Supplementary Materials

Table S8. Peaks coordination and GREAT annotation for FOXC1 and FOXC2 ChIP-seq.

Table S9. Target genes annotated for FOXC1 or FOXC2 sites using Homer or Cistrom.

Table S10. Enrichment of podocyte-specific genes in FOXC1/2 target genes.

Table S11. Clinical characteristics of patient cohort used in Figure 5.

Table S12. Primers and probes.

Figure S1

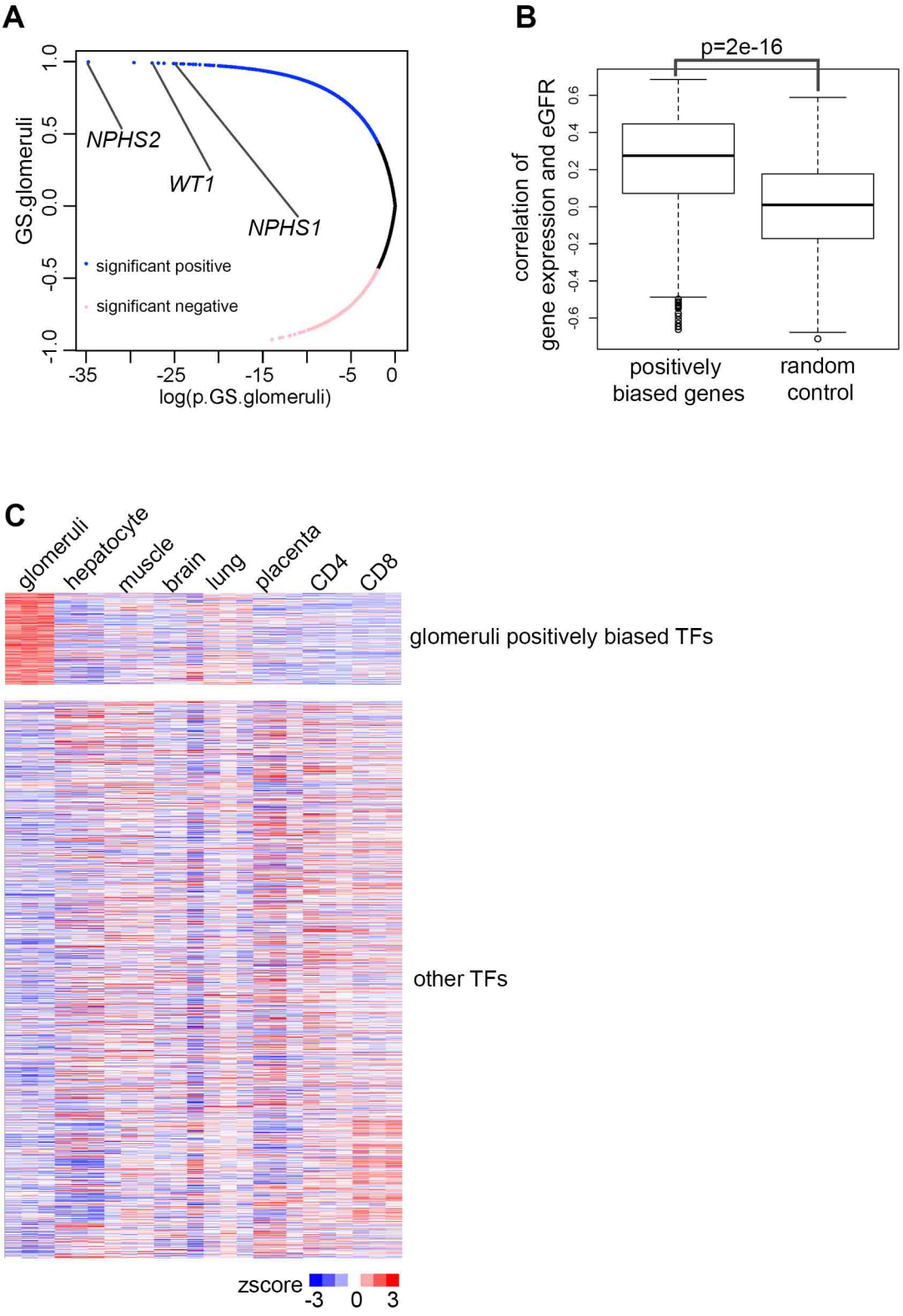
A
GS.glomeruli
log(p.GS.glomeruli)
NPHS2
WT1
NPHS1
significant positive
significant negative
B
p=2e-16
correlation of gene expression and eGFR
positively biased genes
random control
C
glomeruli
hepatocyte
muscle
brain
lung
placenta
CD4
CD8
glomeruli positively biased TFs
other TFs
zscore
-3
0
3

Figure S1. Glomeruli-biased expression in human. (A) Distribution of GS.glomeruli and p.GS.glomeruli of all genes. Podocyte markers as *NPHS1*, *NPHS2*, *WT1* are highlighted. Blue dots indicate glomeruli positively biased genes, and pink dots indicate glomeruli negatively biased genes. (B) Correlation between gene expression and eGFR are calculated for positive biased genes or randomly selected genes. (C) Heatmap of TF genes expressions. Glomeruli positivitely-biased TFs are those TF genes with WGCNA output GS.glomeruli>0 and p.GS.glomeruli<0.01.

