

Non-additive effect of simultaneous presentation of different sensory stimuli on PC Ca^{2+} events. Related to Figure 1.

(A) In the same mouse, a visual or a somatosensory stimulus was presented alone or in simultaneous combination.

(B) Summary plots showing the change in frequency and size of PC dendritic Ca^{2+} events both to individual sensory modalities or to their simultaneous presentation. Data are mean \pm SEM with individual mice in gray; ns, not significant, p = 0.45 and 0.40; paired Student's t-test (n = 6-7 mice).



Lack of coincident MLI activity with spontaneous Ca^{2+} events in PC dendrites. Related to Figure 2.

(A) MLIs in *Kit::Cre* mice were transduced with AAV containing Cre-dependent RCaMP2, a red genetically encoded Ca^{2+} indicator. A second AAV containing the green Ca^{2+} indicator GCaMP6f, expressed under control of the Pcp2 promoter, transduced PCs. (B) Left: Image of an isolated, GCaMP6f-expressing PC dendrite surrounded by RCaMP2-expressing MLIs. Right: Simultaneous, dual-color Ca^{2+} activity measurements from PCs and MLIs during quiescence. An isolated dendritic Ca^{2+} event is demarcated by the black tic mark. (C) Average of MLI Ca^{2+} activity aligned to the occurrence of spontaneous CF-evoked dendritic Ca^{2+} events (n = 212; 3 mice) in nearby PCs (within 50 µm).



Granule cell-mediated excitation contributes to the sensory-evoked activation of MLIs. Related to Figure 2.

(A) Trial-averaged ensemble responses of MLIs to different sensory modalities, measured in two different lobules across mice (n = 6-20 trial blocks; 5-7 mice per condition).

(B) Ca^{2+} activity measurements from all identified MLIs in a single field of view from one mouse. The responses are an average of the repeated stimulation of whiskers with an air puff (n = 15).

(C) AAV containing Cre-dependent hM4d was injected into *Math1::Cre* mice. A second virus transduced MLIs with GCaMP6f.

(**D**) Average sensory-evoked Ca^{2+} response in MLIs in control and after CNO administration to suppress the activity of hM4d-expressing granule cells.

(E) Chemogenetic suppression of granule cell activity significantly reduced peak MLI responses to sensory stimuli (p = 0.005, Student's paired t-test; n = 9 regions; 3 mice). Black symbols: mean. Gray symbols: individual regions.



MLIs do not account for differences in sensory-evoked Ca^{2+} *events in PC dendrites. Related to Figure 2.*

(A) Left: Simultaneous dual-color activity measurements from PCs and MLIs during repeated stimulation of the whiskers with an air puff. Yellow bars indicate timing of the sensory stimuli. Right: Superimposition of sensory-evoked Ca²⁺ events in PC dendrites and the response elicited in the surrounding MLI ensemble.

(B) Relationship between the cue-dependent enhancement of Ca^{2+} events in PC dendrites and the peak amplitude of the sensory-evoked response in MLIs. Each point represents the average response from simultaneous activity measurements of MLI ensemble activity and the change in PC dendritic Ca^{2+} size from a block of trials (measurements from 3 mice).



Brief suppression of inferior olive activity does not affect PC simple spiking. Related to Figure 3.

(A) Activity measurements from PC units in Crus I recorded with a silicon probe while animals sat in quiescence. *Gt*ACR2-expressing neurons in the inferior olive were suppressed using blue light illumination with an implanted optical fiber ($\lambda = 473$; 250 µW; 10 s).

(B) Top: Continuous record from a PC unit before, during, and after optogenetic suppression of the inferior olive. Complex spikes are marked with an asterisk. Bottom: Average (black traces) and individual examples (gray traces) of complex spikes (CS) and simple spikes (SS). (C) Loss of complex spikes were used to classify PC units sensitive to inferior olive activity suppression. In these same units (n = 15, 3 mice), simple spiking was not significantly affected by the optogenetic stimulus (p = 0.66, ANOVA with Tukey's multiple comparison test).



Post-hoc deletion of CF-evoked events does not reveal a residual, dendritic Ca^{2+} *response. Related to Figure 3.*

Average spontaneous Ca^{2+} events as well as those evoked by sensory stimuli in a sample of PC dendrites from control mice. For the latter condition, all algorithmically identified events (putative CF-evoked responses) were eliminated from the analysis (see Star Methods). The remaining response, in blue, fails to indicate a residual component attributable to the activity of parallel fibers (n = 15 trial blocks from 7 mice).



Specific effects of inferior olive activity suppression on CF-evoked signaling in PCs. Related to Figure 3.

(A) Left: In a field of view, PC dendrites demarcated in red were sensitive to suppression of the inferior olive as determined by the complete loss of spontaneous Ca^{2+} events during the illumination period. All other neighboring PCs demarcated in blue were insensitive to this manipulation (maintained normal spontaneous Ca^{2+} event rates). Right: Example traces from corresponding PCs marked in the image during sensory cues (timing in yellow) show either the presence or absence of stimulus-evoked activity in insensitive and sensitive cells, respectively. (B) For sensory-triggered Ca^{2+} events in the two classified populations of PCs, comparisons of the peak amplitudes of integrated responses. Black symbols mark means ± SEM. Gray lines show individual experiments (n = 10 experiments, 4 mice). Note that in PCs insensitive to olivary suppression, there were no significant differences in activity during the optogenetic stimulus (p = 0.53, paired Student's t-test).





(A) Left: In acute cerebellar slices, PCs were filled with a Ca^{2+} indicator dye during whole-cell, current-clamp recordings. Activity was elicited in PCs by CF stimulation. Representative simultaneous measurements of somatic complex spikes (middle) and volume-averaged dendritic Ca^{2+} signals (right) elicited in a PC in response to increasing numbers of CF stimuli delivered at 300 Hz.

(B) After the first stimulus, the average peak amplitude of the PC Ca^{2+} response increased proportionally with each additional presynaptic spike in the burst. Mean dendritic responses, normalized to a single CF stimulus, are indicated \pm SEM (n = 10 dendritic regions, 5 cells).