SUPPLEMENTARY MATERIAL

Visual Map Development Depends On The Temporal Pattern of Binocular Activity in Mice

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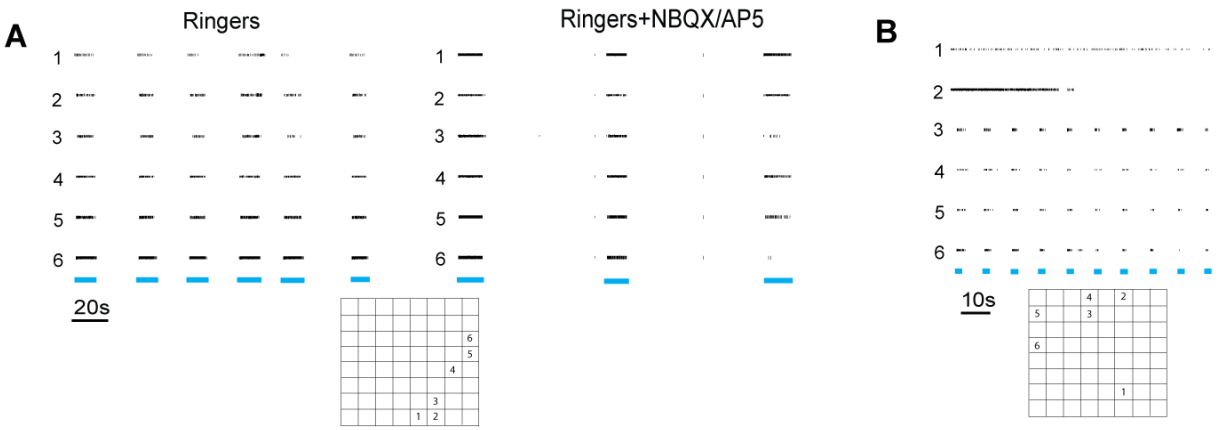
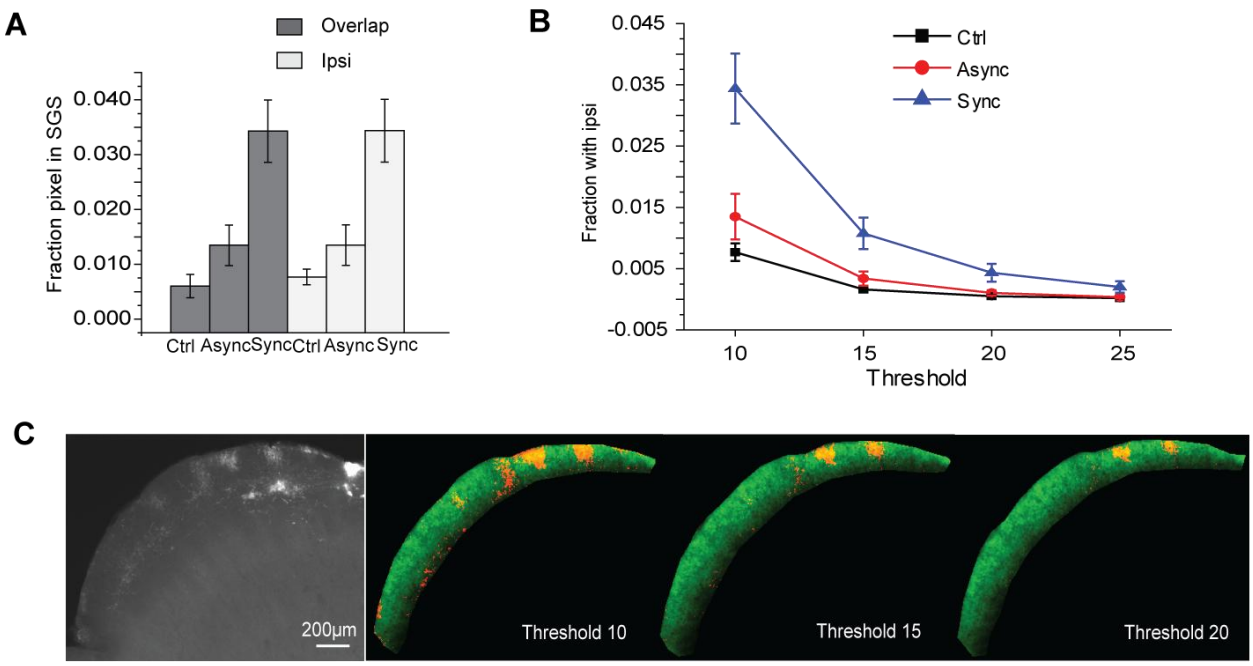
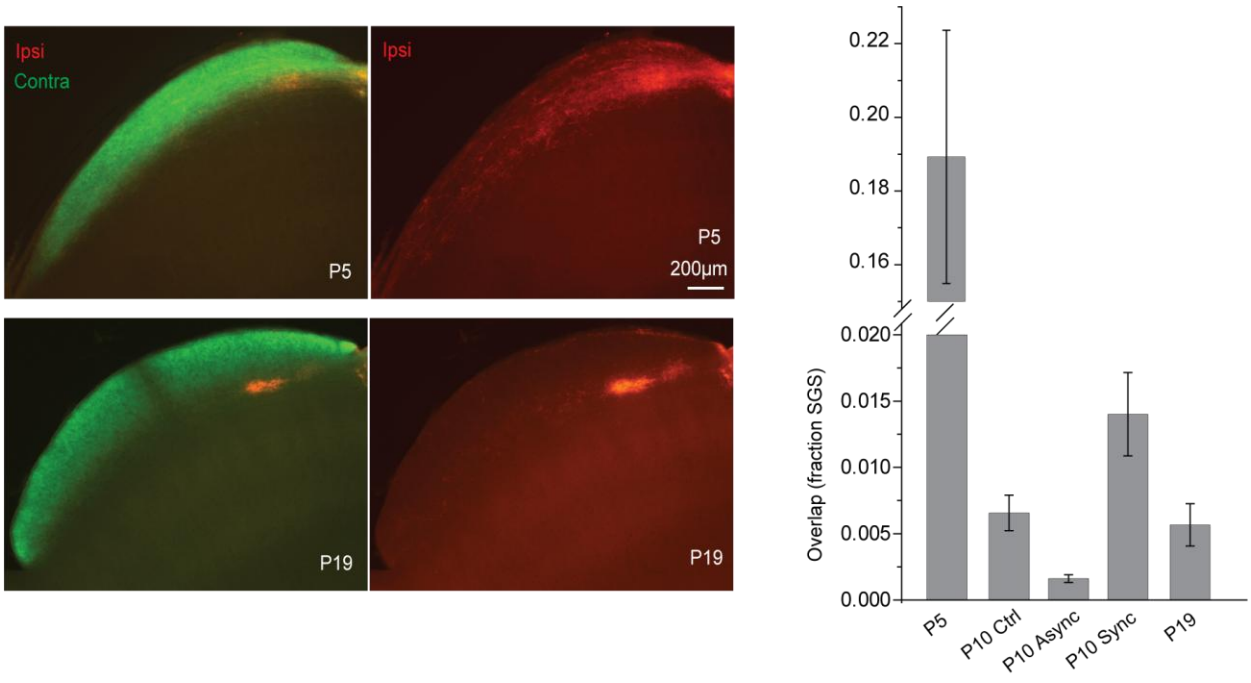
SUPPLEMENTARY FIGURES