Figure S2

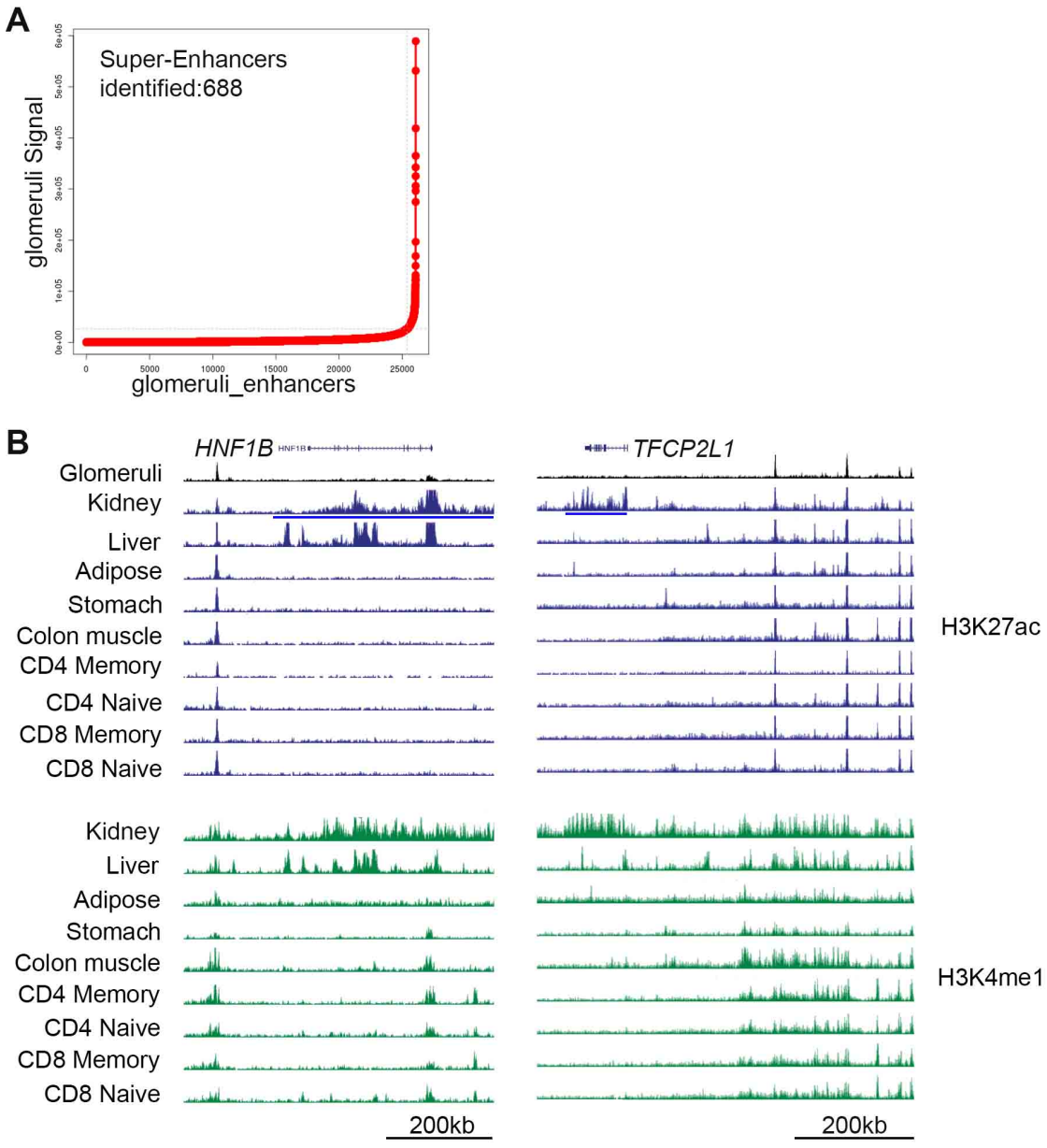

Figure S2. Super enhancer in adult human glomeruli. (A) Super enhancers called on H3K27ac in isolated human glomeruli. Grey line indicates the cutoff for accumulated H3K27ac signal intensity and rank determined by ROSE. Red dots in right up corner are called as super enhancers. (B) H3K27ac distribution at tubule-specific gene loci (*HNF1B* and *TFCP2L1*) in glomeruli, kidney and other tissue or cell types on UCSC genome

browser. Blue bars indicated SE called on H2K27ac from whole human kidney but not from human glomeruli.

Figure S3

Figure S3. The TFs in top1 CRC called using crcmapper. The regulatory relationship determined by crcmapper (left). The expression of these TF genes in human glomeruli and mice podocyte (right).

