

**Angus Cattle at High Altitude: Genetic Relationships and Initial Genome-Wide Association Analyses of Pulmonary Arterial Pressure**

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**ABSTRACT:** Records from yearling Angus (n = 10,647) cattle from elevation 2,340 m were used in genetic analysis of pulmonary arterial pressure (PAP). Bulls were developed within a grain-supplemented performance test, whereas heifers and steers were grazed. The BovineSNP50 Beadchip was used to genotype a subset of cattle (n = 1,690). Bulls had greater (P<0.001) yearling PAP values as compared to steers and heifers. Heritability ranged from 0.20 in heifers and steers to 0.31 ± 0.15 in bulls. Moderate positive genetic correlations were observed in PAP measures with heifers and bulls (0.64 ± 0.14) and steers (0.74 ± 0.37). Genome-wide association analyses using multi-locus mixed model and Bayesian approaches revealed several QTL for PAP; however, no concordant SNP (P<0.001) were observed among males and females. Therefore, results suggest that PAP appears to be a polygenic trait influenced by sex and (or) growth.

**Keywords:** high altitude disease; genetic correlation; GWAS

**Introduction**

Hypoxic-induced pulmonary hypertension in cattle is a noninfectious disease of cattle at elevated altitudes (>1,500 m). The condition causes increased workload for the heart, ultimately resulting in right heart failure. Clinical signs include distension of the jugular vein, dyspnea, weakness, ascites, and edema of the jaw and brisket region. In the U.S., an estimated \$60 million are attributed each year to losses associated with hypoxic-induced pulmonary hypertension (Williams et al. (2012)). Historically, up to 40% of cattle transported from lower elevations to higher elevations developed the disease compared to only 2% of native populations (Will et al. (1975)). Pulmonary arterial pressure (PAP) is the predominant measure used to evaluate risk of an animal’s susceptibility to develop hypoxic-induced pulmonary hypertension. Initial research estimated a moderate heritability (0.46 ± 0.16; Enns et al. (1992)) for PAP in cattle. Additionally, moderate genetic correlations have been reported between males and females for post-weaning PAP (Shirley et al. (2008)). Because breeders often manage and feed seedstock cattle for maximal performance, nutritional management strategies may subsequently impact cattle response to hypoxic conditions. We hypothesized that sex and (or) growth management will impact genetic analyses of PAP.

**Materials and Methods**

**Animals.** Research procedures were approved by Colorado State University Institutional Animal Care and

Use Committee. Records from yearling Angus (n = 10,647) cattle from the Colorado State University Beef Improvement Center (CSU-BIC; One Bar Eleven, Rouse Ranch in WY; 2,340 m altitude) were used for genetic analyses of PAP, which is a measure used to evaluate an animal’s response to hypoxic conditions (i.e., high altitude). A PAP value > 50 mmHg for an individual suggests risk and susceptibility to developing pulmonary hypertension. Genome-wide association analyses were conducted on a subset of cattle (n = 1,690) where DNA samples and PAP measurements were collected over a 15 year period. Cattle averaged 344 ± 0.86 days of age at time of data collection and the bulls were being developed in a performance test (i.e., ration targeting gain of ~1.5 kg/day), while heifers and steers grazed (i.e., dormant winter forage with potential gain of ~0.5 kg/day).

