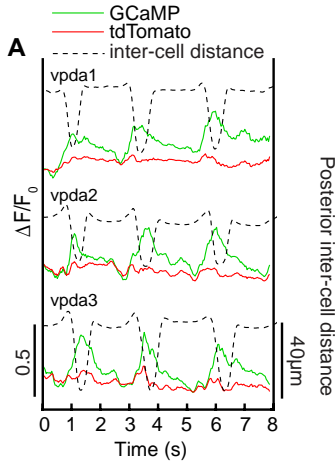
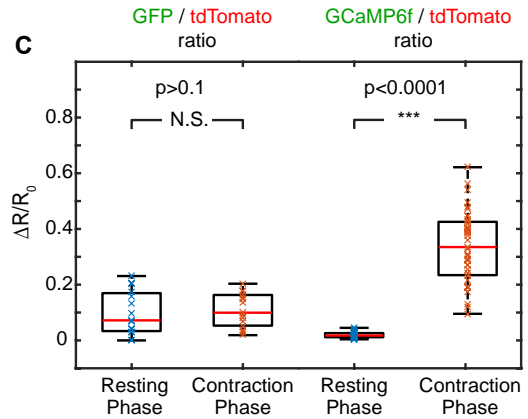
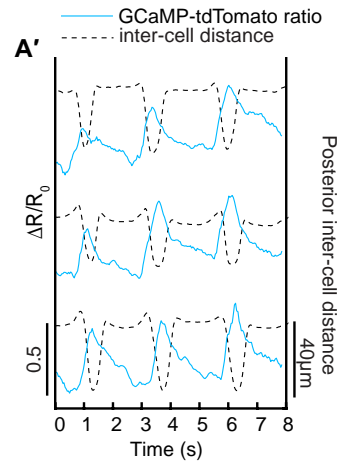


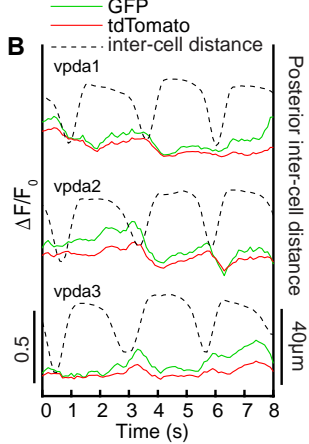
### GCaMP, raw signals



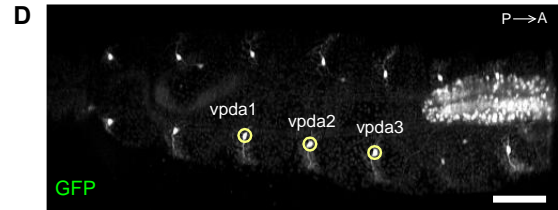
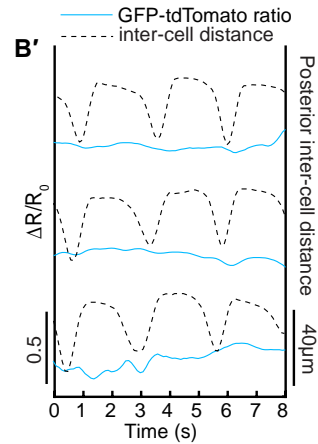
### GCaMP, green/red ratios



### GFP control, raw signals



### GFP control, green/red ratios

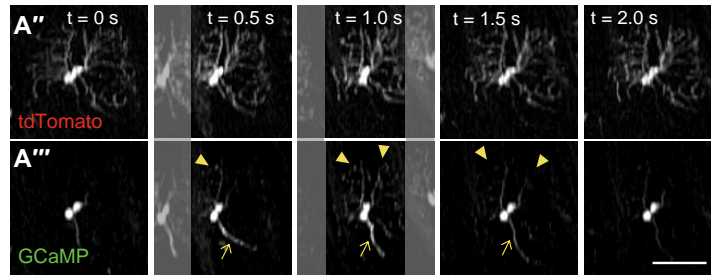
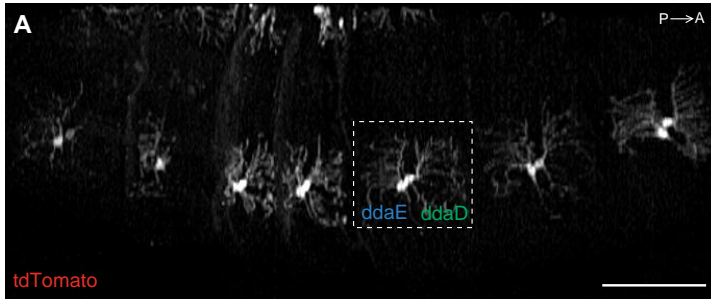


