman, Tropical Composites and Crossbreeds), it is the combination of PC1 and PC2 that allows the separation of substructures within populations. This is particularly the case for the two sub-populations within the crossbreds (Figure 1).

The estimates of heritability were similar to previously described for Brahman or Tropical Composite (Corbet *et al.* 2013), apart from INH that was lower in both cases (0.42, opposed to 0.72-0.74). SC have higher heritabilities in Brahman ( $\sim$ 0.75) compared to Tropical Composite ( $\sim$ 0.43), and in this study was 0.42. PNS on the other hand have higher estimates of heritability in Tropical Composite

(0.27) than Brahman (0.15), which was closer to the estimate of this study (0.18). Often it is observed variation in heritability estimates that can be mostly attributed to sample variation. As SC is easy to measure, relatively inexpensive and highly heritable, it is likely this will remain the reference trait for bull fertility. Considering the GEBV, there is no evidence of bias for any of the observed traits. Also, there is no evidence of over- or under-dispersion for INH (Table 2). However, the GEBV for PNS seems to be over-dispersed, and the ones for SC18 and SC24 seem to be under-dispersed. The population accuracies estimated using method LR are strong, especially for SC measurements.



# Figure 1. Principal component analysis on SNP genotypes for 2,979 bulls of three breed types: Brahman (blue), Crossbreds (orange) and Tropical Composites (grey)

The correlation between the GEBV estimated using all dataset, including the crossbred data, and those estimated setting the crossbred data to missing values varied between traits (Figure 2), from moderate (0.35) for PNS to high (0.77) for SC18.

 Table 2. Estimates of heritability, accuracy, bias and dispersion for GEBV of fertility-related traits in bulls

Trait	Heritability	GEBV accuracy	Bias	Slope
PNS	0.176	0.544	-4.36 x10 <sup>-10</sup>	0.970
Inhibin	0.419	0.685	-3.50 x10 <sup>-9</sup>	1.006
SC18	0.423	0.799	8.15 x10 <sup>-9</sup>	1.033
SC24	0.428	0.811	1.01 x10 <sup>-8</sup>	1.018

\* PNS – Percent of normal sperm at 24 month of age, Inhibin – Blood level of inhibin at around 4 months of age, SC18 and SC24 – Scrotal circumference at 18 and 24 months of age.

#### CONCLUSIONS

There are still some improvements that could be done before implementation of multi-breed genomic selection for bull fertility-related traits e.g. better understanding how to model different populations in different environments, and consistency in trait measurement. The lack of bias and the high accuracy of the estimates are encouraging and warrant further research.

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# Figure 2. GEBV of Crossbred using all data, including own record (x-axis) and without own records (y-axis)

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