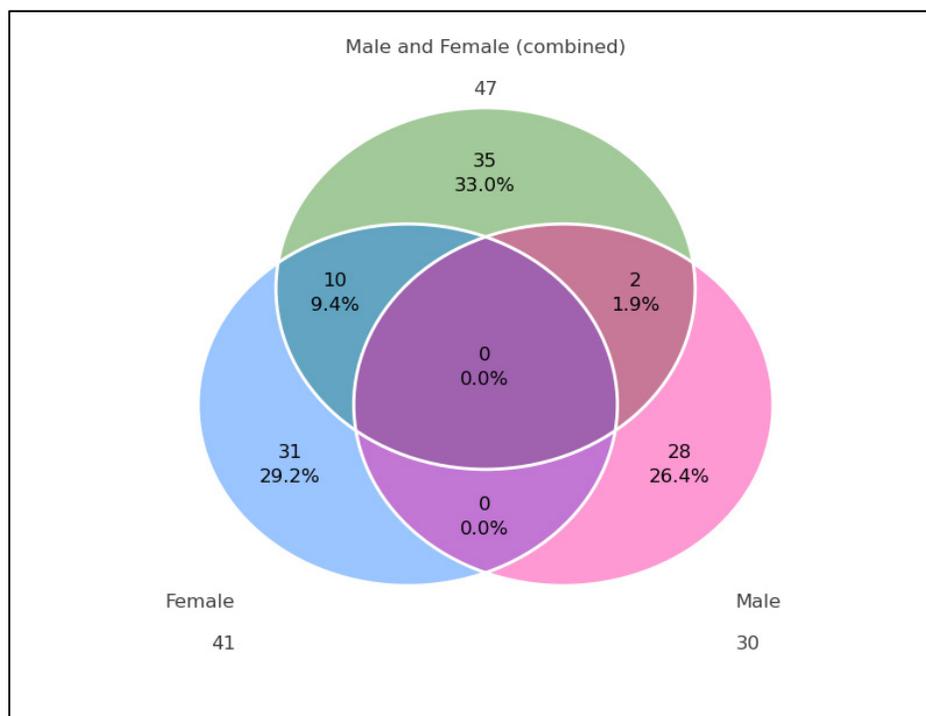
**Table 1.** Descriptive statistics of yearling pulmonary arterial pressure (PAP) for full and subsets of data from Angus cattle managed at high altitude (> 2300 m).

| Item                                 | n     | Min | Mean  | Max | SD    |
|--------------------------------------|-------|-----|-------|-----|-------|
| Full data (n = 10,647)               |       |     |       |     |       |
| Combined                             | 4,812 | 22  | 42.55 | 139 | 10.10 |
| Heifer                               | 2,957 | 22  | 41.38 | 135 | 8.66  |
| Bull                                 | 1,423 | 29  | 45.35 | 139 | 11.99 |
| Steer                                | 430   | 27  | 41.29 | 138 | 10.62 |
| Subset data used in GWAS (n = 1,690) |       |     |       |     |       |
| Combined                             | 1,551 | 26  | 41.88 | 139 | 9.45  |
| Heifer                               | 1,126 | 26  | 41.02 | 116 | 7.72  |
| Bull/Steer                           | 425   | 32  | 44.16 | 139 | 12.99 |

**Table 2.** Heritability estimates (**diagonal**) and genetic correlations (above diagonal) for yearling pulmonary arterial pressure (PAP) in Angus heifers, bulls and steers (n = 10,647) managed at high altitude (> 2300 m).

| Sex    | Heifer             | Bull               | Steer              | Full               |
|--------|--------------------|--------------------|--------------------|--------------------|
| Heifer | <b>0.21 ± 0.04</b> | 0.64 ± 0.14        | 0.74 ± 0.37        | -                  |
| Bull   | -                  | <b>0.38 ± 0.08</b> | 0.82 ± 0.37        | -                  |
| Steer  | -                  | -                  | <b>0.20 ± 0.15</b> | -                  |
| Full   | -                  | -                  | -                  | <b>0.31 ± 0.03</b> |

**Statistical analyses.** Distribution of PAP measures was right-skewed; therefore, the log<sub>10</sub> of yearling PAP measures (n = 10,647) were used in statistical analyses using the Box-Cox procedure in R. Fixed effects in this study included year of birth, date of birth, age of dam, and PAP date. Summary statistics from analyses in R are presented in **Table 1** for the full and subsets of data for heifers, bulls and steers. The word “combined” indicates all



