Research Overview

Bovine Respiratory Disease Complex
Coordinated Agriculture Project

http://www.brdcomplex.org/

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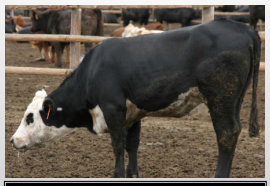
OUTLINE

Introduction
- Aims of project & overview

Research Projects
- Identifying loci for selection
- Development of selection panel
- PTA estimates
- Animal welfare
- Economics
- **Results & Conclusions**



INTRODUCTION

PROBLEM

Same level of morbidity and mortality from BRDC over the past 20 years despite utilizing:

- Best management practices
- Preventative vaccines
- Improved treatments

We need new approaches to reduce the incidence of BRDC in addition to our current approaches!

RESEARCH AIMS

1. Identify genomic regions associated with BRDC susceptibility in beef and dairy cattle
   - Genome wide association, gene set enrichment, copy number variant analyses
     - 2700 Holstein calves
     - 2000 *Bos taurus* steers and heifers
   - Pathogen challenge study for gene expression
2. Develop BRDC genetic selection panel that can be used by the cattle industry
3. Develop PTA estimates
4. Assess how animal welfare is affected by BRDC
5. Assess economic impact of BRDC and selection

RESEARCH PROJECTS

ANIMALS

- Dairy – preweaned calves 2000 from CA, 784 from NM
  - Genome wide association analysis (GWAA)
  - Gene set enrichment analysis (GSEA)
  - Copy number variant analysis (CNV)
- Beef feedlots – 1000 from CO, 1000 from WA
  - GWAA, GSEA
- 46 Angus steers UC Davis
  - RNA-seq for gene expression

Diagnosing BRDC: McGuirk Calf Health Scoring System

Now available as an ipad app!

Calf Health Scoring Criteria			
0	1	2	3
Rectal Temperature (F°)			
100-100.9	101-101.9	102-102.9	≥103
Cough			
None	Induced Single Cough	Induced Repeated Coughs or Occasional Spontaneous Cough	Repeated Spontaneous Coughs
Nasal Discharge			
Normal Serous Discharge	Small Amount of Unilateral Cloudy Discharge	Bilateral, Cloudy, or Excessive Mucus Discharge	Copious Bilateral Mucopurulent Discharge
Eye Scores			
Normal	Small Amount of Ocular Discharge	Moderate Amount of Bilateral Discharge	Heavy Ocular Discharge
Ear Scores			
Normal	Ear Flick or Head Shake	Slight Unilateral Droop	Head Tilt or Bilateral Droop

BRDC PATHOGENS

- Diagnostics for *Mycoplasma*, *P. Multocida, M. Haemolytica, H. Somni,* bovine respiratory syncytial virus, bovine viral diarrhea virus, bovine corona virus completed from swabs

DAIRY GWAA

Results (Neibergs et al., 2014. BMC Genomics 15:1164)

- 4 statistical approaches produced highly concordant results for case-control:

Population	# SNPs	# Chromosomal Regions	Heritability
CA	373	126	21%
NM	370	138	21%
CA & NM	324	116	13%

BEEF AND DAIRY GSEA

- GWAA using principle component analysis (EIGENSTRAT) and 19,723 genes were represented by most significant markers within the gene's haplotype using the method of Gabriel et al. (2002) which averaged 14 kb (beef) and 20 kb (dairy)
- GSEA performed using GenGen software (Holden, 2008)
- 4,389 gene sets were evaluated from Gene Ontology, KEGG, Reactome, Biocarta, and Panther
- Significance was based on normalized enrichment scores >3.0

DAIRY GSEA

Gene Set	# Genes (# LEG)	NES	P value
CA			
Nitric oxide stimulates guanylate cyclase	25 (18)	3.2	0.0005
NM			
Fatty acid biosynthetic process	53 (36)	4.0	0.0001
Monocarboxylic acid biosynthetic process	65 (41)	3.7	0.003
Fatty acid metabolic process	93 (50)`	3.6	0.0003
Organic acid biosynthetic process & carboxylic acid biosynthetic process	103 (56)	3.4	0.0008
Monocarboxylic acid metabolic process	143 (72)	3.4	0.001

NM: 86 unique leading edge genes; CA 18 unique leading edge genes

RNA-SEQ

Gene expression after challenges with *Mycoplasma bovis*, *Pasteurella multocida, Mannheimia haemolytica,* bovine respiratory syncytial virus, bovine viral diarrhea virus, and infectious bovine rhinotracheitis (bovine herpes virus 1) as described (Gershwin et al., 2015 PLosOne)

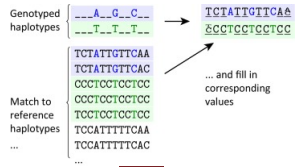
Tizioto et al., 2015 PlosOne

- 142 differentially expressed genes in bronchial lymph nodes were from GWAA identified QTL
- Pathogen specific expression profiles