Figure S4

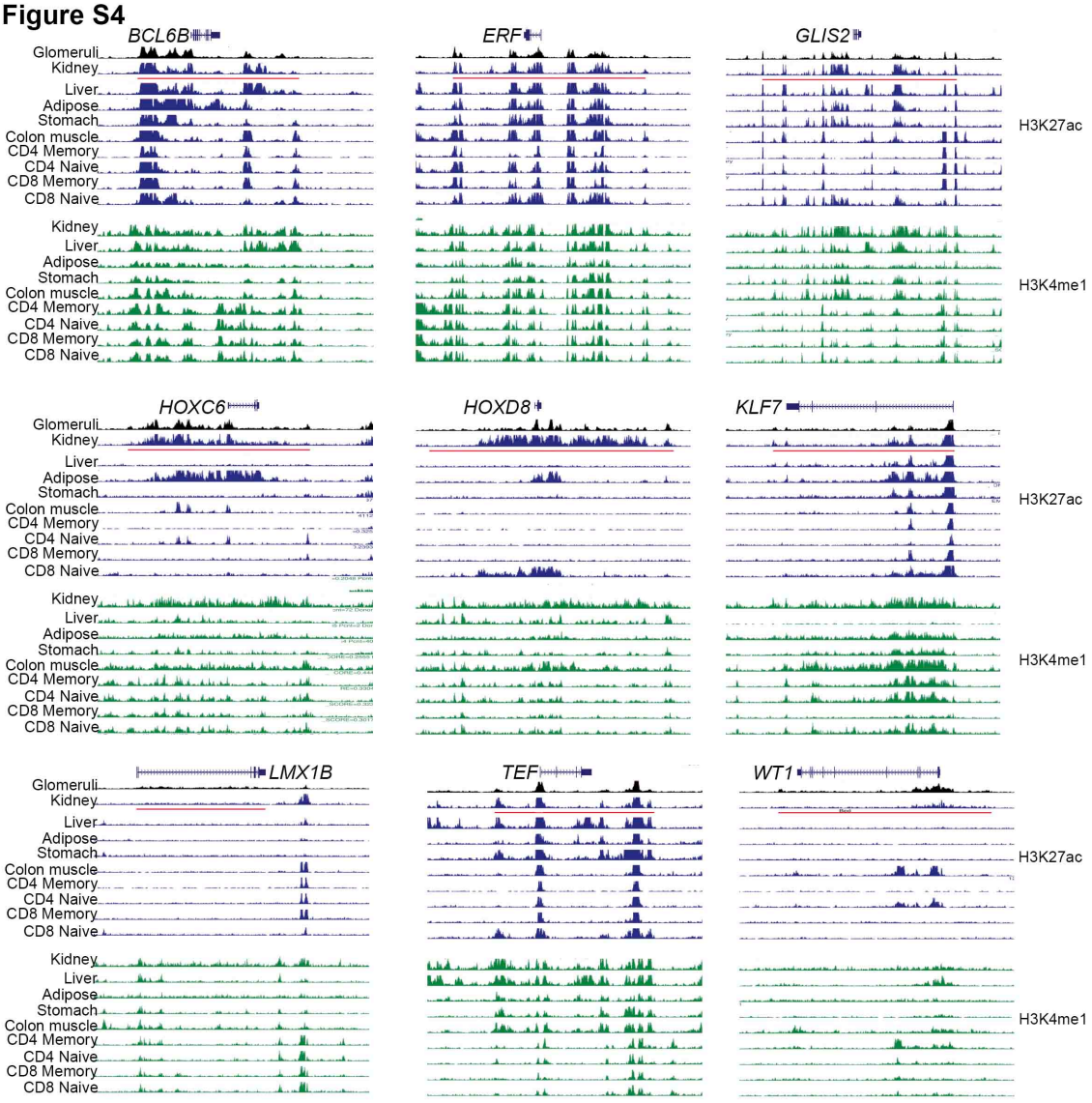

Figure S4. H3K27ac and H3K4me1 distribution at CRC TF gene loci except FOXC1 and FOXC2. Red bars indicate super enhancers called using ROSE on H3K27ac from human glomeruli. Blue tracks are H3K27ac signals and green tracks are H3K4me1 signals. The tissue types which are best matches of those used in WGCNA analysis are displayed.

Figure S5

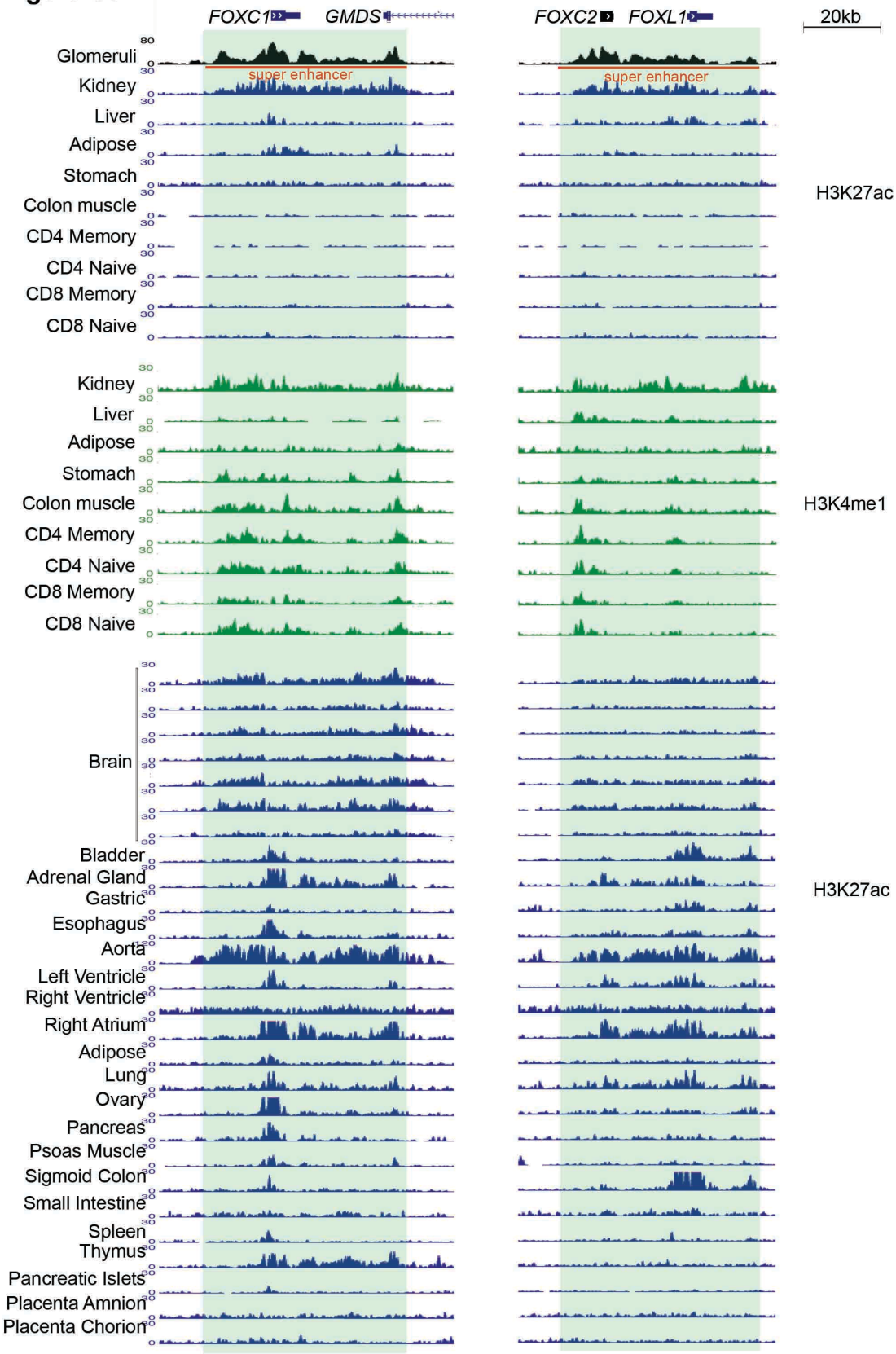
FOXC1
GMDS
FOXC2
FOXL1
20kb
super enhancer
super enhancer
Glomeruli
Kidney
Liver
Adipose
Stomach
Colon muscle
CD4 Memory
CD4 Naive
CD8 Memory
CD8 Naive
H3K27ac
Kidney
Liver
Adipose
Stomach
Colon muscle
CD4 Memory
CD4 Naive
CD8 Memory
CD8 Naive
H3K4me1
Brain
Bladder
Adrenal Gland
Gastric
Esophagus
Aorta
Left Ventricle
Right Ventricle
Right Atrium
Adipose
Lung
Ovary
Pancreas
Psoas Muscle
Sigmoid Colon
Small Intestine
Spleen
Thymus
Pancreatic Islets
Placenta Amnion
Placenta Chorion
H3K27ac

Figure S5. H3K27ac and H3K4me1 distribution at *FOXC1* and *FOXC2*. Red bars and green shades indicate super enhancers called using ROSE on H3K27ac from human glomeruli. Blue tracks are H3K27ac signals and green tracks are H3K4me1 signals. Tissue types that are best match for those used in WGCNA analysis are displayed on top for both H3K27ac and H3K4me1. All additional tissue types that are available in Roadmap are displayed for H3K27ac at bottom.

