An analysis of differential gene expression profiles of half-sibling Limousin bulls divergent in intramuscular fat measured post-slaughter

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I. INTRODUCTION

Intramuscular fat is an important characteristic influencing beef sensory quality and nutritive value [1]. There is a genetic component to beef sensory quality suggesting a possibility to select for animals with improved meat quality [2, 3]. However despite its fundamental importance, meat quality is a trait missing from national breeding objectives. The objective of this study was therefore, to identify differentially expressed genes associated with meat quality as a step towards providing a basis for the identification of functionally relevant polymorphisms. Single nucleotide polymorphisms (SNPs) in the regulatory regions of these genes could potentially be incorporated into breeding programs if significant associations are found.

II. MATERIALS AND METHODS

Paternal half sibling Limousin steers were slaughtered and muscle (*M. longissimus thoracis et lumborum* (LTL)) samples were collected immediately post mortem for intramuscular fat analysis and RNA preservation. A half sibling model was utilized to attempt to reduce the polygenic variation not associated with the meat quality phenotype, while ensuring sufficient genetic variation in the study. Immediately following slaughter, the IMF% of the LTL muscle in the half sibling bulls was assessed using AOAC methods. Tissue for RNA extraction was also collected in RNA-later. The 20 half-sibling bulls most divergent for IMF% were chosen for RNA extraction and RNA-sequencing. One hundred and fifty base paired-end sequencing was run on an Illumina HiSeq2000 platform Following read alignment differential gene expression analysis was conducted using the R package DESeq2. The list of DE genes was investigated for enrichment analysis of gene ontology (GO) terms for Biological Processes (BP) and KEGG pathways, using the functional annotation tool and functional annotation clustering of Database for Annotation, Visualization and Integrated Discovery (DAVID) bioinformatics resources.

III. RESULTS AND DISCUSSION

Overall, 882 genes were differentially expressed between the half sibling bulls divergent for IMF%. Three of the top differentially expressed genes, *RYR1*, *MYOM2* and *MYBPC2* are known to influence meat quality due to their role in sarcomere structure along with skeletal muscle function and development. To further assess how changes in gene expression impact the animal biologically, the differentially expressed gene list was submitted to DAVID and the differentially expressed gene ontology and KEGG terms are presented in Table1. The differentially expressed terms suggest changes in the regulation of ‘transcription’, ‘cell to cell adhesion’

and ‘insulin signaling’ between steers divergent in meat quality. The top differentially expressed GO term was ‘transcription DNA-templated’. Within this GO term many zinc finger genes were differentially expressed. The zinc finger proteins have previously been associated with adipogenesis [4]. Changes in ‘cell to cell adhesion’ could potentially impact meat quality by affecting the deposition of fat within the muscle. Insulin signaling pathway was the top KEGG term. Insulin signaling is important in growth and muscle development with *IGF2R* a differentially expressed gene and the genes *TFRC, RAB11FIP2, VPS26B, GRK2* and *CLTB* involved in vesicle mediated transport, essential for insulin signaling.

Table1: Gene ontology & KEGG terms overrepresented among differentially expressed genes

Term	Count	*P* value
GO:0006351~transcription, DNA-templated	130	6.51E-04
GO:0098609~cell-cell adhesion	30	0.007079
GO:0045944~positive regulation of transcription from RNA polymerase II promoter	71	0.014369
hsa04910: Insulin signalling pathway	23	0.044796

IV. CONCLUSION

The results from this study identify differentially expressed genes, KEGG pathways and gene ontologies differentially expressed between steers with high and low IMF%. These panels of genes present an opportunity to identify functional SNPs associated with meat quality in the promoter region of these genes with potential to be subsequently utilized as genetic markers of meat quality in beef.

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