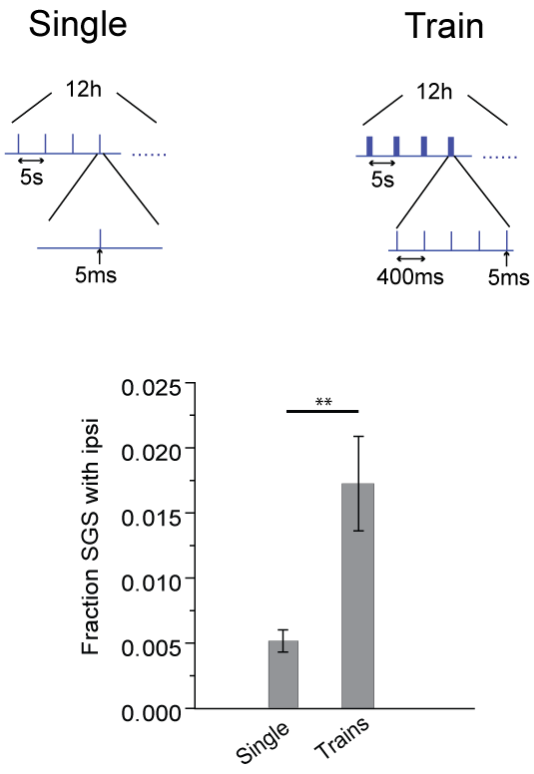
Supplementary Figure 1. Light–driven activity of RGCs is not dependent on synaptic input. (A) Left panel: Raster plot of spiking responses to light in a *Thy1*–ChR2 retina under normal *in vitro* recording conditions in Ringer's solution with an MEA; Right panel: Raster plot of spiking responses to light from the same retina after application of a mixture of glutamate receptor antagonists NBQX (10 µM) and AP5 (50 µM). Blue bars represent light stimuli. (B) Channel 1 and 2 had sustained spiking responses after the onset of the first light pulse reminiscent of ipRGC–like activity; all the other channels had responses that were precisely aligned with the light stimuli.



