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Sensory modality–specific homeostatic plasticity in the developing optic tectum

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We found a previously unknown form of homeostatic synaptic plasticity in multisensory neurons in the optic tectum of *Xenopus laevis* tadpoles. Individual tectal neurons are known to receive converging inputs from multiple sensory modalities. We observed that long-term alterations in either visual or mechanosensory activity *in vivo* resulted in homeostatic changes specific to each sensory modality. In contrast with typical forms of homeostatic synaptic plasticity, such as synaptic scaling, we found that this type of plasticity occurred in a pathway-specific manner that is more reminiscent of Hebbian-type plasticity.

Homeostatic synaptic plasticity is a type of plasticity in which synaptic strength is uniformly adjusted throughout a neuron to compensate for long-term changes in neural activity. Typically, homeostatic plasticity is global and multiplicative, such that all synapses in a cell are either increased or decreased by the same fraction, preserving the relative weights between synapses and optimizing the neuron's dynamic range[1]. Experimentally, homeostatic synaptic plasticity is typically studied by altering global levels of neural activity over several days. For example, experimentally decreasing neural activity for 48 h in cultured hippocampal neurons can lead to the strengthening of excitatory synapses, whereas chronically increasing activity can result in scaling down[2]. Similarly, *in vivo* sensory manipulations that increase or decrease activity have been used to induce scaling[3,4]. However, it is not clear whether selective long-term changes to a select subset of synaptic inputs to a neuron could result in a local form of homeostatic plasticity specific to these inputs. Here we describe a previously unknown, pathway-specific form of homeostatic synaptic plasticity in the optic tectum of *Xenopus laevis* tadpoles.

The *Xenopus* tadpole optic tectum is a multisensory area in which multiple sensory modalities converge onto individual neurons throughout development[5,6]. Using an *in vitro* whole-brain preparation, we can electrically stimulate separate pathways conveying visual and mechanosensory input[5]. We performed a series of *in vivo* sensory manipulations over the course of 48 h and then examined whether they resulted in homeostatic plasticity specific to the manipulated pathway.

We performed whole-cell recordings from 83 optic tectal neurons using isolated whole-brain preparations from stage 49 tadpoles, as described previously[7] (see **Supplementary Methods**). We first tested whether glutamatergic synapses in the optic tectum could express conventional forms of homeostatic plasticity[1] by exposing tadpoles for 48 h to the AMPA receptor antagonist 6-nitro-2,3-dioxo-1,4-dihydrobenzo[f]quinoxaline-7-sulfonamide (NBQX, 30 μM) or $GABA_A$ receptor blocker picrotoxin (PTX, 100 μM). Decreasing neural activity with NBQX resulted in a significant increase in spontaneous excitatory postsynaptic current (sEPSC) amplitudes, whereas increasing activity with PTX resulted in a significant decrease in sEPSC amplitude (control, 5.33 ± 0.53 pA, $n = 7$; NBQX, 7.24 ± 0.37 pA, $n = 8$, $P < 0.01$; PTX, 3.9 ± 0.22 pA, $n = 8$, $P < 0.05$; **Supplementary Fig. 1**). Neither manipulation resulted in changes in sEPSC frequency. These data are consistent with global homeostatic plasticity being present in this system.

To assess synaptic strength at both types of sensory inputs independently, we placed a stimulating electrode on the optic chiasm to activate visual inputs and another in the contralateral hindbrain to activate mechanosensory inputs[5] (**Fig. 1a**). We found that most tectal neurons received converging monosynaptic input from both pathways[5]. To measure quantal amplitude of a given synaptic input, we replaced extracellular Ca^{2+} with Sr^{2+}, resulting in asynchronous

