Refinement of the DST Locus Associated with Bovine Respiratory Disease Complex in Holstein Calves

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ABSTRACT

Despite best management practices including vaccination and treatment programs, bovine respiratory disease complex (BRDC) continues to be a major cause of morbidity and mortality in cattle. An additional approach to reduce BRDC is to select animals that are less susceptible to respiratory disease. A previous genome-wide association analysis (GWAA) identified a 10kb QTL region on chromosome 23 (BTA23) that included the dystonin (DST) gene that plays a major role in herpes virus infections in cattle. The objective of this study was to determine if there were additional variants on BTA23 with a greater association with BRDC that could be used in the selection of cattle with enhanced BRDC resistance. An analysis was conducted using genotypes imputed to whole genome sequence (WGS) for preweaned Holstein calves from California that consisted of 996 controls and 982 cases as defined by the McGuirk health scoring system. Illumina BovineHD BeadChip genotypes on BTA23 were imputed to WGS using Run S data from the 2010 Bull Genomes Project and accuracy was checked by using the WGS of 30 Holstein calves included the study. Imputation was conducted using Fimpute and errors were corrected by using Beagle 4.1 software. SNPs were filtered for low minor allele frequency (<1%) and low call rate (<90%) resulting in 2,196 biallelic markers being used for analysis in a one Mb region surrounding DST. Analyses were performed using an allelic model and an additive model with Efficient Mixed Model Association expedited (EMMAX) that included age and sex as covariates. A 2.5kb region, including intron 57 (ENSEMBL) of DST, contained 19 SNPs that were associated with BRDC with both allelic (P = 6.72x10⁻⁷ to P = 5.07x10⁻⁵) and additive models (P = 3.82x10⁻⁶ to P = 6.88x10⁻⁵). Many of the 19 SNPs were highly conserved across species suggesting that they may have a functional or regulatory role in gene expression. These SNPs will be used to confirm the BRDC association in independent cattle populations to determine their value for inclusion in a genomic PTA for BRDC.

Keywords: Bovine respiratory disease complex, dairy, calves, imputation, loci

INTRODUCTION

• Bovine respiratory disease complex (BRDC) is a common infectious disease of cattle worldwide and accounts for 22.5% of preweaned and 46.5% of weaned heifer deaths (NASS, 2006)
• A previous GWAA identified a 10kb QTL region (consisting of 5 significant loci) on bovine chromosome 23 that included the dystonin (DST) gene (Neibergs et al., 2014)
• The proportion of variance explained (PVE) for all 5 SNPs was 0.13
• DST plays an important role in transportation of bovine herpes simplex virus 1 (BHV-1) capsids to the nucleus of the host cell through the microtubule network (McElwee et al., 2013). BHV-1 is the pathogen responsible for infectious bovine rhinotracheitis or IBR.

Figure 1. Schematic representation of DST (BTA23: 3,431,407 – 3,687,495 bp) encompassing 100 exons. A previous GWAA identified a 10kb QTL region associated with BRDC susceptibility in intron 57.

OBJECTIVE

The objective of this study was to determine if there were additional variants on BTA23 with a greater association with BRDC than previously identified that could be used in the selection of cattle with enhanced BRDC resistance.
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MATERIALS AND METHODS

1968 preweaned Holstein calves

Genotyping with Illumina BovineHD BeadChip

Imputation to whole genome level on BTA23

Association analysis in 1 Mb region surrounding DST

Allelic model and an additive model with Efficient Mixed Model Association expedited (EMMAX)

996 controls and 982 cases

777,962 markers

Run 5 data from 1000 Bull Genomes with Fimpute Errors corrected with Beagle 4.1 software

Quality control: Call rate < 0.9 and MAF < 0.01 Remaining SNPs: 2,196

Age and sex as covariates
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RESULTS

Table 1. A 2.5kb region within intron 57 (ENSEMBL) of DST containing 19 SNPs associated with BRDC with both allelic and additive model

<table>
<thead>
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<th>Marker Position (bp)</th>
<th>Allelic Model</th>
<th>EMMAX Additive Model</th>
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<td>3.82E-06</td>
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<td>3.83E-06</td>
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<td>4.45E-06</td>
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<tr>
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</table>

Figure 3. Significant markers plotted along intron 57 of DST with neighboring exons

Figure 4. Sequence alignment of 2.5kb region within intron 57 of DST with 17 eutherian mammals

- 93% sequence alignment with sheep
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**DISCUSSION**

- **DST** has 59 homologs in different species including sheep, pig, horse, dog, human, fish, fruit fly and *C. elegans*. This conservation across diverse species indicates evolutionary significance of the **DST** gene.

- Down regulation of **DST** gene expression in single pathogen challenge with common BRDC pathogens (BRSV, BVDV, IBR, *M. haemolytica* and *M. bovis*) (Tizioto et al., 2016) supports its role in BRDC susceptibility.

- The closest upstream and downstream splice site variants to these markers are on exon 53 and exon 63.

- The sequence underlying the QTL (Figure 3) is conserved across 17 eutherian mammals.

- Both the allelic model which tests the differences in allelic frequencies, and the additive model which tests if allele effects are additive are associated with BRDC susceptibility.

**CONCLUSIONS**

- Imputation facilitated refinement of the 10kb QTL region to a 2.5kb region with intron 57 of **DST**.

- SNPs associated with BRDC were highly conserved across species suggesting that they have a functional or regulatory role in gene expression.

- Accuracy of imputation and markers will be further validated by genotyping of these markers in Holstein populations.

- Future research will validate BTA23 markers in an independent population so that they may be included in a genomic PTA for BRDC.

**ACKNOWLEDGEMENTS**

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**REFERENCES**