Figure S6

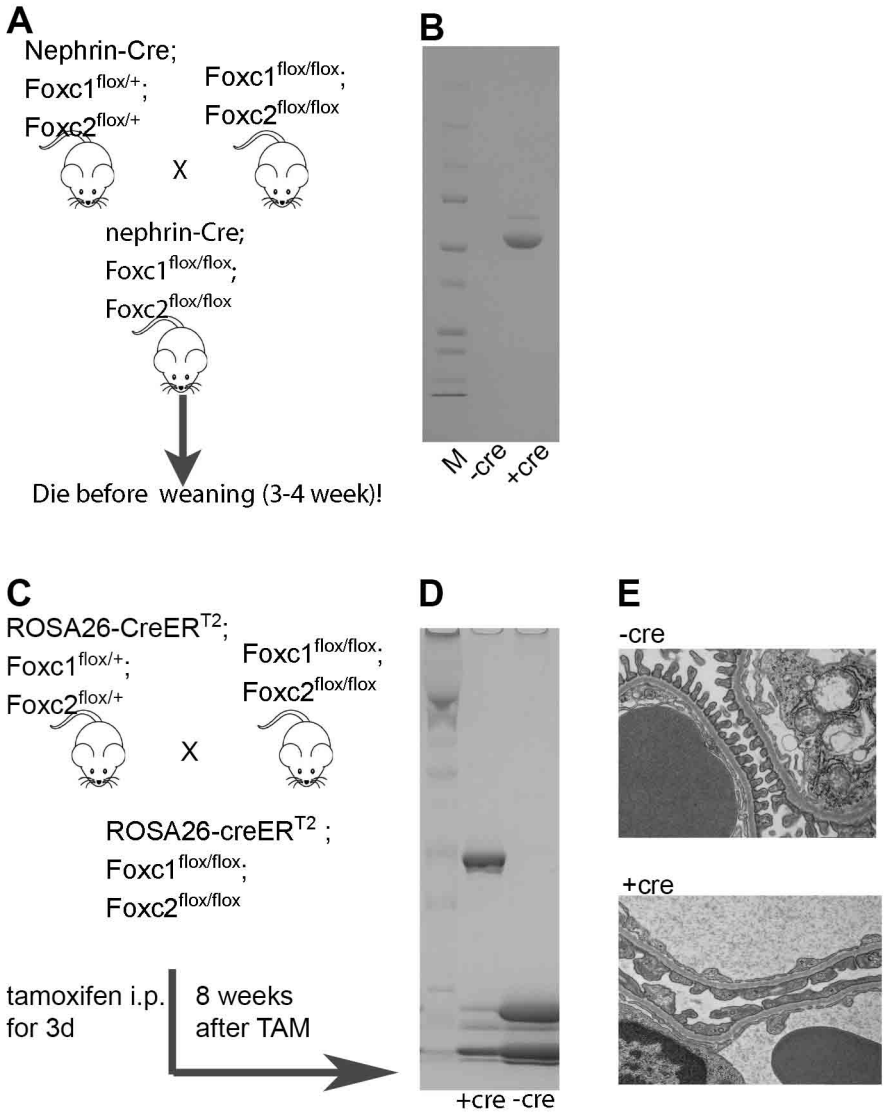

Figure S6. *Foxc1/2* KO in mice. (A) Scheme outline for podocyte specific KO of *Foxc1/2* under Nephrin-cre. (B) Electrophoresis of urea protein under Nephrin-cre mediated *Foxc1/2* KO. (C). Outline for conditional knockout of *Foxc1/2* in adult mice. (D). Electrophoresis of urea protein. (E). Electron microscope of podocyte focal foot process.

