Epigenomic Landscapes from Various Cells and Tissues of *Gallus gallus*

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General Overview

- Annotate *cis*-regulatory elements and novel transcripts in the chicken genome.

- Identify changes in epigenomic states that correspond to differences in *cis*-regulatory activity and expression.

Hawkins et al., 2010 *Nat Rev Gen*
General Overview

• *Gallus gallus* reference/data production ADOL line 6x7 F1.
  – assay optimization: SPF.

• Targeted tissues and cells for Phase 1:
  – Blood cells: macrophage, B cells, polarized T cells; lung macrophages.
  – Reproductive tissues: ovaries, oviduct, shell gland.

• Assays
  – RNA-seq
  – ATAC-seq
  – ChIP-seq
  – WGBS
  – 3D genome architecture
Regulation of gene expression

• Transcriptional regulatory elements
  – Promoters
  – Enhancers
  – Insulators
  – Silencers
ATAC-seq

Buenrostro, 2013
• ATAC-seq on lung and kidney tissue.
• ~72,000 peaks called per tissue.
• ~4% reads map to chrM.

<table>
<thead>
<tr>
<th>Analysis Step</th>
<th>Read Count</th>
</tr>
</thead>
<tbody>
<tr>
<td>Original</td>
<td>50,204,006</td>
</tr>
<tr>
<td>After trimming</td>
<td>50,204,006</td>
</tr>
<tr>
<td>Remove Unmapped</td>
<td>43,122,572 (14.1% loss)</td>
</tr>
<tr>
<td>Remove Orphaned Reads &amp; non-unique reads</td>
<td>43,122,450</td>
</tr>
<tr>
<td>Remove Duplicates</td>
<td>31,789,256 (22.6% loss)</td>
</tr>
</tbody>
</table>
ChIP-seq

- H3K4me3: novel & alternative promoters
- H3K4me1: enhancer elements
- H3K36me3: validate novel transcripts from RNA-seq
- CTCF: insulators
- H3K27ac: indicative of active promoters/enhancers
- H3K27me3: repressed, bivalent promoters
ChIP-seq

H3K4me3 from lung
ChIP-seq

- Number of peaks called: 27,424
- Number of peaks at annotated TSS: 19,552
DNA methylation: WGBS

ENCODE/ROADMAP DATA
DNA methylation: WGBS

- Unmethylated regions (UMRs) – broad regions largely devoid of DNA methylation
  - UMRs: Largely indicative of promoters (and CGIs)
- Lowly methylated regions (LMRs) – narrow regions of low/modest DNA methylation
  - LMRs = Largely indicative of enhancers
WGBS validation of transcription

Hawkins et al., 2010 Cell Stem Cell
## WGBS QC from 6x7 F1 chickens

<table>
<thead>
<tr>
<th>Cell Type</th>
<th>BSC rate (%)</th>
<th>Mapping efficiency (%)</th>
<th>CpG methylation (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Macrophage rep1</td>
<td>99.7</td>
<td>81.2</td>
<td>62.0</td>
</tr>
<tr>
<td>Macrophage rep2</td>
<td>98.7</td>
<td>77.5</td>
<td>65.0</td>
</tr>
<tr>
<td>B cells rep1</td>
<td>99.6</td>
<td>81.8</td>
<td>63.8</td>
</tr>
<tr>
<td>B cells rep2</td>
<td>99.4</td>
<td>77.9</td>
<td>66.9</td>
</tr>
<tr>
<td>Th cells rep1</td>
<td>99.1</td>
<td>82.5</td>
<td>72.6</td>
</tr>
<tr>
<td>Th cells rep2</td>
<td>99.9</td>
<td>81.7</td>
<td>71.5</td>
</tr>
</tbody>
</table>
QC: mapping bias
Summary

• Expression and Epigenomic assays have been optimized for data production.

• Blood cells and reproductive tissue have been isolated from production/reference line.

• PLAC-seq is being optimized to map promoter-enhancer interactions.
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