The majority of cortical or hippocampal SOM⁺ neurons do not express ErbB4.

SOM⁺ neurons in the hippocampus (top panels) and cortex (bottom panels) were identified on the basis of their nucleus-localized H2B-GFP in the Som-Cre:H2b-GFP mice (left panels). ErbB4 expression was detected using an antibody (middle panels). ErbB4 is largely excluded from SOM⁺ neurons (overlay on right), and about 10.7 ± 0.4% of hippocampal (5 brain sections, 578 SOM⁺ neurons analyzed; 1 mouse) and 6.6 ± 1% of cortical (10 brain sections, 1286 SOM⁺ neurons analyzed; 1 mouse) SOM⁺ neurons expressed ErbB4. The inset in each panel is a higher magnification image of the boxed region.
Training mice in the 2-AC tasks.

(a) Left: the learning curve of different groups of mice in the basic auditory 2-AC task. Right: after having reached criteria in the auditory task, the same mice were further trained in the visual 2-AC task. (b) The number of sessions required for the mice to reach the criteria (75% performance) in the auditory (left) and visual (right) 2-AC tasks. The KO mice needed significantly fewer sessions to reach criteria in the auditory 2-AC task (auditory: WT, 21.5 ± 1.2 sessions, n = 32 mice; HET, 20.5 ± 1.0 sessions, n = 28 mice; KO, 15.6 ± 0.8 sessions, n = 30 mice; F(2,87) = 9.38, ***P = 0.0003, one-way ANOVA followed by Tukey’s test; visual: WT, 9.8 ± 0.8 sessions, n = 25 mice; HET, 9.3 ± 0.7 sessions, n = 22 mice; KO, 8.3 ± 0.8 sessions, n = 20 mice; F(2,64) = 0.96, P = 0.39, one-way ANOVA followed by Tukey’s test). (c) & (d) Selective deletion of ErbB4 in SOM+ TRN neurons facilitates learning in the 2-AC task. “Control”: Som-Flip;Erbb4\textsuperscript{lox/lox} mice in which the TRN was injected with a Flp-dependent AAV expressing GFP. “TRN KO”: Som-Flip;Erbb4\textsuperscript{lox/lox} mice in which the TRN was injected with a Flp-dependent AAV expressing Cre-GFP, so as to delete ErbB4 in SOM+ TRN neurons. (c) Left: the learning curve of these mice in the basic auditory 2-AC task. Right: after having reached criteria in the auditory task, the same mice were further trained in the visual 2-AC task. (d) The number of sessions required for these mice to reach criteria (75% performance) in the auditory (left) and visual (right) 2-AC tasks. The TRN KO mice needed significantly fewer sessions to reach criteria in the auditory 2-AC task (auditory: control, 20.57 ± 1.07 sessions, n = 7 mice, TRN KO, 15.86 ± 1.24 sessions, n = 7 mice, DF = 12, T = 2.88, *P = 0.014, t test; visual, control, 6.71 ± 0.68 sessions, n = 7 mice, TRN KO, 6.57 ± 0.53 sessions, n = 7 mice; DF = 12, T = 0.17, P = 0.87, t test). All mice were first trained in the auditory 2-AC task. Data are presented as mean ± s.e.m.
ErbB4 deficiency in SOM+ neurons does not affect sensory perception.

