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Phenotypic and genetic exploration of first- and second-generation progeny sired by gene bank collection bulls

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Introduction

Livestock gene banks have developed genetically diverse collections to safeguard important breeds and/or unique within breed populations for reconstitution under extraordinary circumstances over the last 20+ years. While additional livestock gene bank sample uses are emerging (Blackburn et al., 2024) animal gene bank use has yet to experience the broad range of use seen by the plant community; for example, since 2012 plant gene banks have distributed approximately one million samples to 120 countries (Crop Trust, 2025). Lack of gene bank use has resulted in some nation's questioning the utility of developing collections of animal genetic resources. Gollin (2020) has discussed issues of forecasting use and return on investment of genetic conservation and highlighted that future genetic resource use and timeframe are unknown, termed the option value, and therefore difficult to quantify. To start addressing these knowledge voids we explore two separate uses of Holstein and Angus samples from a gene bank.

A common perception for not using gene bank samples has been that samples held by the gene bank represent genetics from past generations and therefore may be of limited utility when compared to current genotypic and phenotypic performance. But in 2020 Dechow et al. reported reintroduction of Y chromosomes lost from the current Holstein population using gene bank semen samples. While genomic predictions of progeny performance were below breed average, they were substantially higher than the parental mid-point. Phenotypically cows produced by this first mating outperformed contemporary genetic herd mates for milk production and lifetime performance in the Pennsylvania State University herd.

Due to the unexpected performance of the first-generation cows, we explored possible reasons for this unexpected result by evaluating the performance and genomic composition of first and/or second-generation Holstein and Angus progeny of collection bulls born from 1954 to 1997.

Materials & Methods

Yue et al. (2015) found current Holstein bulls carried Y chromosomes from either Elevation (51%), Chief (49%), or Ivanhoe Star (<1%) with all other sire linages deleted from the in-situ population. In the 1960's era they reported 1,821 bulls with potentially different Y chromosomes, these bulls traced back to at least four Dutch bulls based upon association pedigree data. These researchers identified gene bank samples from Allstar Pilot (born in 1954) and Caliban Cuthbert (born 1989) that carried two additional Y chromosomes linages which were requested to test if these lost Y chromosomes (LY) could be reintroduced into Holstein. Withdrawn semen samples were used via in-vitro fertilization with oocytes from cows with genomic breeding values in the upper 70th percentile for net merit. Resulting bull (n=5) progeny were raised, and semen collected and cryopreserved, heifers (n=8) became part of the Pennsylvania State University herd. First generation bulls were mated to commercial Holstein in New Mexico and Pennsylvania with second generation animals having 25% gene bank sires. Resulting heifers (n = 128) remained in their respective herds for characterization and comparison to their contemporary herd mates (n=172). First and

second-generation progeny were genotyped using high density Bovine SNP 777K bead chip. Runs of homozygosity (ROH) were computed with PLINK v1.9 (Purcell et al. 2007) and ROH metrics were evaluated with PROC GLM in SAS 9.4.

The second request was from an elite Angus breeder typically selling 8,000 bulls per year and wishing to improve maternal characteristics in their nucleus herd of approximately 800 cows. The firm used five gene bank bulls born from 1958 to 1997. The breeder also had acquired additional semen from older bulls of the same time frame via commercial sources. The matings resulted in 214 progeny that were compared to 4,791 contemporary genetic herd mates from the breeder's nucleus herd using a 40K commercial SNP chip for genotyping. A mixed model was used to analyse phenotypic traits with fixed effects of: sex, cow/heifer, IVF, herd-year-season, animal age; and random effects of animal. Runs of homozygosity (ROH) were identified using PLINK v1.9 (Purcell et al. 2007), and the overlapping ROH regions were compared between animals in the top and bottom 10% of contemporary animals for weaning and yearling weight genomic estimated breeding values vs ROH of the gene bank/older bulls' progeny.