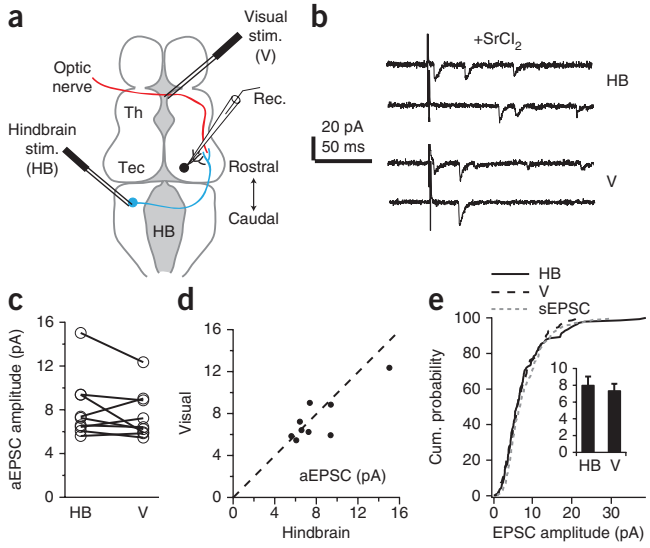

Figure 1 Multisensory convergence in optic tectal neurons. (**a**) Stimulation and recording configuration. Using a whole-brain preparation from *Xenopus* tadpoles, stimulating (stim.) electrodes were placed in the optic chiasm and contralateral hindbrain (HB) to activate visual (V) or mechanosensory inputs to the tectum. Whole-cell recordings (rec.) were performed from optic tectal neurons. (**b**) Extracellular Ca^{2+} was substituted with Sr^{2+} to evoke aEPSCs. (**c**) aEPSCs evoked by either hindbrain or visual stimulation were not significantly different ($P = 0.359$). (**d**) Scatter plot of aEPSC amplitude evoked by each pathway. (**e**) Cumulative probability distribution of aEPSC amplitudes from both pathways superimposed with sEPSC amplitudes showed no differences in amplitude distributions.

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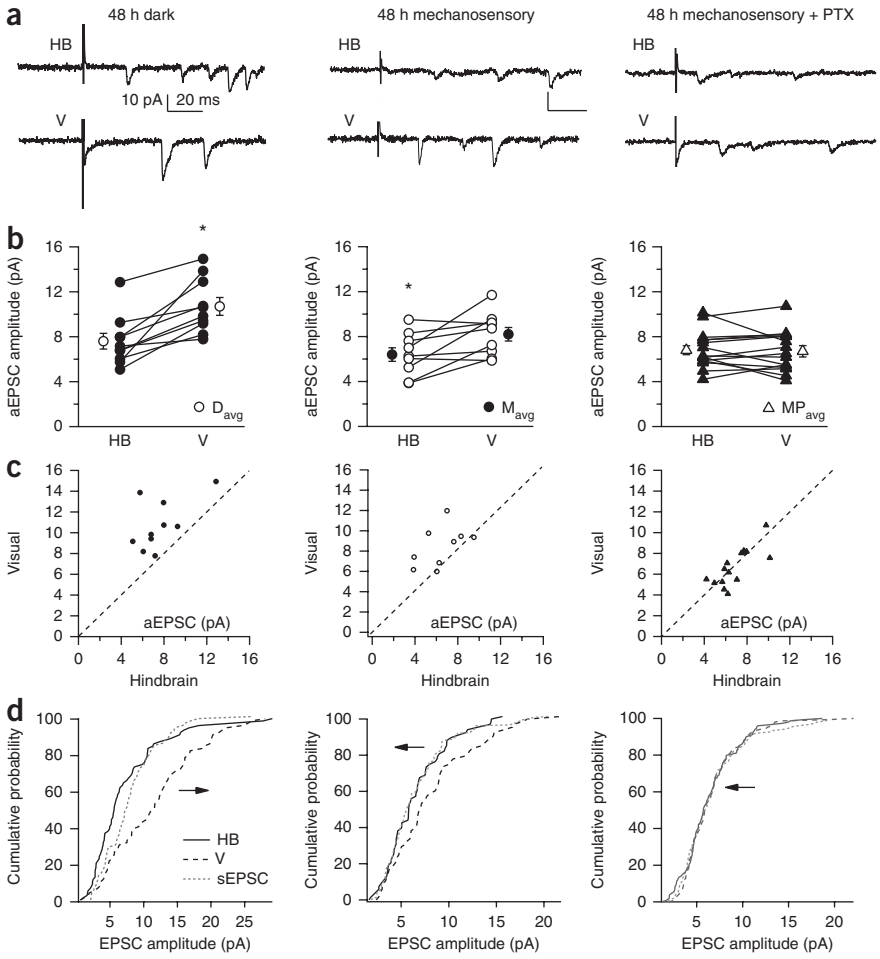