(a) The psychometric function of a representative mouse from each group in an auditory discrimination task. Mice were first trained to criteria in the basic auditory 2-AC task. They were then tested for discrimination of eight frequencies (in kHz): 8, 9.119, 10.39, 11.85, 13.5, 15.39, 17.55, and 20. Data from five consecutive sessions were collected (250-350 trials per session). Ordinate: the percentage of trials that the mice chose the water port on the right side. This data was fitted using a logistic function (see Methods). (b) Quantification of the median threshold, “Xo”, from the psychometric function. There was no significant difference among groups (WT, 13.63 ± 0.02, n = 11 mice; HET, 13.67 ± 0.04, n = 11 mice; KO, 13.67 ± 0.05, n = 10 mice; F(2,29) = 0.40, P = 0.67, one-way ANOVA). (c) Quantification of the parameter “p”. There was no significant difference among groups (WT, 8.3 ± 0.68, n = 11 mice; HET, 9.2 ± 0.97, n = 11 mice; KO, 9.02 ± 1.17, n = 10 mice; F(2,29) = 0.26, P = 0.78, one-way ANOVA). (d-g) Visual perception. (d) A schematic of the experimental setting of a visual discrimination task. A panel with eight individually illuminable LEDs was positioned above the water ports. In each trial, one of the eight LEDs was illuminated (indicated in yellow) for 300 ms. Mice were tested for discrimination of illumination at the eight positions (see Methods for details). (e) The psychometric function of a representative mouse from each group in the visual discrimination task. Data from five consecutive sessions were collected (250-350 trials per session). Ordinate: the percentage of trials...
that the mice chose the water port on the right side. This data was fitted using a logistic function (see Methods). (f) Quantification of the median threshold, \( X_o \), from the psychometric function. There was no significant difference between groups. (WT, -0.24 ± 0.16, n = 8 mice; KO, -0.18 ± 0.11, n = 8 mice; DF = 14, T = 0.31, \( P = 0.75 \), t test). (g) Quantification of parameter \( \theta \). There was no significant difference between groups (WT, 9.52 ± 1.02, n = 8 mice; KO, 9.72 ± 0.58, n = 8 mice; DF = 14, T = 0.17, \( P = 0.86 \), t test). Data are presented as mean ± s.e.m.
ErbB4 deficiency in SOM+ neurons differentially affects sensory selection in the visual/visual and auditory/visual tasks.

(a) A schematic of the experimental setting for the visual/visual task. The target was the illumination of four center LEDs (shown in red), while the distractors were flashing LEDs surrounding the target (see Methods for details). (b) Deficiency of ErbB4 in SOM+ neurons improved performance in the visual/visual task (WT n = 8 mice, KO n = 8 mice; F(1,14) = 36.48, **P = 0.0016; ***P = 0.0001, ****P <0.0001; two-way repeated measures (RM) ANOVA followed by Bonferroni tests). (c) A schematic of the auditory/visual task, in which the auditory cues are behaviorally relevant, whereas the visual cues are irrelevant (see Methods for details). (d) ErbB4 deficiency in SOM+ neurons impaired performance in the incongruent trials of the auditory/visual task (WT n = 10 mice, HET n = 11 mice, KO n = 9 mice; F(2,27) = 9.55; HET compared with WT: session 1, n.s., *P = 0.086, session 2, **P = 0.0023, session 3, *P = 0.033, session 4, n.s., *P = 0.085, session 5, **P = 0.0025; KO compared with WT: session 1, ***P = 0.0007, session 2, **P = 0.0032, session 3, *P = 0.037, session 4, n.s., *P = 0.15, session 5, **P = 0.0014; two-way RM ANOVA followed by Tukey’s tests). (e) ErbB4 deficiency in SOM+ neurons did not affect performance in the congruent trials of the auditory/visual task. Data were collected from the same mice as those in d (WT n = 10 mice, HET n = 11 mice, KO n = 9 mice, F(2,27) = 0.59; P = 0.56, Two-way RM ANOVA). Data are presented as mean ± s.e.m.
Supplementary Figure 5

Selective deletion of ErbB4 in SOM+ TRN neurons.

(a) Representative images of the TRN from a “control” Som-Flp;Erbb4<sup>lox/lox</sup> mouse, in which the TRN was injected with a Flp-dependent AAV expressing GFP (left). ErbB4 was recognized by an antibody (middle). Note the co-expression of ErbB4 and GFP in TRN neurons (overlay in right). The result was repeated in 2 mice. (b) Representative images of the TRN from a “TRN KO” Som-Flp;Erbb4<sup>lox/lox</sup> mouse, in which the TRN was injected with a Flp-dependent AAV expressing Cre tagged with GFP (left). Note the lack of ErbB4 expression in GFP<sup>+</sup> TRN neurons (overlay in right). The result was repeated in 3 mice. The inset in each panel is a higher magnification image of the boxed region.
Supplementary Figure 6

ErbB4 deficiency in SOM+ TRN neurons enhances cortical drive onto TRN across sensory modalities.