neurons tracked in GFP control

**Figure S1. Ratiometrically measured calcium dynamics properly control for motion artifacts, related to Figure 2.** (A) Change in fluorescence from baseline ( $\Delta F/F_0$ ) in GCaMP6f (green) and tdTomato (red) during crawling in vpda neuron somas. Segment contraction is depicted with inter-cell distance (dashed lines). (A') Change in ratio of GCaMP6f to tdTomato fluorescence ( $\Delta R/R_0$ , blue). Increases can be seen during segment contraction. (B) Change in fluorescence from baseline ( $\Delta F/F_0$ ) in GFP (green) and tdTomato (red) during crawling in vpda neuron somas. Segment contraction is depicted with inter-cell distance (dashed lines). (B') Change in ratio of GFP to tdTomato fluorescence ( $\Delta R/R_0$ , blue). No increase is associated with segment contraction. (C) Comparison of GFP-tdTomato and GCaMP6f-tdtomato ratios between resting and contraction phases (see methods). For GFP-tdTomato analysis,  $n = 2$  animals, 7 cells, 14 events, for GCaMP6f-tdtomato analysis,  $n = 3$  animals, 22 cells, 26 events. Note that there is no difference between GFP-tdTomato ratios in the resting versus contraction phases, while there is a significant increase in GCaMP6f-tdTomato ratios during contraction ( $p < 0.001$ , as measured by two-tailed t-test). (D) SCAPE imaging of *410-Gal4*, *20XUAS mCD8::GFP*, *UAS-CD4-tdTomato* animals during forward crawling, Ventral side. Imaging shows vpda neurons. GFP channel is shown. Posterior is to the left. Images are shown on a square root grayscale to reduce dynamic range for visualization of both cell bodies and dendrites. Scale bar = 100µm.

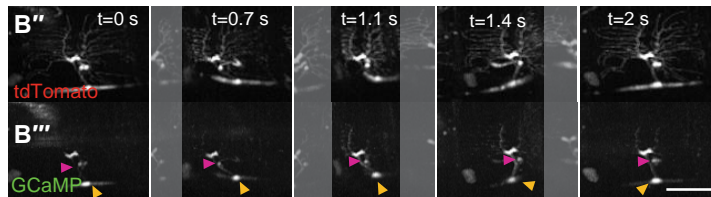
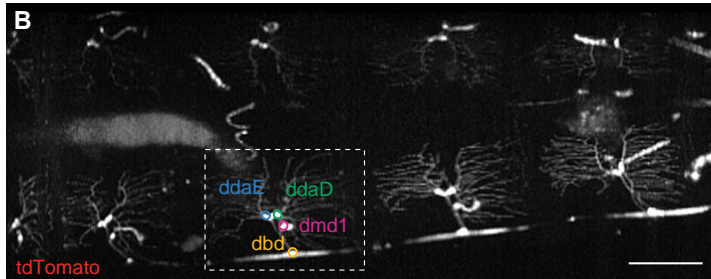
### dorsal class I neurons