Figure S7

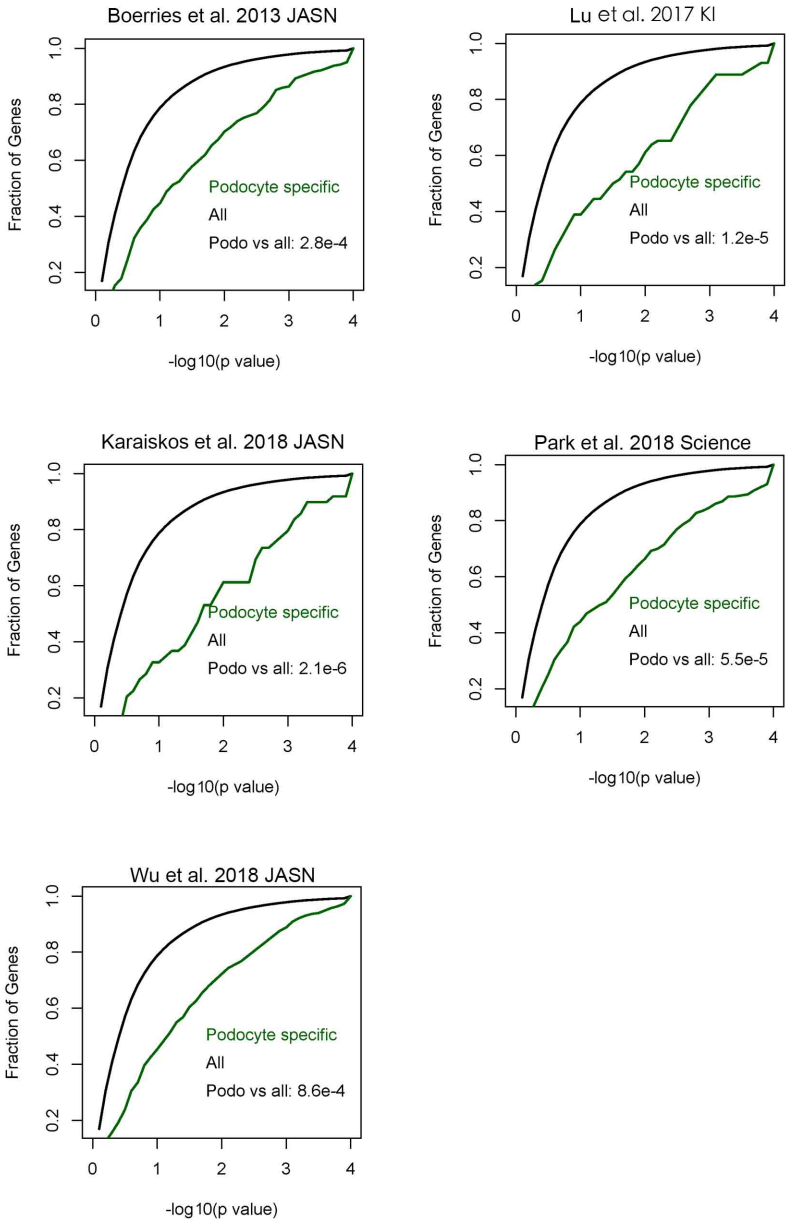
Boerries et al. 2013 JASN
Fraction of Genes
-log10(p value)
Podocyte specific
All
Podo vs all: 2.8e-4
Lu et al. 2017 KI
Podocyte specific
All
Podo vs all: 1.2e-5
Karaiskos et al. 2018 JASN
Podocyte specific
All
Podo vs all: 2.1e-6
Park et al. 2018 Science
Podocyte specific
All
Podo vs all: 5.5e-5
Wu et al. 2018 JASN
Podocyte specific
All
Podo vs all: 8.6e-4

Figure S7. Cumulative curve of –log10(p value) for differential expression analysis from Table S4. Podocyte specific are extracted from previous studies and listed in Table S5. t-test is performed for statistics.

Figure S8

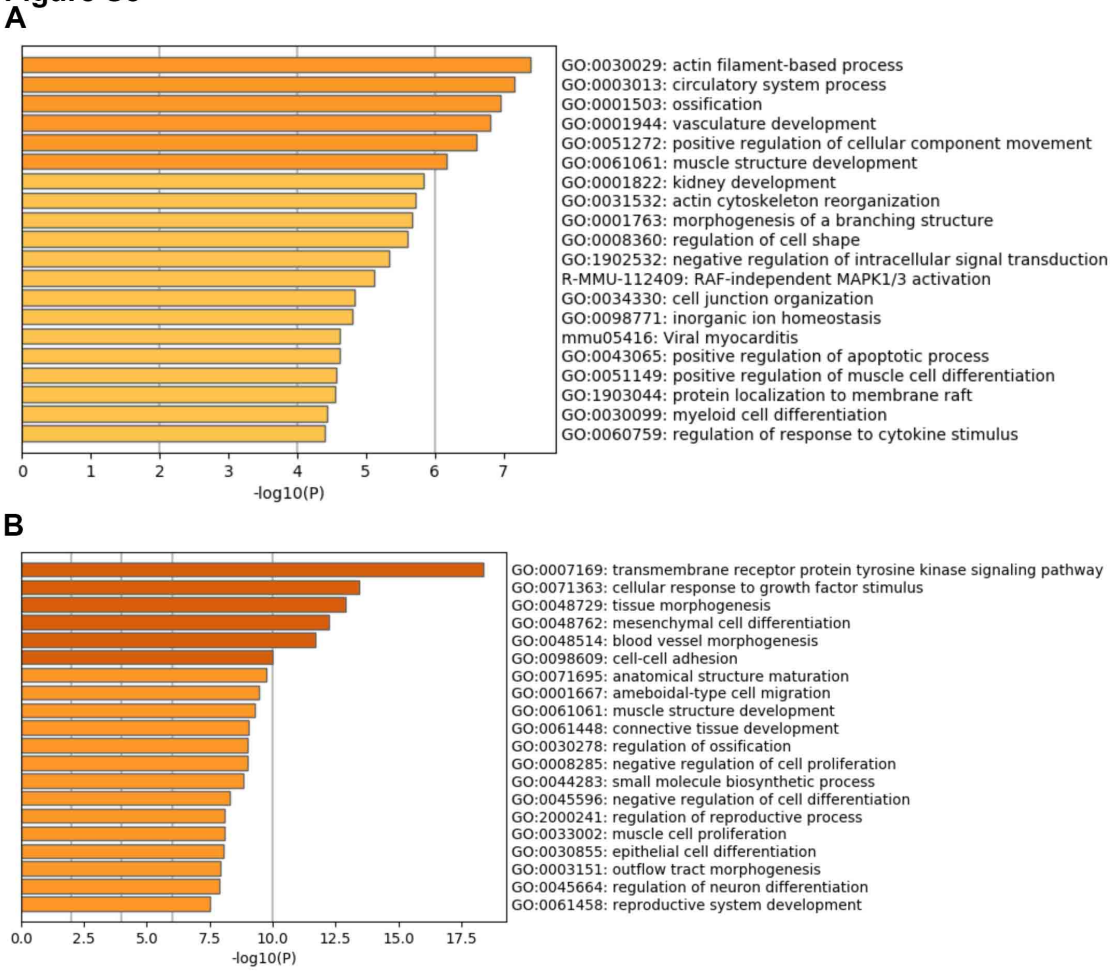

Figure S8. Gene ontology analysis of differential expressed genes in *Foxc1/2* cKO. (A) Top GO terms enriched for down-regulated genes. (B) Top GO terms enriched for up-regulated genes.