Figure 2 Modality-specific changes in aEPSC amplitude after various sensory manipulations. (**a**) aEPSCs evoked by visual or hindbrain stimulation after 48 h of visual deprivation (left), enhanced mechanosensory stimulation (middle) or enhanced mechanosensory stimulation in the presence of the inhibitory blocker PTX (right). (**b**) Comparison of hindbrain and visual stimulation after various experimental conditions. Symbols next to paired data represent average values and error bars represent s.e.m. D_{avg}, dark average; M_{avg}, mechanosensory average; MP_{avg}, mechanosensory average and PTX. (**c**) Scatterplot of aEPSC amplitude evoked by each pathway after various experimental conditions. (**d**) Cumulative probability distribution of aEPSC amplitudes from both pathways superimposed with sEPSC amplitudes after various experimental conditions. $^*P < 0.05$.

release of synaptic vesicles following electrical stimulation of a pathway[8] (**Fig. 1b**). These asynchronous EPSCs (aEPSC) were separately analyzed for each pathway. We also analyzed sEPSCs, which primarily arise from synaptic contacts made by intratectal recurrent axon collaterals[9]. In control tadpoles there was no significant difference between the aEPSC amplitude evoked by either the visual or the hindbrain input (visual, 7.5 ± 0.7 pA; hindbrain, 8.1 ± 1 pA; $n = 9$, $P = 0.359$; **Fig. 1c–e**). Furthermore, the range of amplitudes from both inputs was indistinguishable from the amplitudes of sEPSCs (spontaneous, 8 ± 0.9 pA; **Fig. 1e**), indicating that, in control conditions, the quantal amplitudes of all synaptic inputs are roughly matched to each other. Across cells there was a strong and significant correlation between the amplitude of evoked aEPSCs from different modalities and between evoked aEPSCs and sEPSCs (visual versus hindbrain, $r = 0.71$, $P = 0.037$; visual versus spontaneous, $r = 0.82$, $P = 0.011$; hindbrain versus spontaneous, $r = 0.78$, $P = 0.017$; Spearman R correlation).

We next asked whether long-term alterations in the activity of a specific sensory modality would result in either compensatory homeostatic plasticity specific to that pathway or in global alterations affecting all synapses (**Fig. 2**). First, tadpoles were placed in the dark for 48 h to selectively decrease visual input. We found that aEPSCs evoked from the optic nerve after treatment were significantly larger than aEPSCs evoked by hindbrain stimulation (visual, 10.74 ± 0.76 pA; hindbrain, 7.57 ± 0.7; $n = 10$, $P = 0.002$; **Figs. 2a–c** and **3a**). We also found that visual aEPSCs were significantly larger than sEPSCs (spontaneous, 7.58 ± 0.42 pA, $P < 0.001$; **Fig. 2d**), suggesting that visual deprivation resulted in a selective increase in retinotectal synapses. This change could be reversed by returning tadpoles to a normal rearing environment (12:12 h light/dark cycle) for 48 h (visual, 6.04 ± 0.44 pA; hindbrain, 6.09 ± 0.45 pA; $n = 11$, $P = 0.831$; **Supplementary Fig. 2**). We also tested whether 48 h of dark treatment resulted in changes in paired pulse facilitation (PPF), an index of presynaptic release probability. We found that visual synapses typically exhibit more PPF than hindbrain inputs, but we did not find any effects of dark rearing on PPF (hindbrain: control PPF, 111.2 ± 28.4%, dark PPF, 90.37 ± 11.7%, $P = 0.82$; visual: control PPF, 216 ± 54.2%, dark PPF, 279 ± 53.6, $P = 0.34$; $n = 8$ and 7, Mann-Whitney; **Supplementary Fig. 3a,c**) or in the ratio between PPF in visual versus hindbrain pathways (control, 2.41 ± 0.52; dark, 2.61 ± 0.45; $P = 0.82$; **Supplementary Fig. 3b**). These data indicate that no presynaptic changes occurred following 48 h of dark rearing.