410-Gal4, 20XUAS-IVS-GCaMP6f (x2), UAS-CD4-tdTomato



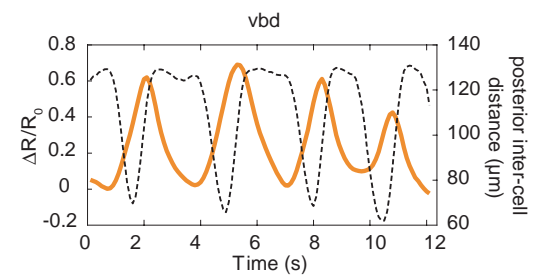
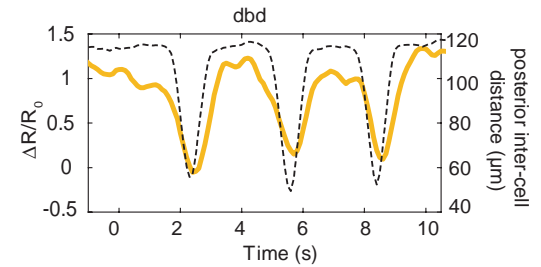
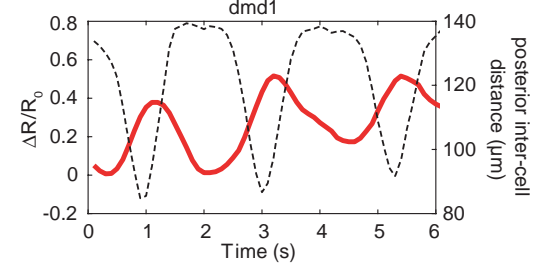
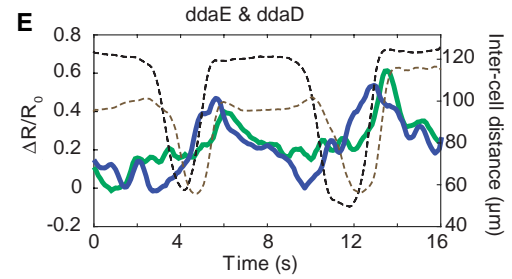
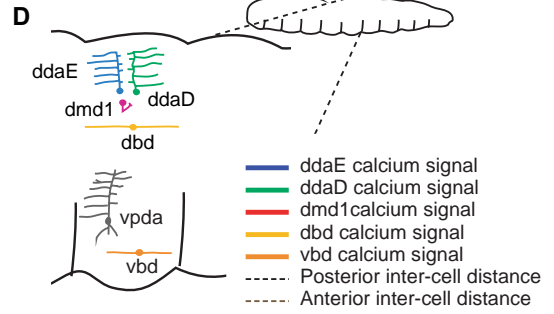
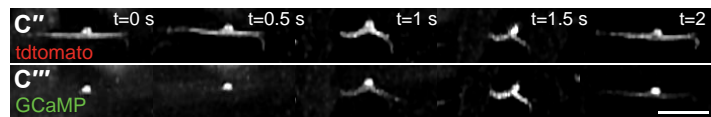
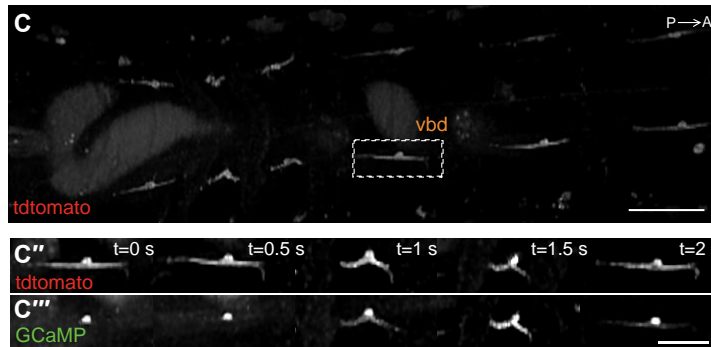
### all dorsal proprioceptors