Figure S9

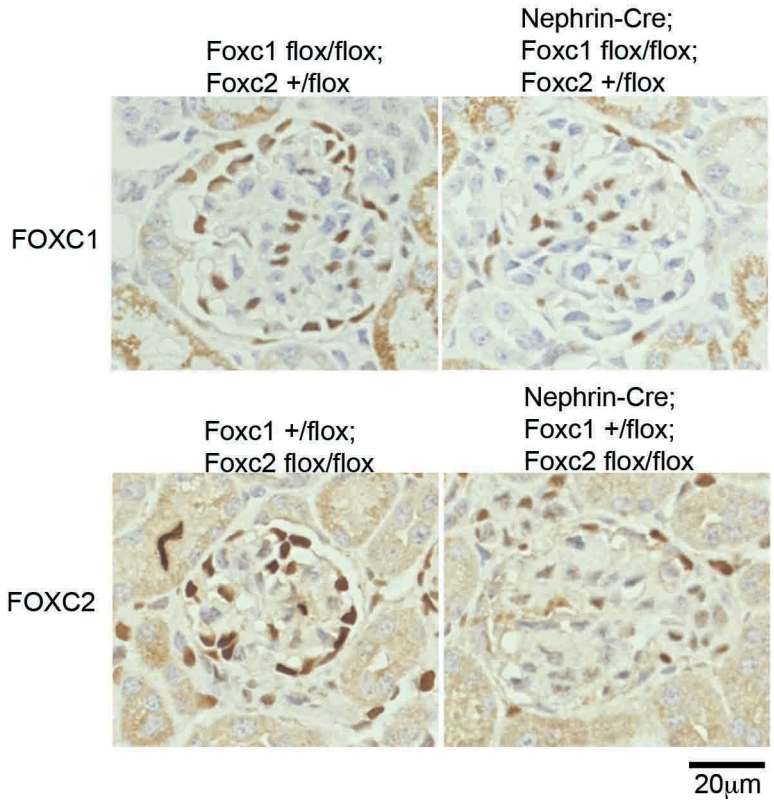

Figure S9. Validation of FOXC1 and FOXC2 antibodies.

Figure S10

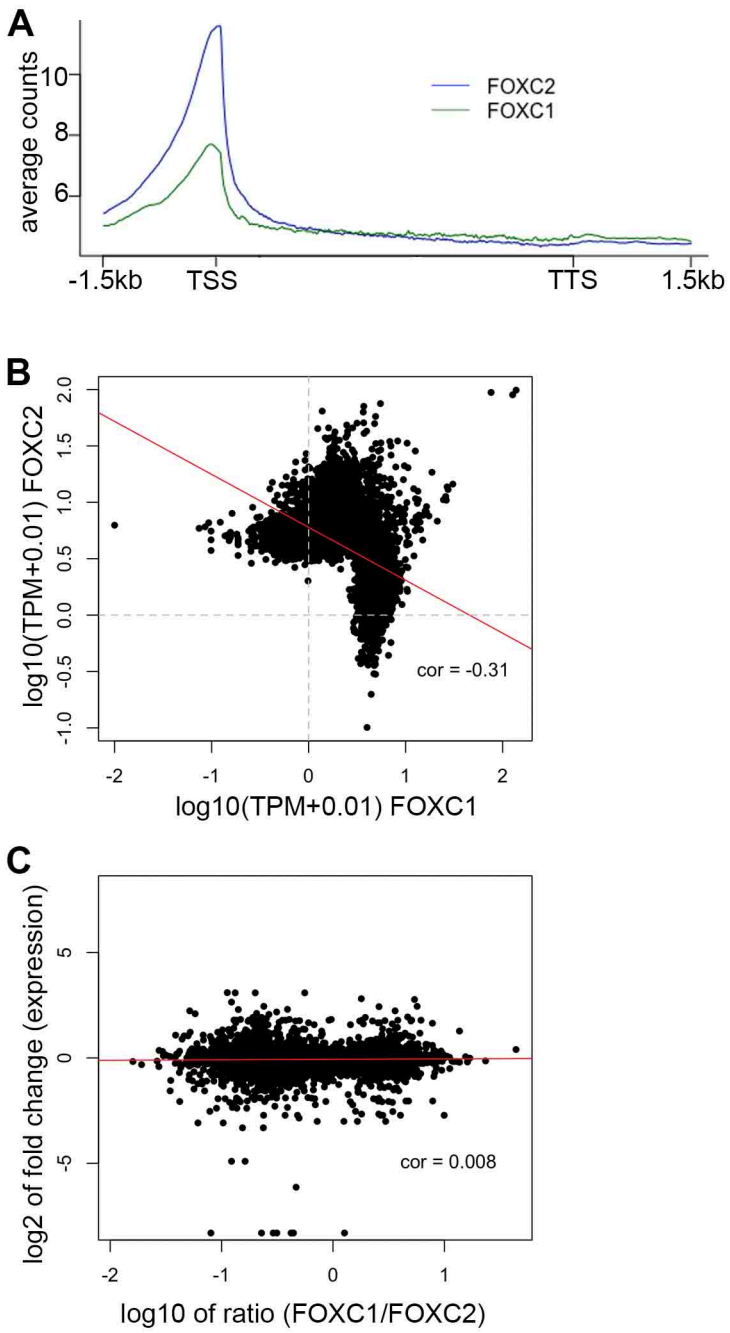

Figure S10. Features of FOXC1/2 binding. (A) Metagene plot of FOXC1 and FOXC2 ChIP-seq signal. (B) Scatter plot of FOXC1 verses FOXC2 signal at combinded FOXC1/2 binding sites. (C) Scatter plot of expression fold change from FOXC-associated genes verses FOXC1/FOXC2 signal ration of these FOXC sites.