Next, tadpoles were kept in normal light/dark conditions, but were stimulated with a constant vibration for 48 h to activate mechanosensory pathways. Vibration was delivered by gently bubbling air into their rearing tank. After treatment, aEPSCs evoked by hindbrain

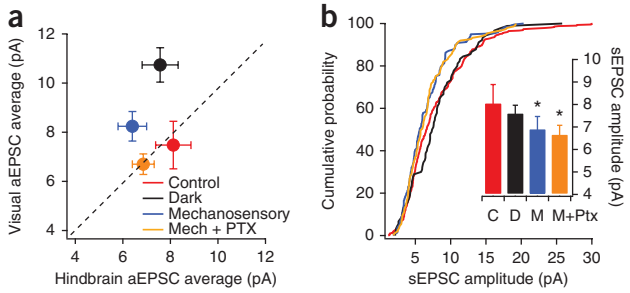

Figure 3 Summary of synaptic changes after *in vivo* sensory manipulations. (**a**) Averaged data comparing aEPSC amplitude evoked by visual and mechanosensory (mech) pathways under various conditions. Notice modality specific changes. (**b**) Cumulative probability plots of sEPSC amplitudes from different experimental groups: C, control; D, dark; M, mechanosensory; M+PTX, mechanosensory plus PTX. Inset shows average sEPSC amplitudes across conditions. Error bars are s.e.m., $^*P < 0.05$. For direct comparisons, see **Supplementary Figure 5**.

stimulation were significantly smaller than visual aEPSCs (visual, 8.24 ± 0.64 pA; hindbrain, 6.41 ± 0.63 pA; $n = 9$, $P = 0.019$; **Figs. 2a–c** and **3a**), indicating a pathway-specific change. Because activating afferent hindbrain input will also drive activity in recurrent intratectal synapses (**Supplementary Fig. 4**), we would expect that these would also be decreased after long-term mechanosensory stimulation. Consistently, sEPSCs were also significantly smaller than retinotectal aEPSCs (spontaneous, 6.89 ± 0.61 pA, $P = 0.02$; **Fig. 2d**). Finally, we tested whether we could eliminate differences between hindbrain and visual inputs by evoking a more global type of homeostatic plasticity[1] by strongly driving the system while simultaneously presenting mechanosensory stimulation. Tadpoles were exposed to 48 h of mechanosensory stimulation in the presence of 100 μM PTX, a $GABA_A$ antagonist, in the rearing media[9]. This would be expected to strongly activate the tectal network without necessarily enhancing visual input. After this treatment, we found no difference between visual and hindbrain-evoked aEPSCs (visual, 6.71 ± 1.8 pA; hindbrain, 6.86 ± 1.6 pA; $n = 15$, $P > 0.99$; **Fig. 2a–c**) and no difference between aEPSCs and sEPSCs (spontaneous, 6.66 ± 1.8 pA; **Fig. 2d**). However, sEPSCs in the mechanosensory and PTX–treated group were significantly smaller than control sEPSCs ($P = 0.005$; **Fig. 3b**). This suggests that stronger activation of the tectal circuitry can lead to global compensatory changes in synaptic transmission that can override local differences in synaptic activity.