10D05-Gal4, 20XUAS-IVS-GCaMP6f (x2), UAS-CD4-tdTomato



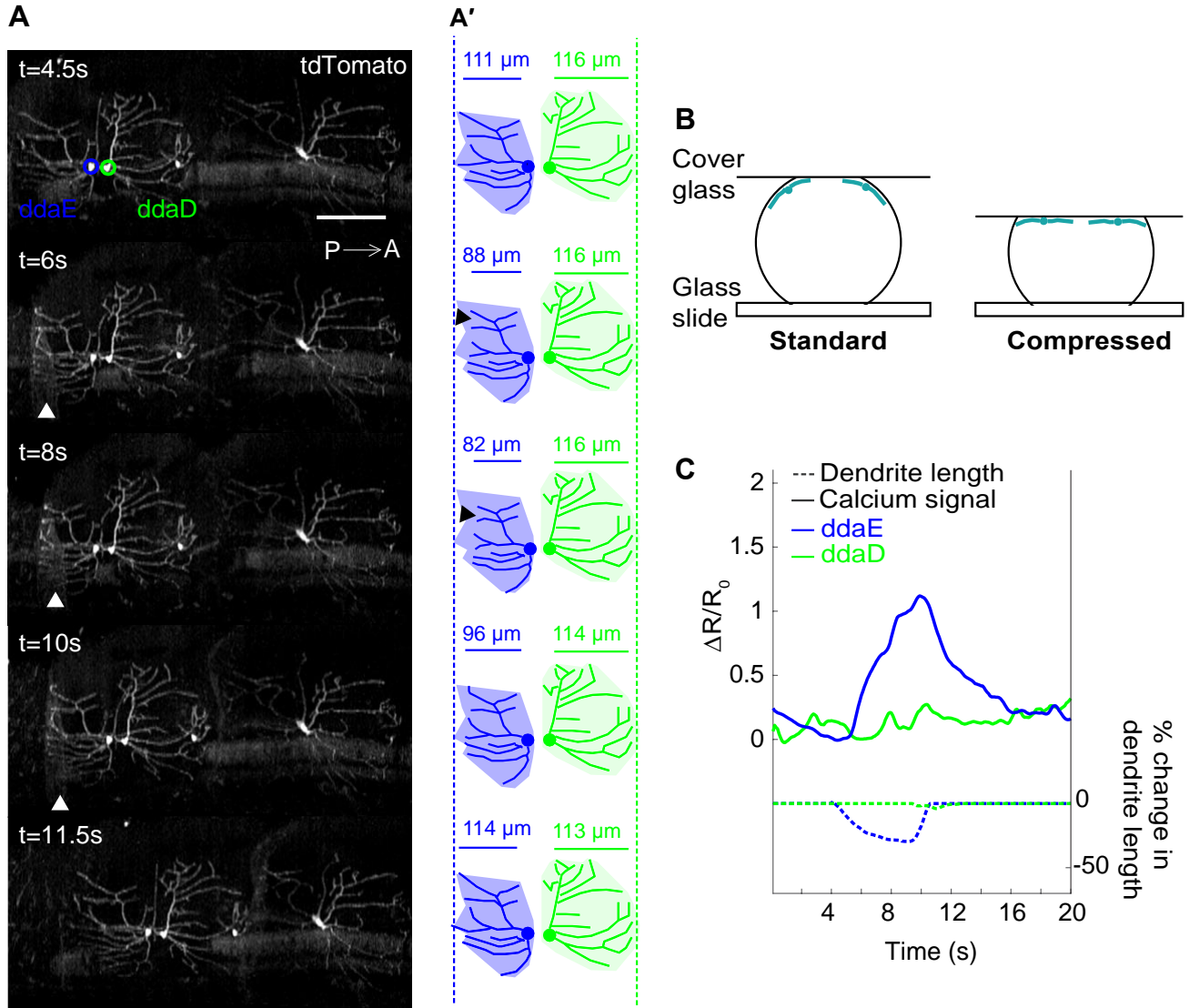
### vbd neurons

1129-Gal4, 20XUAS-IVS-GCaMP6f (x2), UAS-CD4-tdTomato



**Figure S2. Examples of SCAPE imaging of GCaMP dynamics, related to Figure 3.** Posterior is to the left for all images. For (A-C), images show representative SCAPE MIPs over a 35–90  $\mu\text{m}$  depth range from a 160–200 deep volume (to exclude

gut autofluorescence, square root grayscale). Dashed box indicates neurons examined in time lapse sequences below, shown for both tdTomato and GCaMP channels. **(A-A'')** SCAPE imaging of *410-Gal4*, *20XUAS-IVS-GCaMP6f* (x2), *UAS-CD4-tdTomato* larva. See Video S5, first section. Arrowheads indicate increases in dendritic GCaMP6f, arrows indicate increases in axon bundle (containing both ddaD and ddaE axons). Note ddaE dendrites are active before ddaD. **(B-B'')** SCAPE imaging of *GMR10D05-Gal4*, *20XUAS-IVS-GCaMP6f* (x2), *UAS-CD4-tdTomato* larva. See Video S5, second section. Orange arrowhead marks dbd cell body, pink arrowhead marks dmd1 cell body. **(C-C'')** SCAPE imaging of *1129-Gal4*, *20XUAS-IVS-GCaMP6f* (x2), *UAS-CD4-tdTomato* larva. See Video S5, third section. **(D)** Schematic of larval proprioceptive system. **(E)** Examples of single cell calcium activity dynamics during forward crawling. The calcium response is plotted in solid lines (quantified as  $\Delta R/R_0$ ). The distance between the measured neuron and the posterior neuron (posterior inter-cell distance) is plotted in black dashed lines. The distance between the measured neuron and the anterior neuron (anterior inter-cell distance) is also plotted in brown dashed lines on the ddaD plot, since this is a better proxy for dendrite folding.



**Figure S3. Sensory activity does not occur in the absence of dendritic folding, related to Figure 3.**

(A) Time lapse of SCAPE imaging of dorsal class I neurons labeled with *410-Gal4*, *20XUAS-IVS-GCaMP6f* (x2), *UAS-CD4-tdTomato*, in a compressed preparation, which prevents dendritic folding in *ddaD* (see (B) and methods). TdTomato channel is shown to depict dendrite dynamics. Larva is 3<sup>rd</sup> instar. Posterior is to the left. (MIP) over a 50 $\mu\text{m}$  depth range from a 160 $\mu\text{m}$  deep volume. (A') Tracing of time lapse data shown in (A), posterior cells. *ddaE* is blue and *ddaD* is green. Dotted lines and shaded areas represent extent of arbor in a relaxed segment. Measurements represent dendrite length ( $\mu\text{m}$ ), a measure of dendrite folding. Arrows denote frames with dendrite folding. Note that *ddaE* dendrites fold, but not *ddaD*. (B) Schematic of compressed preparation. (C) Calcium responses ( $\Delta R/R_0$ , solid lines) and % change in dendrite length (dotted lines) in a compressed preparation of *ddaE* (blue) and *ddaD* (green) during segment contraction. Activity correlates with dendrite folding. Scale bar=100 $\mu\text{m}$ .