Results

First generation of Holstein gene bank progeny produced 12% more milk than contemporary herd mates (Table 1). While first generation gene bank Angus progeny were 4.5% heavier at birth and 1.7% lighter at weaning compared to the contemporary herd mates.

Table 1. Phenotypic performance for first generation Angus and Holstein progeny.

	Angus		Holstein
Genetic Group	Birth wt, kg	Weaning wt, kg	305-day milk, kg
Contemporary	38.1	261.8	14,194.1
Gene Bank	39.8	257.3	15,895.9

Figure 1 illustrates how second-generation gene bank progeny performed when produced in a commercial dairy herd in Pennsylvania and that 38% of gene bank granddaughters exceeded the average for the contemporary progeny to which they were compared.

Figure 1. Predicted Transmitting Ability (PTA) for milk by 305-day mature equivalent milk yield (ME305d, kg) in a Pennsylvania commercial herd for contemporary (blue) and second-generation gene bank progeny (red) cows. Green dashed line is the mean ME305d for contemporary cows and solid purple is one standard deviation from contemporary cow mean.

Introducing Holstein gene bank bulls and the continued use of their progeny for a second generation resulted in animals with fewer runs of homozygosity; generation 1 = 58.8 ROH & 210 Mb, generation 2 = 86.7 ROH & 320 Mb, and contemporary progeny = 112.1 ROH & 506 Mb. For Angus ROH were similar for top and bottom 10% of contemporary progeny but ROH for gene bank progeny were 72 and 72.8% lower for the group of SNPs associated with weaning weight. The overlap of ROH among gene bank and the two groups of contemporary progeny were $\leq$12% (Fig 2).

Figure 2. Number of ROH (Mb) and percent overlap of ROH for first generation gene bank Angus vs the top 10% and bottom 10% of contemporary Angus progeny for weaning weight.

Discussion

In these two examples of gene bank collection use, the progeny from the first and/or second generation performed at similar levels to progeny from contemporary sires for phenotypic traits despite the gene bank bulls representing genetic combinations that were developed 25 to 70 years ago (potentially more than 10 generations). These phenotypic results are counter to previous findings. However, breeding value estimates were substantially less (Fig 1) when compared to contemporary animals, potentially due to fewer genetic ties currently used in genomic evaluation programs and/or higher than expected levels of heterosis. These findings were surprising given Holstein and Angus are among the most highly selected breeds in the U. S. Additionally, previous work with small breeds and simulation (Eynard et al. 2017; Leroy et al., 2011) have not been as positive. The results obtained may be due to reducing ROH in both the first and second generation. In addition, second generation grand-progeny of collection bulls comprised of 75% contemporary genetics which may also be contributing to higher performance levels (Fig 1). In separate studies using 1960 era Holstein genetics, gene bank progeny increased allele frequencies for resistance to disease (Sarlo Davila et al. 2023). It was noted in the Pennsylvania State University herd first generation Holstein had a longer productive life than contemporary herd mates; but this was based upon a small sample size.

Given the relatively new use of animal gene bank collections and the findings reported, there are several elements that a user may wish to consider when using gene bank collection samples. These include mating collection bulls to relatively high performing females; generate enough progeny to sufficiently utilize recombination effects; and use in vitro fertilization to increase the number of progeny produced per unit of semen. Finally, many animals in a gene bank are likely to have been superior sires or dams during their lifetime. As a result, they may have been more than one standard deviation above the breed average for specific traits of interest. This suggests that the genetic difference between older animals and contemporary genetics may not be as large as initially supposed, indicating their utility when used in current populations.

Conclusions

Results suggest that previous concerns about low performance of gene bank collection germplasm is not well founded. Industry and the research communities should, without reservation and where appropriate, utilize gene bank holdings to support their breeding objectives. Tracking the lifetime performance for first and subsequent generations of gene bank collection progeny will further confirm performance levels and approaches for effectively using this resource.

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