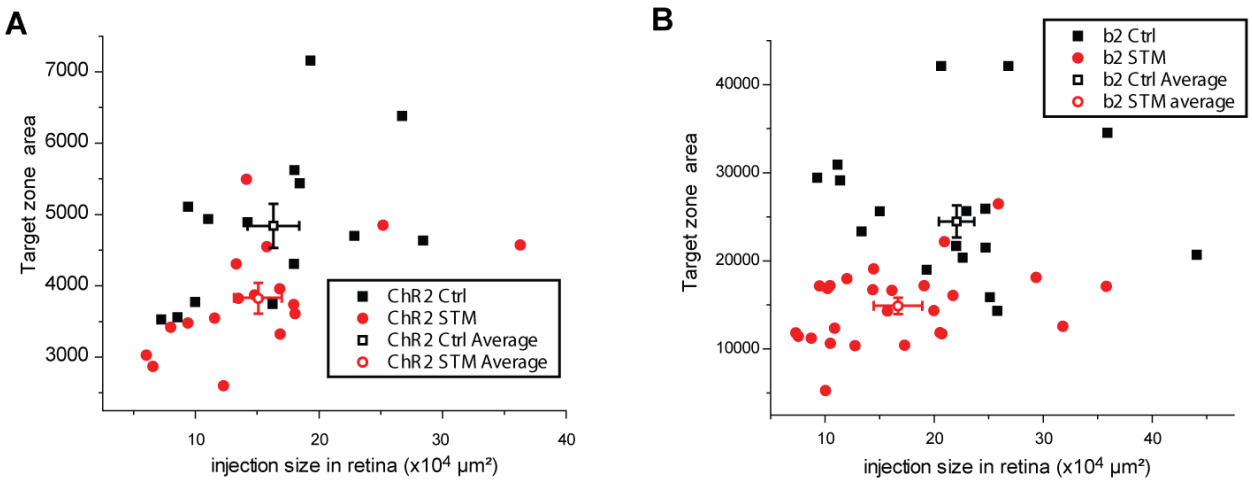
Supplementary Figure 2. Quantification of eye–segregation in the superior colliculus. (A) Quantification of synchronous and asynchronous stimulation phenotype in the superior colliculus using two different measures. Left (dark grey): fraction of pixels in the SGS with overlapping contralateral (green) and ipsilateral (red) fluorescent signal; Right (light grey): fraction of the SGS with ipsi (red) pixels. Both measures give similar results. (B) Quantification of synchronous and asynchronous stimulation phenotype in the superior colliculus at different fluorescent thresholds. (C) Illustration of superior colliculus ipsilateral image processing with different thresholds. Green color is the contralateral (SGS) domain; orange color is the thresholded ipsilateral (red) pixels overlayed on the (green) contralateral image. Threshold 10 captures the cluster phenotype the best. Error bars represent s.e.m..



Supplementary Figure 3. Eye–specific segregation emerges between P5 and P9. At P5, ipsilateral (red) and contralateral (green) axons are largely overlapping in the superior colliculus. Eye–specific segregation is dramatically better at P10 (P10 Ctrl, unstimulated AAV–ChR2 treated Rx–Cre mice), and does not further improve at P19. Asynchronous (P10 Async) stimulation improves eye segregation, whereas synchronous (P10 Sync) stimulation inhibits eye segregation. Error bars represent s.e.m..



Supplementary Figure 4. Burst stimuli disrupt segregation much more than single stimuli. Synchronous, single 5 msec pulses at 0.2 Hz had no effect on eye–specific segregation in the superior colliculusof *Thy1*–ChrR2 mice, but bursts of synchronous 5 msec pulses at the same frequency significantly disrupted segregation. Error bars represent s.e.m.. ** p <0 .005.



Supplementary Figure 5. Target zone is independent of the size of DiI injections in the retina. (A) Dorsal and ventral–temporal injections were grouped together to make stimulated (ChR2 STM) and control (ChR2 Ctrl) data for *Thy1*–ChR2 animals. The injection sizes of the DiI labeled retina were plotted against the corresponding target zone size in the superior colliculus. The average of injection sizes in ChR2 Ctrl mice (red open circle) is not significantly different from ChR2 STM mice (black open circle), but the size of the target zone in the superior colliculus in ChR2 Ctrl mice is significantly larger than ChR2 STM. R^2 is 0.200 for ChR2 Ctrl and 0.253 for ChR2 STM. (B) Dorsal and ventral–temporal injections were grouped together to make stimulated (ChR2;$\beta2^{-/-}$ STM) and control (ChR2;$\beta2^{-/-}$ Ctrl) data for ChR2;$\beta2^{-/-}$ animals. The average of injection sizes in ChR2;$\beta2^{-/-}$ Ctrl (red open circle) is not significantly different from ChR2;$\beta2^{-/-}$ STM (black open circle), but the size of the target zone in the superior colliculus in ChR2;$\beta2^{-/-}$ Ctrl mice is significantly larger than ChR2;$\beta2^{-/-}$ STM mice. R^2 is 0.061 for ChR2;$\beta2^{-/-}$ Ctrl and 0.102 for ChR2;$\beta2^{-/-}$ STM.