(a) Left: a schematic of the recording configuration. The CT-TRN pathway originating from the auditory cortex is selectively stimulated by photo-activation of ChR2 (green), and EPSCs are recorded from SOM+ TRN neurons (red). Right: images of a brain slice used in the recording. The slice was prepared from a SOM-Cre;Ai14 mouse in which the AAV-CAG-ChR2(H134R)-YFP was injected into the primary auditory cortex. Although the auditory cortex was severed, the ChR2-YFP+ (green) fibers originating from auditory cortex can be readily observed projecting to the medial geniculate (MG) complex.

(b) Left: representative emEPSC traces recorded from SOM+ TRN neurons in response to photo-stimulation (blue bars) of the auditory CT-TRN pathway, using the minimal photo-stimulation protocol. Calibrations: 20 pA and 2 ms. Right: quantification of the amplitude of emEPSCs driven by the auditory CT-TRN pathway (WT: 24.07 ± 1.07 pA, n = 8 cells (2 mice); KO: 76.71 ± 5.71 pA, n = 8 cells (2 mice); DF = 14, T = 9.07, ****P < 0.0001, t test). (c and d) Shown in c and d are similar to those in a and b, respectively, except that AAV-CAG-ChR2(H134R)-YFP was injected into the primary visual cortex (ChR2-YFP+ fibers originating from the visual cortex can be observed projecting to the lateral geniculate (LG) complex) (c), and emEPSCs driven by the visual CT-TRN pathway was measured (d) (WT: 35.88 ± 3.24 pA, n = 10 cells (3 mice); KO: 82.05 ± 5.18 pA, n = 11 cells (3 mice); DF = 19, T = 7.38, ****P < 0.0001, t test). Data are presented as mean ± s.e.m.
Supplementary Figure 7

Monosynaptic excitation and disynaptic inhibition of thalamic neurons driven by the cortical inputs.

(a) A schematic recording configuration, in which the CT pathway was selectively stimulated by the optogenetic method. Cortical neurons (green) were infected with the AAV-CAG-ChR2(H134R)-YFP. The synaptic responses were recorded from neurons in the thalamus. (b & c). Representative EPSC (b) or IPSC (c) traces recorded from thalamic neurons in response to the photo-stimulation (blue bars). (b) Monosynaptic EPSCs of short latency, which could be blocked by CNQX, were recorded at –52 mV holding potential that was experimentally measured to be the reversal potential of the IPSCs under our recording conditions. (c) The same photo-stimulation evoked disynaptic IPSCs in thalamic neurons, which could be blocked by either CNQX (left) or picrotoxin (right). The IPSCs were recorded from two different thalamic neurons at 0 mV holding potential. The experiments in b and c were repeated in 2 cells/1 mouse for each condition with consistent results. Calibrations: 50 pA and 50 ms.
Supplementary Figure 8

Targeting TRN neurons with GluA4-C-tail.

(a) Representative images of SOM⁺ TRN neurons expressing GluA4-C-tail-GFP. The TRN of KO (SOM⁺ErbB4–/–) mice were bilaterally injected with the AAV-DIO-GluA4-C-tail-GFP, so that GluA4-C-tail-GFP (green fluorescent) was selectively expressed in SOM⁺ TRN neurons. An antibody recognizing NeuN was used to label all neurons (red). Bilateral coronal TRN sections spanning from Bregma −0.82 mm to −1.82 mm are shown. (b) The infection rate of AAV-DIO-GluA4-C-tail-GFP in the TRN, measured as the ratio of GFP⁺ neurons to NeuN⁺ cells. Note that ~80% of TRN neurons are SOM⁺ (Fig. 1). (c) The performance of mice in the auditory/auditory task significantly correlated with the infection rate by the AAV-DIO-GluA4-C-tail-GFP (n = 8 mice (same as those in Fig. 8a); $R^2 = 0.67$, black line; $F(1,6) = 12.41$, $P = 0.013$ by a linear regression). Data are presented as mean ± s.e.m.
**Supplementary Figure 9**