SEQUENCING

15 Holstein cases and 15 Holstein controls were sequenced to identify additional variants under QTL

- Determine if the new markers are more informative or predictive for BRDC susceptibility for genotyping panel
- Use for imputation to whole genome sequence



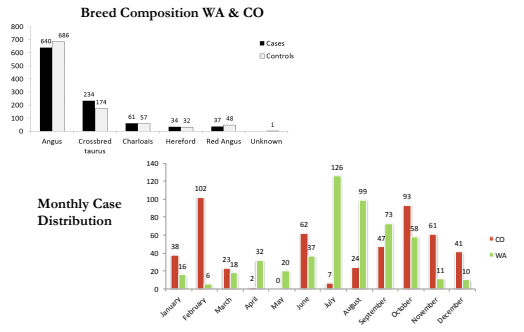
ILLUMINA GGP 250

- Content includes some SNPs associated with BRDC susceptibility in dairy
- 2000 BRDC dairy and beef samples have been genotyped with the GGP 250 and GWAA analysis will be presented at JAM by Dr. Taylor's lab.

IMPUTATION

- Jesse Hoff (Dr. Taylor's lab) has imputated BovineHD BeadChip genotypes to whole genome sequence level for the dairy samples based on the 1000 bull project sequences and BRDC dairy sequences.
- Accuracies checked by comparing to Holstein sequence data and select genotyping of SNPs identified as associated with BRDC

BEEF GWAA



BEEF GWAA

Two analyses (EMMAX & EIGENSTRAT) were conducted for case-control and clinical score phenotypes (most QTL were shared across phenotypes)

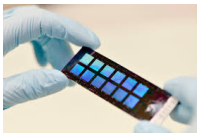
- Case-control shared QTL
  - CO: 9 QTL
  - WA: 10 QTL
  - CO & WA: 24 QTL
- Clinical score shared QTL
  - CO: 9 QTL
  - WA: 6 QTL
  - CO & WA: 20 QTL
- Estimated heritability ≈ 24% for case-control, ≈ 20% for clinical scores

BEEF GSEA

Gene Set	# Genes (# LEG)	NES	P value
CO & WA			
Integral component of plasma membrane	137(59)	3.6	0.001
Negative reg. cellular protein metabolic proc	207 (79)	3.1	0.003
Steroid binding	20 (7)	3.1	0.003
Reg. G protein coupled receptor protein signaling	40 (17)	3.1	0.002
CO			
Reg. peptidase activity	110 (45)	3.1	0.0015
Reg of proteolysis	169 (73)	3.0	0.002
WA			
Alzheimer disease amyloid secretase pathway	61 (27)	3.5	0.0005
Reg RNA pathways	23 (11)	3.3	0.0003

WA: 38 unique LEG; CO: 74; combined: 155; 79 unique LEG with challenged calves; 6 LEG overlap with dairy

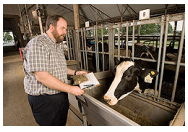
DAIRY & BEEF GENOTYPING CHIP



- Custom chip designed from QTL with major individual or collective effects
- Variants surrounding QTL based on sequencing data of beef and dairy cattle
- Genotypes on all animals will be done
- Markers from this, GGP 250 and BovineHD BeadChip used to choose best markers to be used commercially

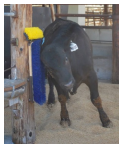
PTA DAIRY

- Predicted transmitting abilities are being estimated for Holsteins and these data will be presented at JAM by Drs. Gordon Spangler and Curt Van Tassell
- Can be incorporated into Net Merit selection indexes for use in selection of dairy



ANIMAL WELFARE

- NSAID (meloxicam) and antibiotic treatment vs antibiotic alone in BRSV and *H. somni* challenged steers was evaluated via behavior and clinical exams used to determine if the use of NSAID benefited BRDC affected cattle (Toaff-Rosenstein et al. 2016, Applied Animal Behaviour Science)



- Access to environmental enrichment was assessed to determine if it was helpful in diagnosis of BRDC animals

ECONOMICS

Economic cost of the disease in dairy and beef

- Estimates of economic gain from selection to reduce BRDC incidence in dairy calves (Neibergs et al. 2014)
- Economic benefits of using genetic selection to reduce the prevalence of bovine respiratory disease complex in beef feedlot cattle (Neibergs et al. 2014)

RESULTS AND CONCLUSIONS

TRANSLATION TO INDUSTRY

- Common phenotype – standardize and report
- Marker panel enabling the identification of susceptible cattle
- Predicted transmitting ability for AI dairy sires for BRD susceptibility
- Ability to identify and select against cattle with BRD susceptibility
- Identify break-even point for BRD prevention based on economic analyses

WHICH KEY SECTORS HAVE BEEN ADDRESSED?

- Beef industry – feedlot (25 million cattle/year)
- Dairy industry – large dairies; pre-weaned calves; milk production and fertility of mature cows

ACKNOWLEDGEMENTS

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BOVINE RESPIRATORY DISEASE CONSORTIUM