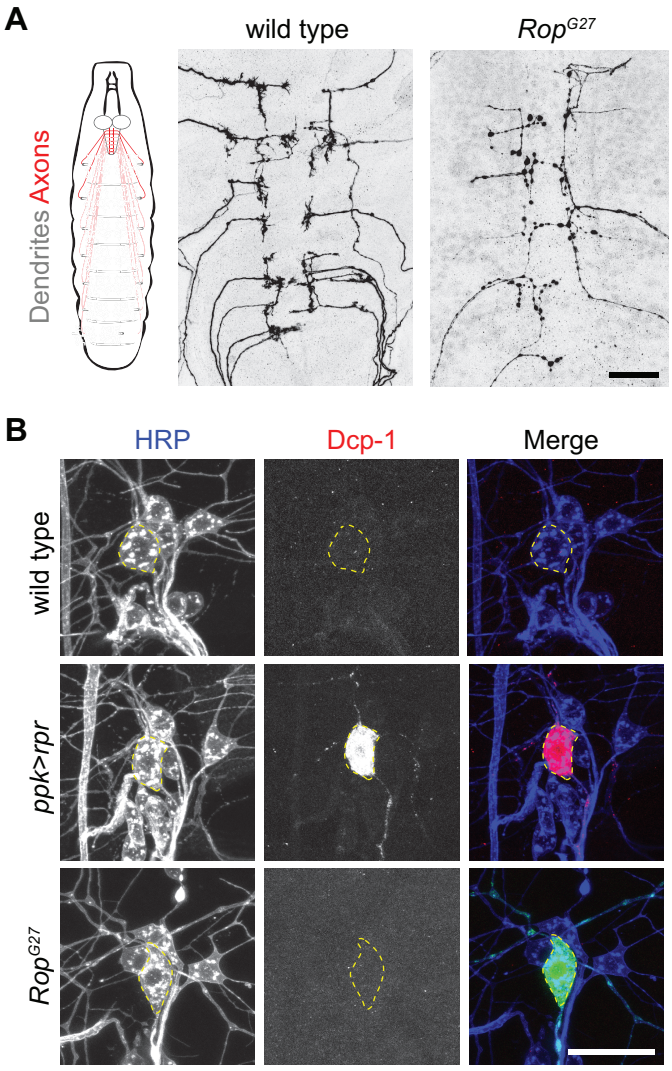

Figure S1. Axon and dendrite phenotypes of *Rop* mutant neurons are unlikely to be a consequence of cell death. (A) Axons of *Rop*^{G27} da neuron MARCM clones appear thin with varicosities along the terminals at 144 h AEL compared to wild-type controls. (B) Dorsal cluster of da neurons as revealed by staining with horseradish peroxidase (HRP). Wild-type C4da neurons and *Rop*^{G27} C4da-ddaC MARCM clones (labeled in green) show no detectable signal for Death caspase-1 (Dcp-1). By contrast, C4da neurons expressing the apoptosis activator Reaper (*ppk>rpr*) stain positive for Dcp-1. Scale bars, 25 μm.

