Cell Reports, Volume 42

## **Supplemental information**

## Jarid2 promotes temporal progression

## of retinal progenitors via repression of *Foxp1*

Jianmin Zhang, Jacqueline M. Roberts, Fei Chang, Joon Schwakopf, and Monica L. Vetter



## Figure S1: Jarid2 is conditionally deleted in the retina, related to STAR Methods.

(A-D) *Jarid2* expression by *in situ* hybridization (ISH) on E14.5 and P0 mouse retinal sections. (E) Schematic diagram of the generation of *Jarid2* conditional knockout allele. (F) RT-PCR of whole retina from *Six3-Cre Jarid2* cKO at P0. Forward and reverse primers used are marked in (E). Arrow indicates the *Jarid2* cKO band cut for DNA sequencing. Upper band is *Jarid2* floxed allele that remains due to mosaic activity of Six3-Cre in whole retina. (G) Western blot for Jarid2 and GAPDH (as loading control) in the whole retina from *Six3-Cre Jarid2* cKO and control littermates at P0. NBL: neuroblastic layer; GCL: ganglion cell layer. Scale bar: 100µm.





At P0/P3 *Six3-Cre Jarid2* cKO retina shows (A, B, G) increased Brn3+ RGCs within GCL (bracket), (C, D, H) increased Calbindin+ amacrine cells (white bracket) and horizontal cells

(yellow bracket) and (E, F, I) increased Rxry+ cone photoreceptors (bracket). At E16.5 there was no change in (J, K, P) Brn3-labeled RGCs, (L, M, Q) Otx2-labeled photoreceptor precursors (bracket), or (N, O, R) Rxry labeled cone photoreceptors (bracket) in *Six3-Cre Jarid2* cKO retina. At P14 (S, T, Y) AP2 $\alpha$ + amacrine cells, (U, V, Z) Brn3a+ RGCs, and (W, X, AA) Rxry+ cones remain elevated in the *Six3-Cre Jarid2* cKO. Graphs represent mean ± SEM. n = 6 retinas in (G, Y, Z, AA) and *Jarid2* cKO in (Q, R). n=4 in (H, I, P) and control in (Q, R). ns, not significant; \**p*<0.05; \*\**p*<0.01 by Student's T test. Scale bar: 100 µm. See also Table S6.





(A) Schematic diagram of the experimental design. EdU labeled (B, C, H) Brn3a+ RGCs, (D, E, I) AP2 $\alpha$ + amacrine cells, and (F, G, J) Rxr $\gamma$ + cone photoreceptors are increased in *Rax-Cre Jarid2* cKO retina. Arrowheads indicate co-labeled cells. Graphs represent mean ± SEM. n = 3 retinas in (I, J) and control in (H). n = 4 for Jarid2 cKO in (H) \*\*p<0.01, \*\*\*p<0.001, \*\*\*\*p<0.0001 by Student's T test. Scale bar: 50 µm. See also Table S6.



Figure S4: Dimensional reduction distinguishes retinal cell types, and not batch effects,

related to Figure 3

(A) UMAP dimensional reduction of scRNA-seq results for whole retina, colored by relative gene expression, distinguishes RPCs (*Ccnd1*, *Vsx2*) and neurogenic cells (*Neurog2*, *Atoh7*). (B) UMAP colored by retinal cell type. (C) UMAP colored, or separated, by experiment batch. (D) UMAP colored, or separated, by genotype.



Figure S5: scRNA-seq Expression of Early and Late RPC Genes, related to Figures 3, 4, 5

and 7

Genes cited in this study as having early or late gene expression are plotted. At each age, E11-P5, the proportion of RPCs expressing the gene and average gene expression are represented by circle size and color respectively. This data is represented both scaled (z-score across ages) and unscaled. scRNA-seq data is from Clark et al.<sup>11</sup>.



Figure S6: *Foxp1* is Regulated by Histone Methylation During Retinal Development, related to Figure 4

Genome tracks for log2 fold enriched coverage, compared to IgG, over gene loci of interest. Negative log2FE values are not shown. *Foxp1* (A, B), *Pcdh7* (C), *Fign* (D), *Efna5* (E), and *Reln* (F) are all associated with reduced H3K27me3 peaks in *Jarid2* cKO and are expressed at higher levels in *Jarid2* cKO RPCs. (A, C-F) H3K27me3 CUT&RUN tracks for control (orange) and *Jarid2* cKO (purple) retinas at E18.5. Aggregated peaks from these datasets are indicated by black bars. Peaks significantly reduced in *Jarid2* cKO are marked with a red asterisk. H3K27me3 CUT&RUN tracks for wild type whole retina at E14 and P0 are in red. (B) H3K27me3 ChIP-seq tracks for whole retina at E17, P0, P3 (raw data from Aldiri et al.<sup>29</sup>). Red asterisks show peaks that are increased across development in a pairwise comparison between any two timepoints. Significance is q-value <= 0.05. Scale bar: 50 kb.



**Figure S7: Retinal neurogenesis is affected in** *Foxp1* **cTG**, **related to Figure 6** Foxp1 immunostaining on retinal cross sections from P0 *Foxp1* cTG and control littermates confirms that (A-B) Foxp1 expression is increased in *Foxp1* cTG. (C-E) Foxp1 and Brn3a immunostaining shows mosaic levels of *Foxp1* expression, with higher levels coinciding with increased Brn3a labeled cells. White dash lines demarcate higher Foxp1 expression region. (F-H) Rosette structures are visible in the outer nuclear layer in a subset of *Foxp1* cTG retinal sections. ONL: outer nuclear layer. Scale bar: 100 μm.