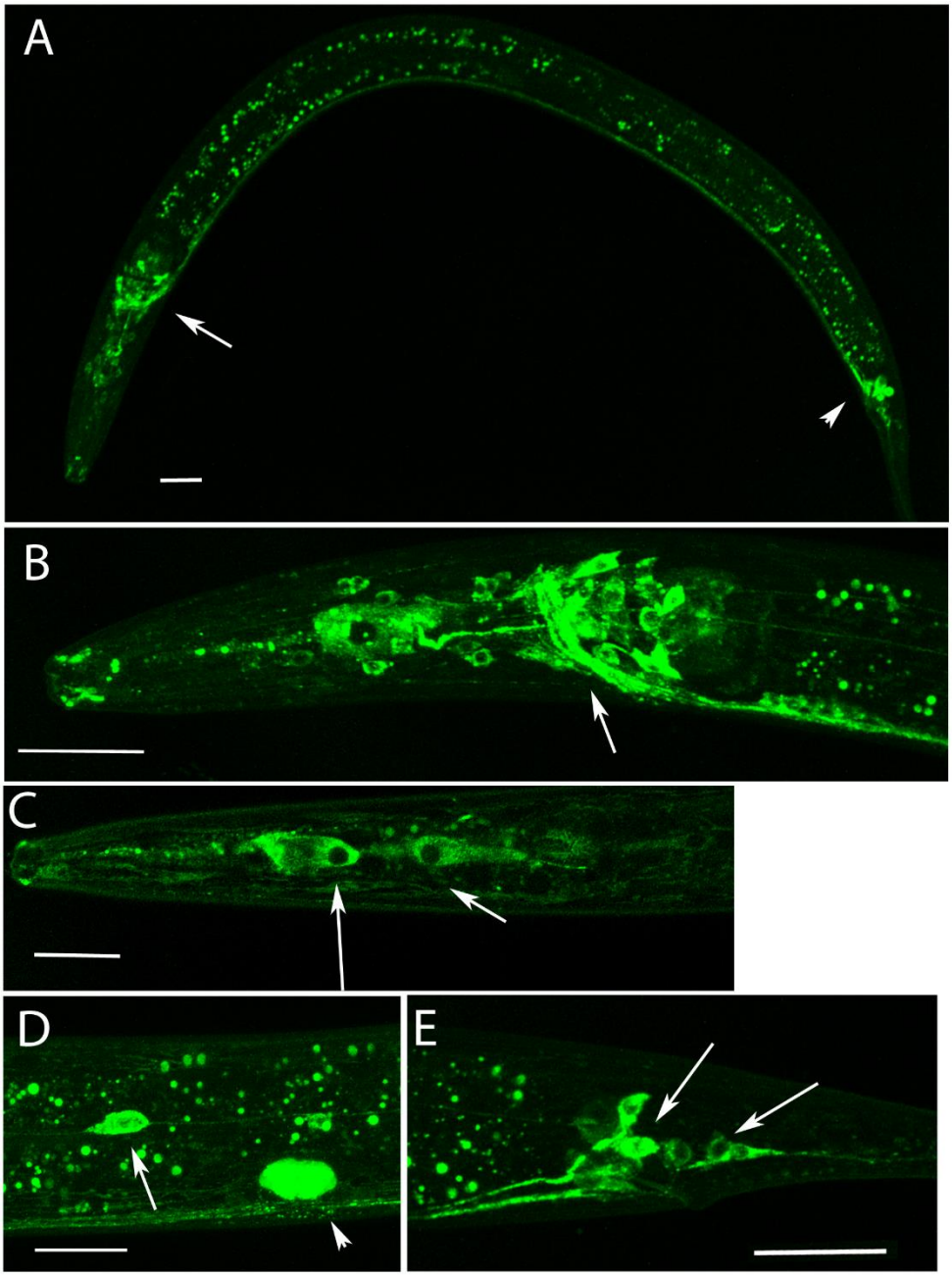

Figure S1: *efn-4* expression in a L4-staged larva
(A) Confocal micrographs showing EFN-4::GFP expression in *lin-15(n765); juIs109* animals, which carry an integrated EFN-4::GFP transgene (Chin-sang et al., 2002). As previously reported, EFN-4::GFP is expressed in the nervous system and can be detected in head neurons (A, B), the nerve ring (B, arrow) and the ventral nerve cord (A, B), seam cells (C, arrows), lateral neurons, including the CAN neuron (D, arrow), vulval cells (D, arrowhead), and tail neurons (A, arrowhead; E, arrows). Scale bar, 20 μm.

