

**Neuron, Volume 94**

**Supplemental Information**

**Homeostatic Plasticity Shapes**

**Cell-Type-Specific Wiring in the Retina**

**Nai-Wen Tien, Florentina Soto, and Daniel Kerschensteiner**

## **SUPPLEMENTARY INFORMATION**

### **Homeostatic plasticity shapes cell-type-specific wiring in the retina**

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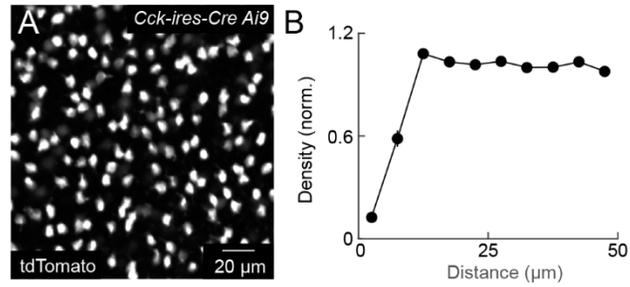
**Figure S1** (related to Figure 1)

**Figure S2** (related to Figure 1)

**Figure S3** (related to Figure 2)

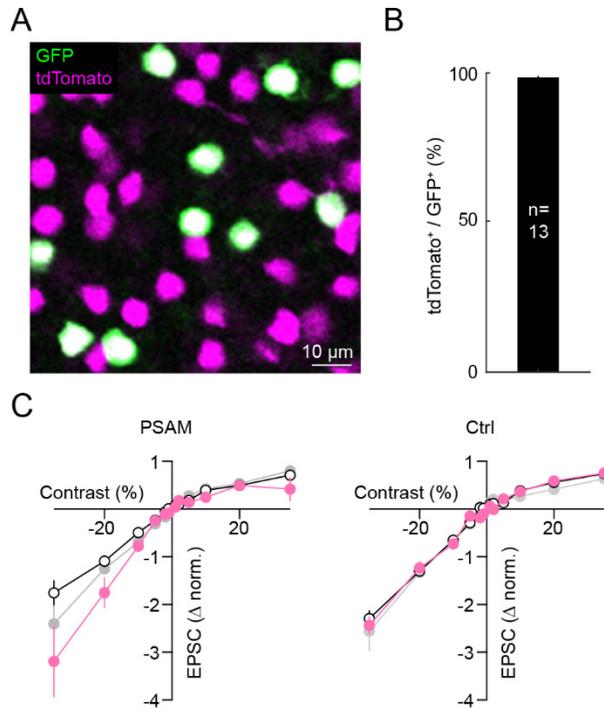
**Figure S4** (related to Figures 3, 4 and 5)

**Figure S5** (related to Figures 6 and 7)



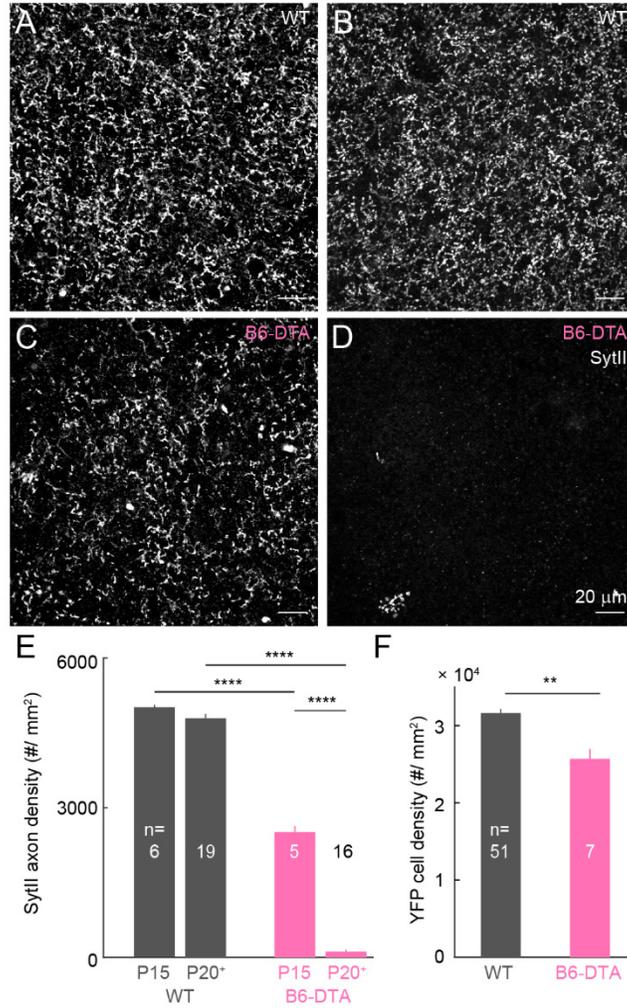
**Figure S1. Distribution of tdTomato-expressing bipolar cells in *Cck-ires-Cre Ai9*** (related to Figure 1)

(A) Representative confocal image of tdTomato-expressing cells in the bipolar cell body layer of a P21 whole-mount *Cck-ires-Cre Ai9* retina. (B) Summary data of the density recovery profiles of tdTomato-expressing cells in the bipolar cell body layer (n = 5 retinas) show an exclusion zone characteristic of labeling of a single cell type.