Figure S11

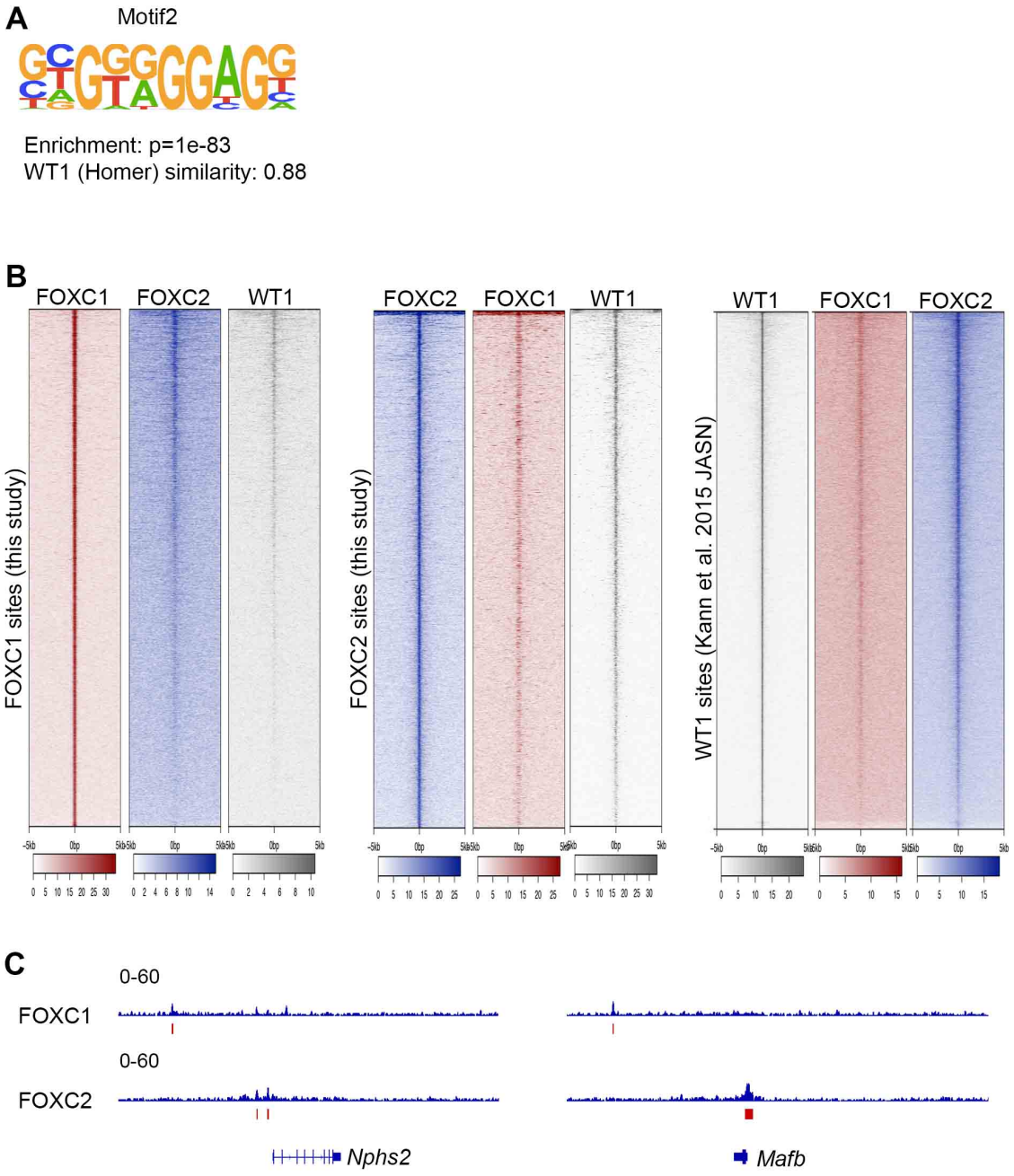

Figure S11. Distribution of FOXC1 and FOXC2 on genome. (A) Top 2nd motifs discovered at FOXC1/2 binding site. (B) Heatmap of signal from FOXC1, FOXC2 and WT1 ChIP-seq at FOXC1, FOXC2 or WT1 (Kann 2015) sites. (C) IGV tracks show

FOXC1 and FOXC2 distribution at *Nphs2* and *Mafb* loci. Red bars indicate the binding peaks.

Figure S12

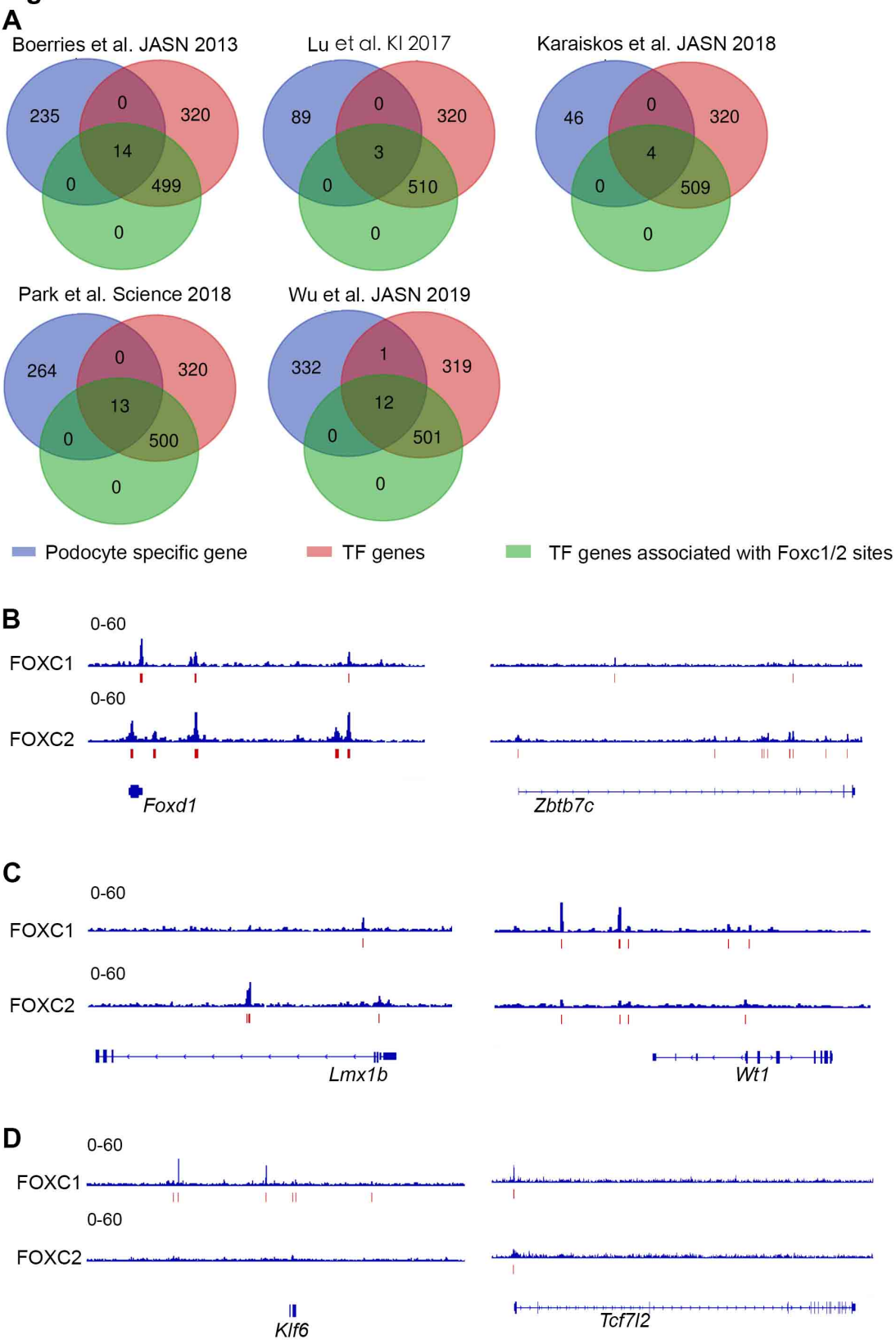
A
Boerries et al. JASN 2013
235
0
320
14
0
499
0
Lu et al. KI 2017
89
0
320
3
0
510
0
Karaiskos et al. JASN 2018
46
0
320
4
0
509
0
Park et al. Science 2018
264
0
320
13
0
500
0
Wu et al. JASN 2019
332
1
319
12
0
501
0
Podocyte specific gene
TF genes
TF genes associated with Foxc1/2 sites
B
0-60
FOXC1
0-60
FOXC2
Foxd1
Zbtb7c
C
0-60
FOXC1
0-60
FOXC2
Lmx1b
Wt1
D
0-60
FOXC1
0-60
FOXC2
Klf6
Tcf7l2