To the best of our knowledge, this is the first example of homeostatic synaptic plasticity that is pathway specific and that can be induced by long-term *in vivo* manipulations of sensory activity. A synapse-specific form of homeostatic plasticity has been described in mossy fiber and recurrent collateral inputs to CA3 neurons in organotypic cultures[10]. In that experiment, however, it was not possible to manipulate different pathways independently and experiments were performed *in vitro*. Cross-modal homeostatic plasticity has been also described after visual deprivation, but these changes occurred in separate sensory cortices and not in individual neurons receiving converging inputs[11].

Our data indicate that pathway-specific homeostatic synaptic plasticity can be overridden by stronger global activation. What are the potential mechanisms for these types of plasticity? Inputs to individual tectal neurons from visual and mechanosensory pathways are anatomically segregated: visual inputs terminate primarily in the distal portion of the dendritic arbor, with mechanosensory inputs terminating more proximally[5,6]. The *in vivo* sensory manipulations that we used in this study could use this segregation to restrict activity to a local portion of the dendrite, creating a local signal for homeostatic plasticity. In contrast, global manipulations, such as adding PTX and mechanosensory stimulation, may override local signals to produce conventional forms of global homeostatic plasticity. The presence of dendritic spiking may mediate the transition between local and global forms of homeostatic plasticity. It is unclear whether local and global homeostatic plasticity require separate mechanisms or whether they share a common mechanism that is normally spatially restricted, but can also be expressed globally. Known plasticity mechanisms, such as BDNF release from dendrites[12,13] or local dendritic protein synthesis[14], are both activity dependent and could be harnessed to induce either global or local changes in synaptic strength.

Note: Supplementary information is available on the Nature Neuroscience website.

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AUTHOR CONTRIBUTIONS

K.E.D. and C.D.A. worked on the experimental design, performed the experiments and wrote the manuscript.

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

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1. Turrigiano, G.G. & Nelson, S.B. *Curr. Opin. Neurobiol.* **10**, 358–364 (2000).
2. Turrigiano, G.G., Leslie, K.R., Desai, N.S., Rutherford, L.C. & Nelson, S.B. *Nature* **391**, 892–896 (1998).
3. Desai, N.S., Cudmore, R.H., Nelson, S.B. & Turrigiano, G.G. *Nat. Neurosci.* **5**, 783–789 (2002).
4. Tyler, W.J., Petzold, G.C., Pal, S.K. & Murthy, V.N. *J. Neurosci.* **27**, 9427–9438 (2007).
5. Deeg, K.E., Sears, I.B. & Aizenman, C.D. *J. Neurophysiol.* **102**, 3392–3404 (2009).
6. Hiramoto, M. & Cline, H.T. *Dev. Neurobiol.* **69**, 959–971 (2009).
7. Pratt, K.G. & Aizenman, C.D. *J. Neurosci.* **27**, 8268–8277 (2007).
8. Xu-Friedman, M.A. & Regehr, W.G. *J. Neurosci.* **20**, 4414–4422 (2000).
9. Pratt, K.G., Dong, W. & Aizenman, C.D. *Nat. Neurosci.* **11**, 467–475 (2008).
10. Kim, J. & Tsien, R.W. *Neuron* **58**, 925–937 (2008).
11. Goel, A. *et al. Nat. Neurosci.* **9**, 1001–1003 (2006).
12. Kuczewski, N., Porcher, C., Lessmann, V., Medina, I. & Gaiarsa, J.L. *Mol. Neurobiol.* **39**, 37–49 (2009).
13. Rutherford, L.C., Nelson, S.B. & Turrigiano, G.G. *Neuron* **21**, 521–530 (1998).
14. Steward, O. & Schuman, E.M. *Neuron* **40**, 347–359 (2003).