**Behavioral effects of GluA4-C-tail expression in SOM+ TRN neurons.**

(a & b) Expression of GluA4-C-tail in SOM+ TRN neurons normalizes learning of KO mice in the basic 2-AC tasks. (a) Left: the learning curve of different groups of mice in the basic auditory 2-AC task. Right: after having reached criteria in the auditory task, the same mice were further trained in the visual 2-AC task. (b) The number of sessions required for the mice to reach criteria (75% performance) in the auditory 2-AC task (left) (“KO, GFP”, n = 8; “KO, C-tail-GFP”, n = 8; DF = 14, T = 2.36, *P = 0.034, t-test) and visual 2-AC task (right) (DF = 14, T = 2.23, *P = 0.043, t-test). The WT data is the same as that in Supplementary Fig. 2a & b, which is not significantly different from that of the “KO, C-tail-GFP” group (P = 0.66 for the auditory task, P = 0.29 for the visual task; t-test). All mice were first trained in the auditory 2-AC task. (c – e) Expression of GluA4-C-tail in SOM+ TRN neurons does not affect auditory perception of KO mice. (c) The psychometric function of a representative mouse from each group in the auditory discrimination task. (d) Quantification of the median threshold “X₀” from the psychometric function (“KO, GFP”, n = 8 mice; “KO, C-tail-GFP”, n = 8 mice; n.s., not significant, DF = 14, T = 0.12, P = 0.91, t-test). (e) Quantification of the parameter “p” (n.s., not significant, DF = 14, T = 1.81, P = 0.1, t-test). Data are presented as mean ± s.e.m.
Supplementary Figure 10

A model for TRN function in the selection of behaviorally relevant sensory inputs.

Schematics of the cortico–TRN–thalamic circuitry in the sensory selection tasks are shown. The thickness of each line represents the strength of a specific output. Dashed lines denote the outputs that are less active or largely suppressed. Arrows and bars denote excitatory outputs and inhibitory outputs, respectively. For simplicity, only relevant connections are indicated. The existence of these anatomical or functional connections has been described previously (for a detailed connectivity of TRN circuitry, see Zikopoulos and Barbas 2007 & 2012). Only the possible role of SOM+ TRN neurons in the auditory/auditory task (a) and visual/auditory task (b) is indicated; however, the same can also be applied to the visual/visual task and auditory/visual task, respectively. (a) Left: the open-loop circuits between TRN and thalamus provide the anatomical basis for lateral inhibition, which suppresses thalamic neurons that might respond to distractors. In this scenario, thalamic responses driven by behaviorally relevant stimuli (the “targets”) will have preferential access to the cortex. Note that the targets are associated with reward. Therefore they are behaviorally relevant and can engage goal-directed (top-down) attention, as indicated by an arrow from the cortex to a relevant thalamic neuron and the corresponding TRN neuron. In contrast, the distractors in this task act primarily through a sensory-driven (bottom-up), rather than a top-down process because they do not predict reward. Right: in conditions in which the cortical synaptic transmission onto TRN neurons is strengthened (indicated by a thicker arrow), such as that in the ErbB4 mutant mice, the lateral inhibition described above is enhanced, leading to increased signal-to-noise ratio in the thalamus and therefore improved performance. (b) Left, in the visual/auditory task, activation of neurons in the visual TRN leads to inhibition of neurons in the auditory sector. Unlike the auditory/auditory task, in which distractors mainly interfere with attention through a bottom-up process, in the visual/auditory task both the relevant (visual) and the irrelevant (auditory) sensory cues have been associated with reward during the initial training phase of the task. This prior knowledge can allow not only the relevant, but also the irrelevant cues to engage goal-directed attention (indicated by arrows from the cortex to thalamic neurons and the corresponding TRN neurons). This may lead to the activation of irrelevant neurons and suppression of relevant...
neurons in the thalamus and TRN, resulting in performance error. Right: in ErbB4 mutant mice in which the cortical synaptic transmission onto TRN neurons is excessively strengthened (indicated by thicker arrows), the probability of an auditory (irrelevant) TRN neuron to escape inhibition is increased, leading to decreased signal-to-noise ratio in the TRN and thalamus, and thus impairment in behavioral performance.