Figure S12. Target genes of FOXC1 and FOXC2 binding sites. (A) Venn diagram for overlapping among podocyte-specific genes (Table S5), TF genes in mouse genome, and FOXC1/2 target genes (annotated using Cistrom). (B) FOXC1 and FOXC2 binding at podoyte-specific TF genes (*Foxd1* and *Zbtb7c*). Red bars indicate the binding peaks. (C) FOXC1 and FOXC2 binding at other CRC members *Lmx1b* and *Wt1*. (D) FOXC1 and FOXC2 binding at reported TF genes important for podocyte homeostasis (*Klf6* and *Tcf7l2*).

Figure S13

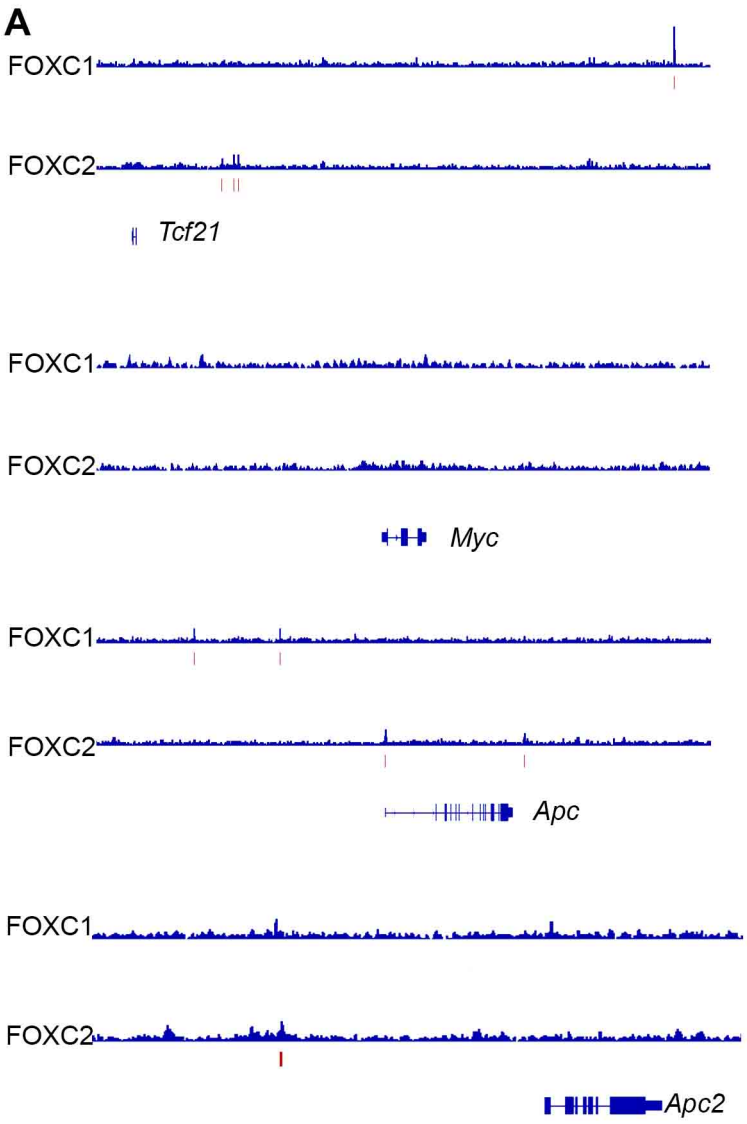
A
FOXC1
FOXC2
Tcf21
FOXC1
FOXC2
Myc
FOXC1
FOXC2
Apc
FOXC1
FOXC2
Apc2
B
Apc
relative expression
1.5
1
0.5
0
CTL
TAM
Apc2
*
relative expression
1.2
1
0.8
0.6
0.4
0.2
0
CTL
TAM

Figure S13. FOXC1 and FOXC2 distribution at *Tcf21*, *Myc*, *Apc* and *Apc2*. (A) FOXC1 and FOXC2 signal at *Tcf21*, *Myc*, *Apc* and *Apc2*. Red bars indicate the binding peaks. (B) Relative expression of *Apc* and *Apc2* mRNA. * stands for $p<0.05$.

Figure S14

Figure S14. Conservation of Foxc in human, mice and zebrafish. Red box highlights forkhead domain. Stars highlight DNA binding sites.