ABSTRACT

ASSESSMENT OF STATE-SPECIFIC DNA METHYLATION PATTERNS TO IMPROVE FUNCTIONAL ANNOTATION OF FARM ANIMAL GENOMES

By

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Over the past several decades, genetic advancements in the domestic pig (*Sus scrofa*) and other farm animal species have resulted in increased economic output and expanded use of these organisms as biomedical models to study human disease. However, limited functional annotation of the porcine genome—particularly in non-coding regulatory regions—hinders both identification of causal genes for complex traits and translational research capabilities. The Functional Annotation of Animal Genomes consortium seeks to map functional elements in domesticated animal genomes in part by performing sequencing assays to characterize the animal epigenome, as specific chromatin modifications have been shown to be predictive of regulatory regions. DNA methylation is the most ubiquitous epigenetic modification made to the DNA molecule, and in mammals occurs almost exclusively at cytosines in CpG dinucleotides. DNA methylation exerts regulatory effects through numerous mechanisms, including the occlusion of transcription factors at activating regulatory regions, and as such has been shown to play important roles in establishing spatiotemporal gene expression. Furthermore, differential methylation has been associated with genomic imprinting and stress-induced physiological changes in mammals. Assessment of DNA methylation in the pig and other farm animal species has thus far been limited in scope. In this dissertation, I have characterized state-specific DNA methylation patterns in farm animal genomes across a diverse collection of cell types, developmental stages, and environmental conditions, to enhance understanding of epigenetic gene regulation in livestock and poultry. **First**, I demonstrate that sorted porcine immune cells exhibit unique DNA methylation landscapes that are strongly

correlated with local and distal gene expression as well as binding sites for transcription factors regulating immune cell-specific functions. The co-localization of immune cell differentially methylated regions with GWAS SNPs for immune-related traits supports the use of epigenomics assays to increase functional annotation of economically relevant genomic regions. **Second**, I show that development of four porcine fetal tissues (whole brain, liver, loin muscle, and placenta) is associated with increased differentiation of DNA methylation profiles that likely contributes to tissue-specific transcriptomes and transcription factor regulatory potential. I also report widespread allele-biased methylation in fetal tissues associated with breed-specific gene regulation as well as putative regions of genomic imprinting events. **Third**, I characterize associations between environmental stimuli and DNA methylation patterns in two studies. I show that piglet weaning correlates with changes in peripheral blood mononuclear cell DNA methylation, and that increased weaning stress is associated with increased methylation and decreased expression of T cell-enriched genes, suggesting a diminished adaptive immune response. Lastly, I assess the impact of broiler chick incubation parameters on cardiac DNA methylation and observe significant temperature-associated differential methylation of genes involved in heart morphogenesis. I identified differentially methylated and expressed genes between temperature treatments that may influence environment-driven differences in cardiovascular development. In conclusion, I have performed the most expansive survey of whole-genome DNA methylation in farm animal species to date and have identified thousands of putative regulatory elements influencing state-specific gene and phenotype expression. These data will be a valuable resource for future functional annotation efforts seeking to identify mechanistic links between genetic and phenotypic variation in animal species.