**Figure 1.** Venn diagram of the number of SNP detected with multi-locus mixed model GWAS of yearling pulmonary arterial pressure (PAP) for combined (males and females;  $n = 47$  SNP), female ( $n = 41$  SNP), and male (bulls and steers;  $n = 30$  SNP) Angus cattle in high altitude ( $> 2300$  m). Percentages represent the number of SNP as per the total ( $n = 118$ ; 100%).

three animal classes (i.e., bulls, steers, and heifers) were pooled. Genetic parameters for the animal classes were obtained using ASReml (Table 2). A univariate animal model was fitted to the full dataset to obtain heritability estimates for yearling PAP and a multivariate model was fitted to the full dataset to obtain genetic correlations between sexes.

**Genome-wide association study.** A commercial laboratory (Zoetis, Kalamazoo, MI) isolated DNA from each sample ( $n = 1,690$ ), which was then genotyped using the Illumina BovineSNP50 BeadChip. A genome-wide association study (GWAS) was conducted using a single SNP association analysis (MLMM: multi locus mixed model) and a SNP-window ( $\sim 1$  Mb) association analysis (BayesC). Both analyses were conducted for the female (heifers), male (bulls and steers), and combined (bulls, steers, heifers) data. The SNP & Variation Suite 7 (SVS7; Golden Helix, Bozeman, MT) was used for the single-SNP association GWAS analyses and the GenSel software (Iowa State University) was used for the SNP-window association analyses. Venn diagrams were generated as to compare the number of QTL observed among data from females, males, and combined (males and females).

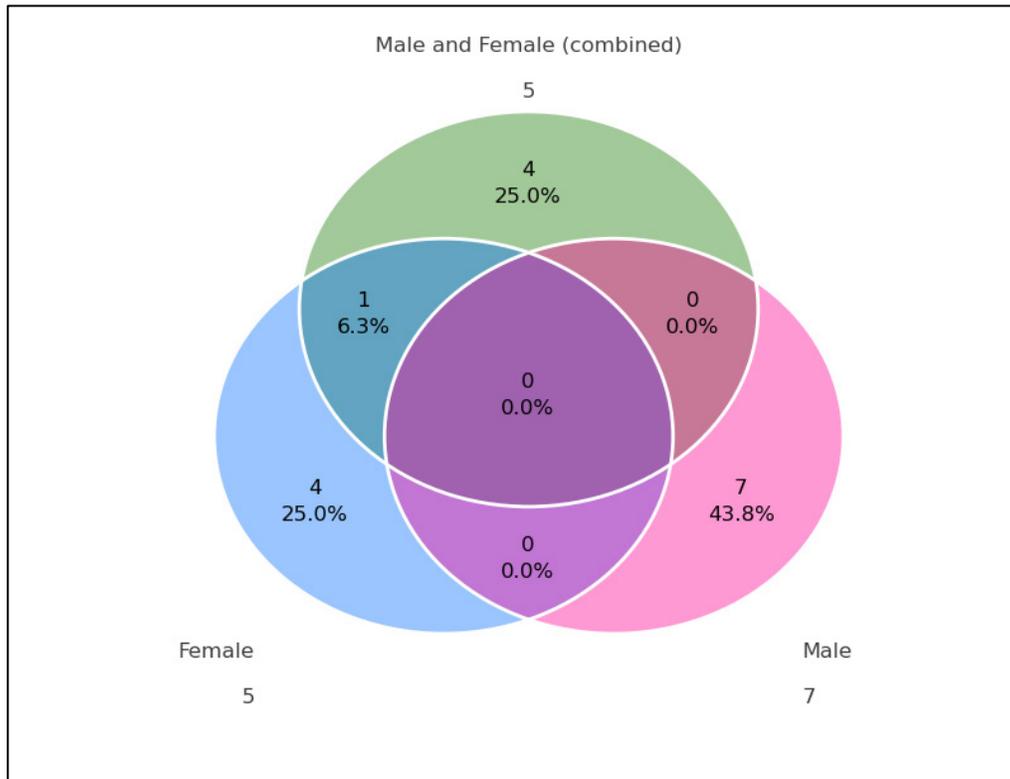
## Results and Discussion

**Descriptive statistics.** Bulls had increased ( $P < 0.001$ ) PAP scores compared to heifers and steers in the full dataset (Table 1). A somewhat similar result was observed in the data used for GWAS; however, due to