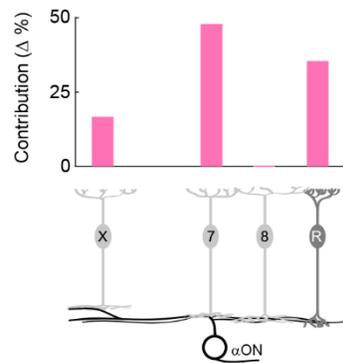
**Figure S2. Pharmacogenetic silencing of B6 cells changes contrast response functions of ON $\alpha$ -RGCs (related to Figure 1)**

(A) Representative image of the INL in a *CCK-ires-Cre Ai9* mouse injected with *AAV-Grm6<sub>s</sub>-GFP*. (B) 98.5%  $\pm$  0.5 of GFP-positive cells in the INL were also tdTomato positive. (C) Contrast response functions of the normalized excitation of ON $\alpha$ -RGCs with (*PSAM*, n = 10) or without (*Ctrl*, n = 9) *AAV-Grm6<sub>s</sub>-PSAM<sup>con</sup>* injection before (*Before*, *black*), during (*PSEM*, *pink*) and after (*Wash*, *grey*) the addition of PSEM<sup>308</sup>. Differences between responses before and during PSEM<sup>308</sup> application in the *AAV-Grm6<sub>s</sub>-PSAM<sup>con</sup>* injected retinas were significantly different (95% confidence interval, see STAR Methods).



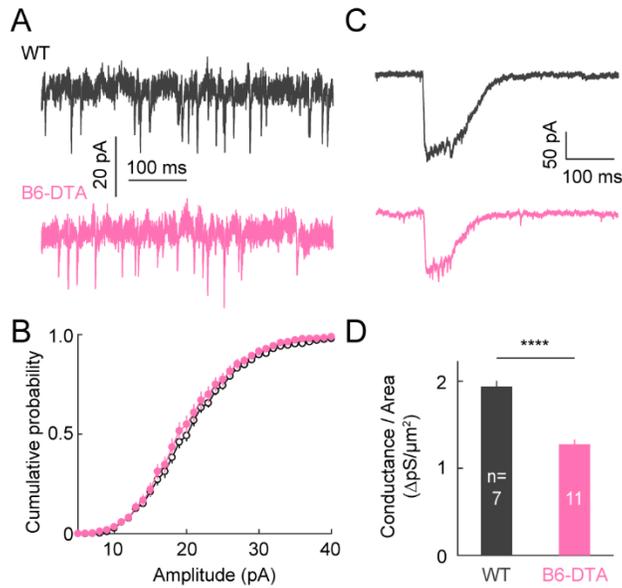
**Figure S3. Cell-type-specific removal of B6 cells in *B6-DTA* mice** (related to Figure 2)

(A-D) Representative images of SytII staining in the ON sublamina of the inner plexiform layer in P15 (A), P20<sup>+</sup> (i.e. P20 – P30) (B) wild-type and P15 (C) and P20<sup>+</sup> (D) *B6-DTA* retinas. (E) Summary data of the density of SytII-positive axons in the ON sublamina in wild-type and *B6-DTA* retinas during development. In (E), *P15 WT*: 5015 ± 50 # / mm<sup>2</sup>; *P20<sup>+</sup> WT*: 4792 ± 89 # / mm<sup>2</sup>; *P15 B6-DTA*: 2511 ± 124 # / mm<sup>2</sup>; *P20<sup>+</sup> B6-DTA*: 115 ± 40 # / mm<sup>2</sup>; *P15 WT* vs *P20<sup>+</sup> WT* p = 0.48; *P15 WT* vs *P15 B6-DTA* p = 5 \* 10<sup>-6</sup>; *P20<sup>+</sup> WT* vs *P20<sup>+</sup> B6-DTA* p = 8 \* 10<sup>-26</sup>; *P15 B6-DTA* vs *P20<sup>+</sup> B6-DTA* p = 10<sup>-5</sup>. (F) Summary data of the density YFP-expressing cells in wild-type (31624 ± 548 # / mm<sup>2</sup>) and *B6-DTA* (25699 ± 1269 # / mm<sup>2</sup>, p = 0.002) retinas.



**Figure S4. Bipolar cell types rewire with ON $\alpha$ -RGC in cell-type-specific ratios in *B6-DTA* mice** (related to Figures 3, 4 and 5)

In *B6-DTA* mice, the number of excitatory synapses numbers on ON $\alpha$ -RGCs is reduced by 29.5 % compared to wild-type littermates. Given that B6 cells were shown to account for 70 % of the excitatory synapses on ON $\alpha$ -RGCs in wild-type retinas (Schwartz et al., 2012), it follows that non-B6 bipolar cells restore 40.5 % of the lost synapses in *B6-DTA* retinas. We calculated the approximate convergence of each bipolar cell type to ON $\alpha$ -RGCs by dividing the dendritic territory of the later by the axonal territory of the former. We then multiplied the convergence factor by the change in synapses numbers between pairs of individual bipolar cells and ON $\alpha$ -RGCs, to estimate the relative contribution each bipolar cell type to rewiring (XBC 17%, B7 47.4 %, RBC 35.6 %).



**Figure S5. Postsynaptic strength in ON $\alpha$ -RGCs of *B6-DTA* mice** (related to Figures 6 and 7) (A and B) Representative traces (A) and cumulative distribution curves (B) of sEPSCs recorded from ON $\alpha$ -RGCs in wild-type (n = 10) and *B6-DTA* (n = 5) retinas. (C and D) Representative traces (C) and summary data (D) of currents elicited by glutamate puffs in ON $\alpha$ -RGCs in wild-type (1.9  $\pm$  0.07 pS /  $\mu$ m<sup>2</sup>) and *B6-DTA* (1.3  $\pm$  0.05 pS /  $\mu$ m<sup>2</sup>, p = 2 \*10<sup>-13</sup>) retinas. The area of each application was estimated by 2-photon imaging of a fluorescent dye (Alexa 488) included in the puff solution. Extrasynaptic glutamate receptors may contribute to the currents observed in puff experiments.