Supplemental Figures

Single-cell chromatin and gene-regulatory dynamics of mouse nephron progenitors Sylvia Hilliard,¹ Giovane Tortelote,¹ Hongbing Liu,¹ Chao-Hui Chen,¹ and Samir S. El-Dahr^{1*} ¹Section of Pediatric Nephrology, Department of Pediatrics, Tulane University School of Medicine, New Orleans, LA 70112, USA

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Supplemental Figure 1. Quality control for the Singleome scATAC-seq data (E16.5/P2).

- (A) Insert size distribution of the scATAC-seq samples showing characteristic nucleosomal periodic patterns.
- (B) Transcription start sites (TSS) signal enrichment of the scATAC-seq samples.

Supplemental Figure 2. TSS and unique nuclear fragments enrichment in Singleome scATAC-seq data (E16.5/P2).

- (A) Violin plots showing TSS enrichment scores per sample at E16.5 and P2.
- (B) Violin plot showing the log(base10) of unique nuclear fragments per sample.
- (C) scRNA seq: median number of genes (left) and reads (right) per cell.

Supplemental Figure 3. Quality control of Multiome scATAC/GEX (E16.5/P2).

(A) scATAC-seq: unique fragments/TSS, TSS enrichment scores, unique nuclear fragments, fragment size and TSS enrichment profiles

(B) scRNA-seq: median genes/reads per cell.

Supplemental Figure 4. Differential gene-specific chromatin accessibility across cell clusters. Representative genes of progenitors (*Six2*), early (*Wnt4, Lef1*) and late (*Lhx1, Hnf1b*) differentiating, proximal (*Hnf4a*), LOH/distal (*Pou3f3 and Tfap2b*) segments are shown. Supplemental Figure 5. Chromatin accessibility identifies new markers of tubular differentiation.

(**A**, **B**) scATAC-seq UMAPs: *Mcmdc2* chromatin accessibility is higher in proximal than distal precursors. *Nabp1* chromatin accessibility is seen in both proximal and distal precursors.

(**C-J**) Confocal images of RNAscope. *Mcmdc2* is expressed in LTL-positive proximal tubules (C, D) and in Slc12a3+ distal tubules (H,I). *Nabp1* is expressed in LTL-positive as well as LTL-negative/Slc12a3-positive segments (E, F, H, J). *Mcmdc2* and *Nabp1* are not expressed in nascent nephron of the nephrogenic zone (NZ) (G).

Supplemental Figure 6. *Meis2* is expressed in stroma and NPCs.

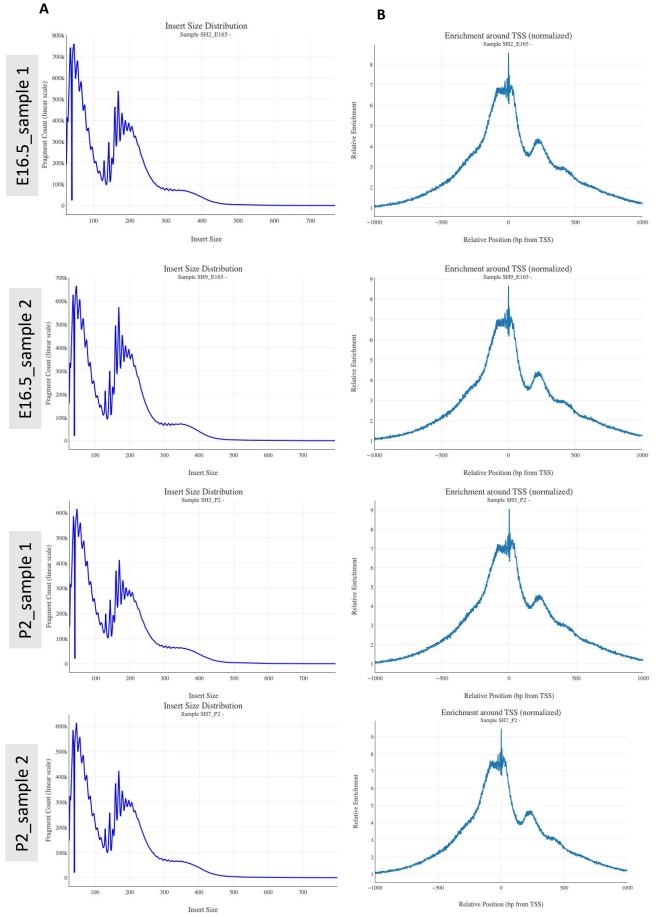
(**A**) Section IF for Six2 and Meis (1/2/3) factors in E14.5 kidney. In addition to expression in stroma, there is a low-level expression in Six2⁺ cells.