limited number of males with genotypes, the bull and steer (i.e., male) classes were combined.

**Heritability.** Moderate heritability estimates of PAP were observed in heifers, bulls, steers, and combined analyses of yearling PAP (Table 2). Previous research involving Angus cattle in western Colorado reported a moderate to high yearling PAP heritability ( $0.46 \pm 0.16$  Enns et al. (1992)), whereas in another study a moderate estimate ( $0.34 \pm 0.16$ ) was observed (Shirley et al. (2008)). In general, heritability estimates in the current study were lower than previous reports, which may be attributed to variety of reasons such as increased animal numbers, different population of cattle and (or) ranch locations, etc. Regardless, genetic trends for yearling PAP at CSU-BIC have consistently decreased over the past 15 years (Enns et al. (2011)). Heritability estimates and genetic trends across these studies suggest it is possible to make genetic improvement in PAP score.

**Genetic correlations.** Moderate positive genetic correlations were observed for yearling PAP between heifers and either bulls or steers (Table 2). Furthermore, there was a high positive genetic correlation for yearling PAP between bulls and steers. Shirley et al. (2008) also observed a moderately positive genetic correlation ( $0.64 \pm 0.12$ ) for PAP measured post-weaning between male and female calves. These types of results involving male and female cattle were observed in a previous study by Garrick et al. (1989). Specifically, genetic correlations for post-weaning gain between males and females differed greatly among expected and observed values (Garrick et al.



**Figure 2.** Venn diagram of the number of Bayesian-GWAS SNP-windows ( $\geq 1\%$  genetic variance) of yearling pulmonary arterial pressure (PAP) in combined (male and female;  $n = 4$ ), female ( $n = 5$ ), and male ( $n = 7$ ) Angus cattle in high altitude ( $> 2300$  m). Percentages represent the number of SNP as per the total ( $n = 17$ ; 100%).

(1989)). Therefore, similar to post-weaning gain, results of the current study suggest that yearling PAP represents a different trait for heifers and bulls; however, additional research is needed to delineate differences among sexes and their physiology versus differences that are due to nutritional management and rate of gain.

**Quantitative trait loci.** The single SNP GWAS approach identified 41, 30, and 47 loci across the 30 chromosomes associated ( $P < 0.001$ ) with yearling PAP in the female, male and combined data, respectively (**Figure 1**). The SNP-window approach detected 5, 7, and 5 QTL associated with PAP for female, male, and combined analyses, respectively (**Figure 2**). Results from MLMM and BayesC analyses did not reveal any SNP-QTL concordant to the male and females groups. However, the single SNP and SNP-window analyses revealed several similar loci. Specifically, there were concordant loci detected on chromosomes 7 and 28. Results from the two GWAS approaches support the findings of the genetic correlation analyses and suggest that additional research is needed to demarcate differences in genetic regulation of PAP due to sex and (or) growth rate, which appear confounded in this study.

## Conclusions

Pulmonary arterial pressure measures appeared polygenic in Angus cattle sampled at high altitude. Also, this study supported our hypothesis that sex and (or) growth management influences genetic analyses of PAP. Specifically, varied heritabilities, moderate genetic correlations, and lack of concordant QTL suggest that PAP is a different trait among males and females growing at different rates. Future research is necessary to elucidate the influence of growth management on the genetic response of cattle to hypoxia.

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