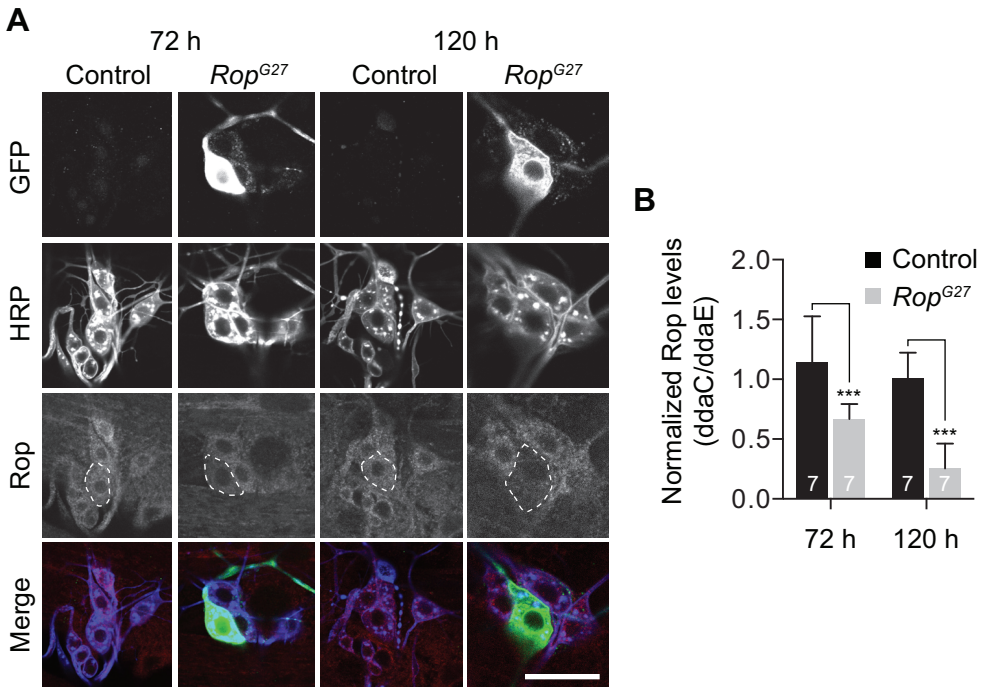

Figure S2. Rop protein perdures through early larval stages in *Rop*^{G27} mutant C4da MARCM clones. (A) *Rop*^{G27} mutant C4da MARCM clones and *Rop*^{G27}/+ heterozygous control C4da neurons (from the neighboring segment in the same larva) were stained with antibodies for HRP (to label sensory neurons) and Rop. Representative images and (B) quantification (mean and standard deviation, number of neurons indicated for each sample) of Rop levels (mean Rop immunoreactivity in the C4da soma, outlined with white hatched line) are shown for each genotype-timepoint combination. Scale bar, 25 μm.

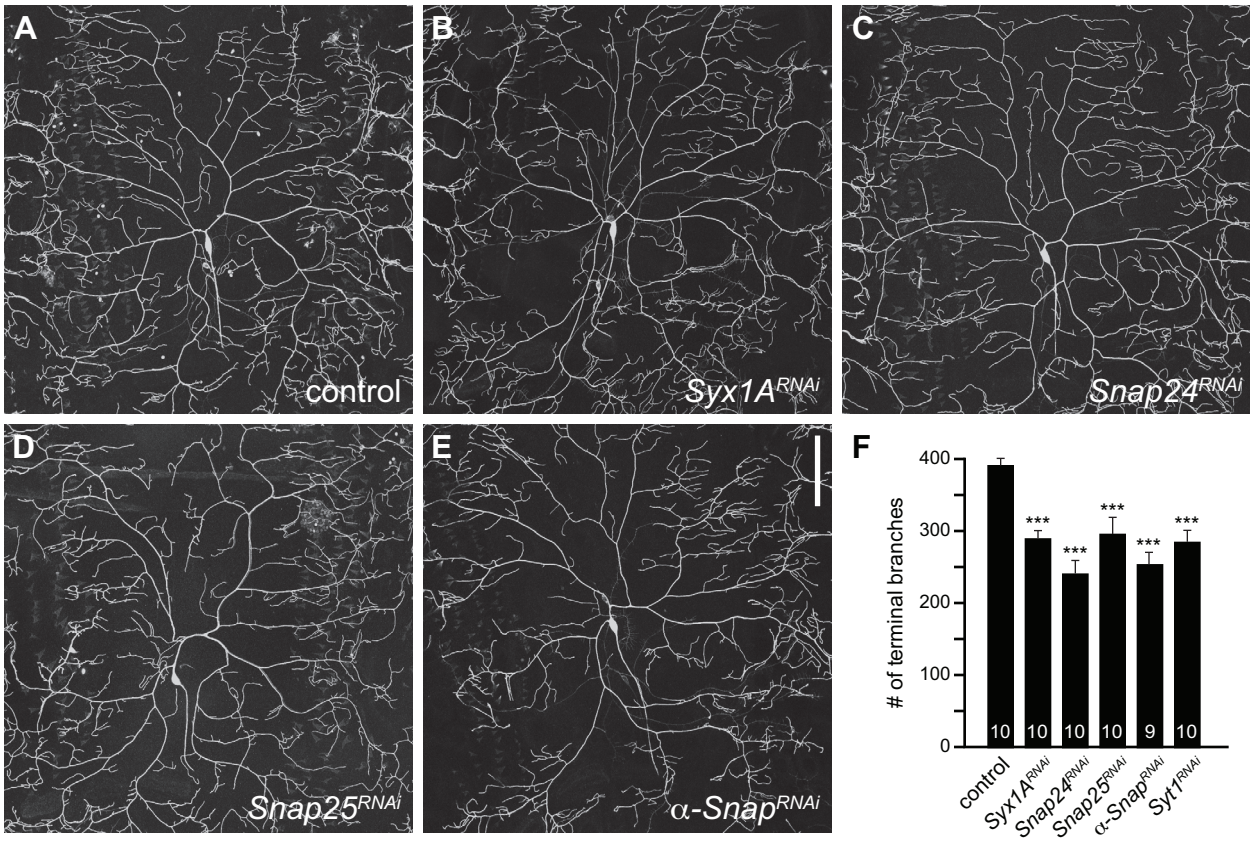

Figure S3. SNARE proteins regulate terminal dendritic branching. Dendritic morphologies of representative C4da neurons in a *UAS-Dicer-2* control (A) or expressing RNAi transgenes targeting *Syx1A* (B), *Snap24* (C), *Snap25* (D), and α-*Snap* (E). (F) Mean and standard deviation of terminal branch number with the number of neurons analyzed for each genotype indicated. *** $p < 0.001$, compared to control (one-way ANOVA with Tukey's HSD *post hoc* analysis). Scale bar, 50 μm.

