A164 · Integrated analysis of Chromatin Accessibility and Regulon Activity suggests candidate Regulatory Programs in Polarized Porcine Monocyte-derived Macrophages (MDM)

Presenter: Mehak Kapoor — Iowa State University

Authors: USDA-ARS-NADC

Session: Day 1 · Thu Oct 1, 2026 · 2:15–4:15 PM

Macrophage polarization is governed by signaling to transcription factors (TFs) controlling gene expression. While chromatin accessibility is necessary for TF binding, whether footprint-inferred TF occupancy corresponds to TF-driven target gene expression within polarized states remains unresolved. Thus, we performed ATAC-seq and scRNA-seq on MDMs comparing resting (M0, GM-CSF), pro-inflammatory (M1, IFN-ɣ) and anti-inflammatory (M2, IL-4) states. We found SCENIC-predicted regulon activity revealed distinct, state-specific transcriptional signatures. IRF-family regulon activity was more pronounced in M1 compared to M0 and M2, while FOSL2 activity was more elevated in M2 compared to M0 and M1. We further identified 41,105 differentially accessible peaks (DAPs) across pairwise comparisons, predominantly in distal intergenic/intronic regions. Notably, 24.6% of DAPs were consistently differential in M1 (vs. M0 and M2) and 9.2% in M2 (vs. M0 and M1), versus only 0.07% consistently differential in M0 (vs. M1 and M2), indicating M1 and M2 each establish a distinct consistent chromatin signature. Pathway enrichment linked M1 vs. M0 DAPs to inflammatory pathways, M2 vs. M0 DAPs to hypoxia-associated pathways, and M2 vs. M1 DAPs to TNFα/TGF-β signaling. Differential TF footprinting showed increased IRF-family binding distinguishing M1 from M0, AP-1 (FOS, JUNB) binding distinguishing M2 from M1, and KLF-family/STAT6 binding distinguishing M2 from M0, consistent with canonical IFNɣ and IL-4 effector activity and partially agreeing with SCENIC-derived regulon scores. To further test whether chromatin-level occupancy predicts transcriptional output, we plan to integrate SCENIC-derived regulons with footprint-derived TF-target gene sets and determine target-gene-level concordance across polarization states.

Integrated analysis of Chromatin Accessibility and Regulon Activity suggests candidate Regulatory Programs in Polarized Porcine Monocyte-derived Macrophages (MDM)

Mehak Kapoor[1*], Carrie S. Meeks-Patience[2], Kristen A. Bryne[3], Crystal L. Loving[3#], Christopher K. Tuggle[1,2#]

[1]Bioinformatics and Computational Biology Program and Department of Animal Science, Iowa State University, Ames, IA
[2]Immunobiology Program, Department of Animal Science, Iowa State University, Ames, IA
[3]USDA-ARS-NADC, Food Safety and Enteric Pathogens, Ames, IA

Macrophage polarization is governed by signaling to transcription factors (TFs) controlling gene expression. While chromatin accessibility is necessary for TF binding, whether footprint-inferred TF occupancy corresponds to TF-driven target gene expression within polarized states remains unresolved. Thus, we performed ATAC-seq and scRNA-seq on MDMs comparing resting (M0, GM-CSF), pro-inflammatory (M1, IFN-γ) and anti-inflammatory (M2, IL-4) states.

We found SCENIC-predicted regulon activity revealed distinct, state-specific transcriptional signatures. IRF-family regulon activity was more pronounced in M1 compared to M0 and M2, while FOSL2 activity was more elevated in M2 compared to M0 and M1. We further identified 41,105 differentially accessible peaks (DAPs) across pairwise comparisons, predominantly in distal intergenic/intronic regions. Notably, 24.6% of DAPs were consistently differential in M1 (vs. M0 and M2) and 9.2% in M2 (vs. M0 and M1), versus only 0.07% consistently differential in M0 (vs. M1 and M2), indicating M1 and M2 each establish a distinct consistent chromatin signature. Pathway enrichment linked M1 vs. M0 DAPs to inflammatory pathways, M2 vs. M0 DAPs to hypoxia-associated pathways, and M2 vs. M1 DAPs to TNFα/TGF-β signaling. Differential TF footprinting showed increased IRF-family binding distinguishing M1 from M0, AP-1 (FOS, JUNB) binding distinguishing M2 from M1, and KLF-family/STAT6 binding distinguishing M2 from M0, consistent with canonical IFNγ and IL-4 effector activity and partially agreeing with SCENIC-derived regulon scores.

To further test whether chromatin-level occupancy predicts transcriptional output, we plan to integrate SCENIC-derived regulons with footprint-derived TF-target gene sets and determine target-gene-level concordance across polarization states.