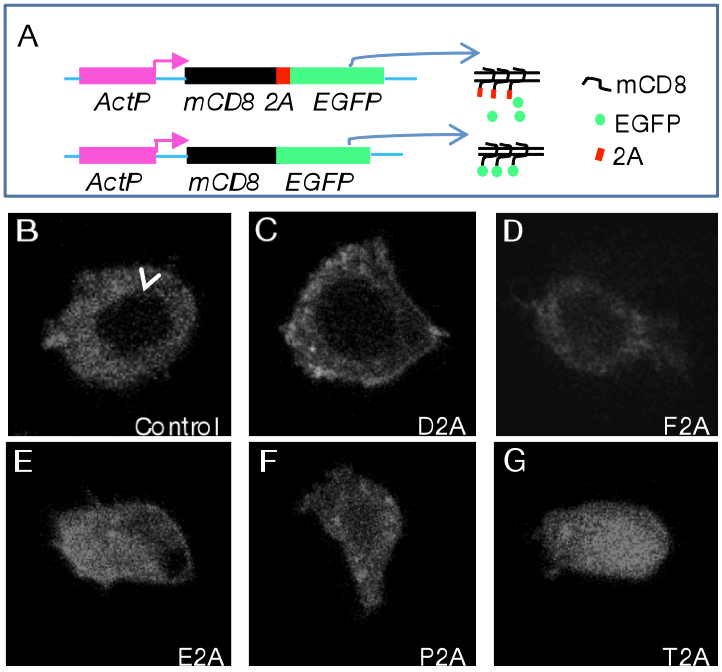

Figure S1 T2A and other 2A peptides facilitate ribosomal skipping in *Drosophila* SL2 cells. (A) Schematic of the mCD8-2A-EGFP construct used to test the faciliation of ribosomal skipping by five 2A peptides (top) and the control mCD8-EGFP construct (bottom). Both constructs are expressed under the control of the *actin* promoter. (B-G) The expression and localization of EGFP in cells transfected with the constructs shown in (A). The images are confocal micrographs of the EGFP distribution in representative individual cells. (B) Tethering EGFP to the membrane by fusing it to mCD8 restricts access of the fluorophore to the nucleus (arrowhead), as is evident in cells made with the control construct lacking a 2A peptide. Constructs made with the D2A (C) or F2A (D) peptides had non-nuclear labeling, similar to controls indicating poor facilitation of ribosomal skipping. In contrast, transfection with the E2A (E), P2A (F), and T2A (G) constructs showed nuclear EGFP localization, with the T2A construct yielding the most uniform labeling patterns suggesting high levels of ribosomal skipping, and the production of soluble, rather than membrane-bound, EGFP. The 2A peptides tested are as indicated in the Materials and Methods.