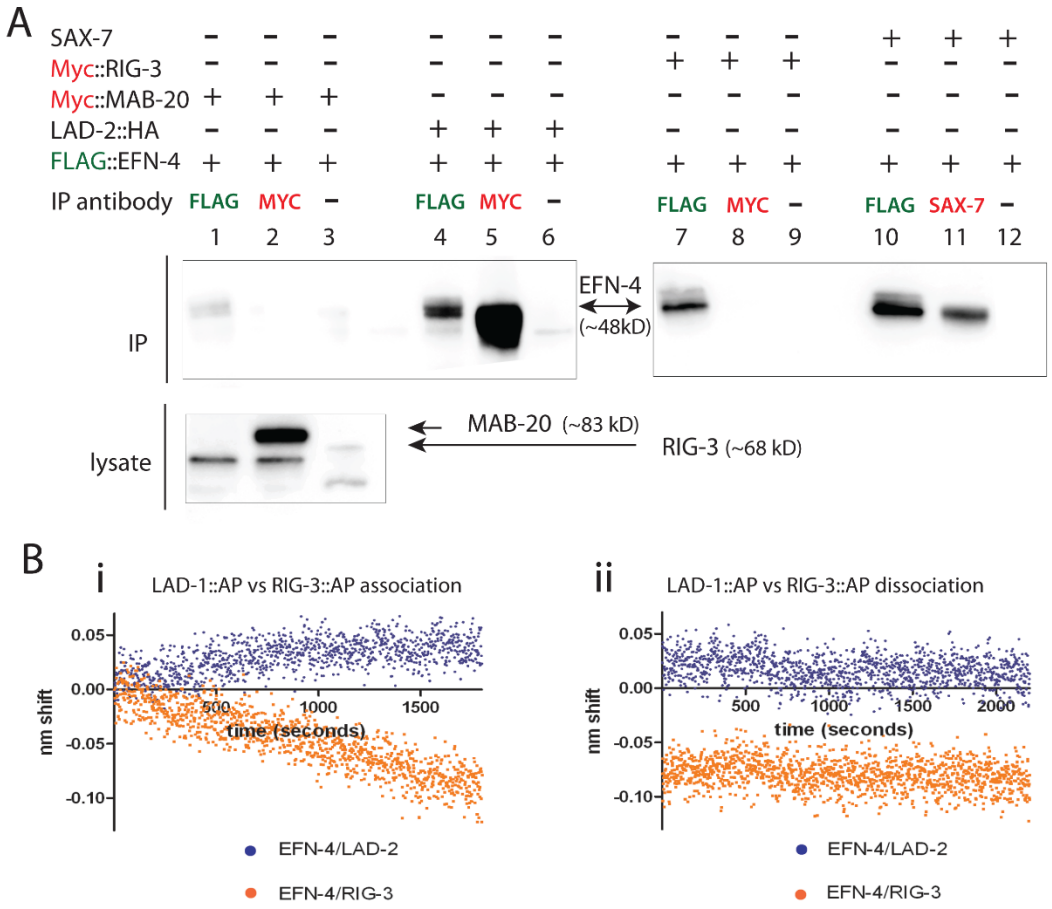

Fig S2: LAD-2 can biochemically interact with EFN-4.
(A) A Western blot showing results of a co-immunoprecipitation assay performed on lysates of HEK293T cells co-transfected with FLAG::EFN-4 and LAD-2::HA, Myc::MAB-20, Myc::RIG-3, or SAX-7. Western blot analysis reveals that FLAG::EFN-4 is pulled down in anti-HA but not anti-Myc immunoprecipitates, revealing that EFN-4 can interact with LAD-2 but not MAB-20 or RIG-3, which serves as negative controls. FLAG::EFN-4 is also pulled down in immunoprecipitates that use the 6991 anti-SAX-7 antibody (Chen et al., 2001), revealing that EFN-4 can also interact with SAX-7, which has a similar extracellular domain as LAD-2. (B) BioLayer Interferometry (BLI) reveals a fast-on, slow-off binding interaction between LAD-2-AP and EFN-4-Fc but not between RIG-3-AP and EFN-4-Fc. A BLI probe against human Fc was used to capture secreted EFN-4-Fc in parallel with a secreted Fc control. After baseline measurements in Opti-MEM media alone, the captured EFN-4 ligand was dipped into tissue culture supernatant from cells transiently transfected with a secreted LAD-2-AP or RIG-3-AP expression plasmid during the association phase (i), then dipped into Opti-MEM media during the dissociation phase (ii). BLI measurements of the Fc control interaction with LAD-2-AP showed no appreciable binding and were used to correct for instrument drift. The negligible dissociation observed in (ii) is consistent with a high affinity interaction between EFN-4 and LAD-2. In contrast, the dissociation observed for RIG-3 reveals that RIG-3 does not bind to EFN-4.