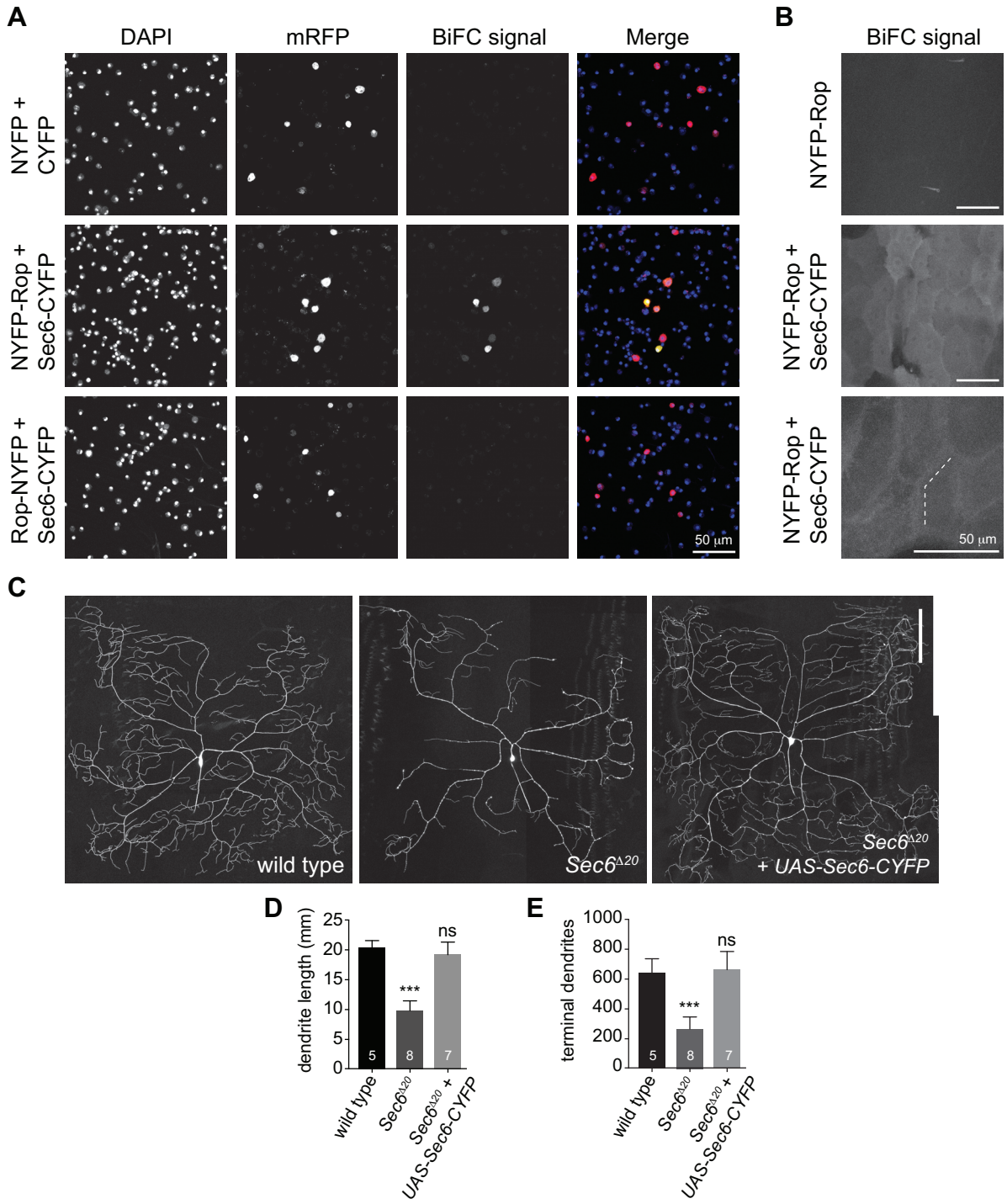

Figure S4. Monitoring Rop-Sec6 complex formation using BiFC. (A) S2 cells co-transfected with Actin-Gal4 + UAS-RFP (to mark transfected cells) and the indicated expression constructs (e.g. UAS-NYFP + UAS-NYFP) were DAPI stained and imaged using epifluorescence microscopy to assay for YFP reassembly (BiFC signal). (B) Co-expression of UAS-NYFP-Rop and UAS-Sec6-CYFP under control of the epidermal A58-Gal4 driver, but not expression of UAS-NYFP-Rop alone, results in YFP fluorescence in the body wall, indicative of Rop-Sec6 complex formation. YFP fluorescence is enriched at cell-cell junctions (dashed line).(C) Dendritic morphologies of representative wild-type, *Sec6*$^{\Delta 20}$, or *Sec6*$^{\Delta 20}$ + *UAS-Sec6-CYFP* C4da-ddaC MARCM clones. Scale bar, 100 μm. (D,E) Mean and standard deviation of total dendrite length (D) and terminal dendrite number (E) is shown, with the number of neurons analyzed for each genotype indicated. Bars represent mean values *** $p < 0.001$, ns, not significant, compared to wild type controls (one-way ANOVA with Tukey's HSD *post hoc* analysis).