(B,C) UMAPs of E16.5/P2 multiome showing expression of *Meis2* in NPC clusters.

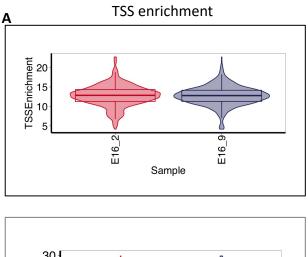
(**D**) scRNA derived UMAPs from published (GSE108291, GSE149134) and our unpublished (E14.5, P0) databases showing co-expression of *Meis2/Six2* especially prominent prior to P0.

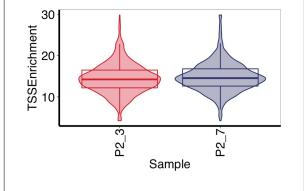
2

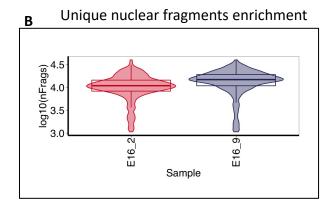
Quality control: Singleomes Six2^{GFP} cells - scATAC-seq

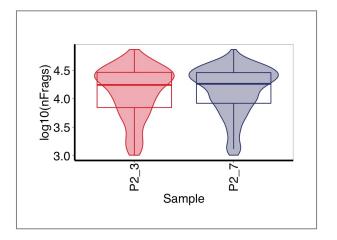


Singleomes: Six2^{GFP} scATAC - TSS and unique nuclear fragments enrichment

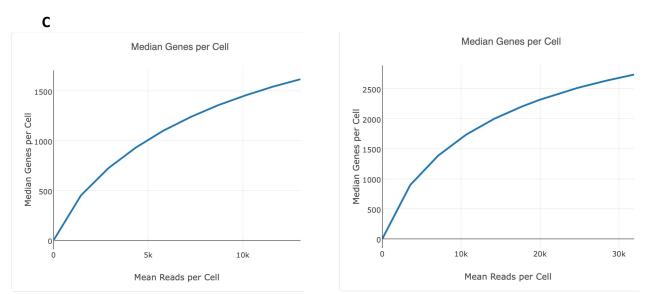








Singleomes: Six2^{GFP} scRNA-seq



Multiome: scATAC/GEX (E16.5/P2) QC metrics

В

scATAC-seq Α log10(Unique Fragments) vs TSS enrichment score 20 TSS Enrichment 10 3.5 4.0 Log10 Unique Fragments TSS enrichment score 30 25 TSSEnrichment 05 15 10 P2dual-E165dual Log10(unique nuclear fragments) 4.5 log10(nFrags) 6 3.5 P2dual-E165dual Fragment size TSS enrichment profiles distribution profiles per sample 15 Normalized Insertion Profile ^{group} - E165dual - P2dual 0.00

0.7

Percentage of Fragm

0.2

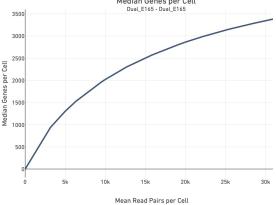
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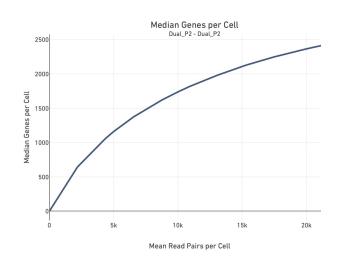
-200

-1000 0 1000 Distance From Center (bp)

2000

scRNA-seq Median Genes per Cell Dual_E165 - Dual_E165







Differential gene-specific chromatin accessibility

Fig. S4

