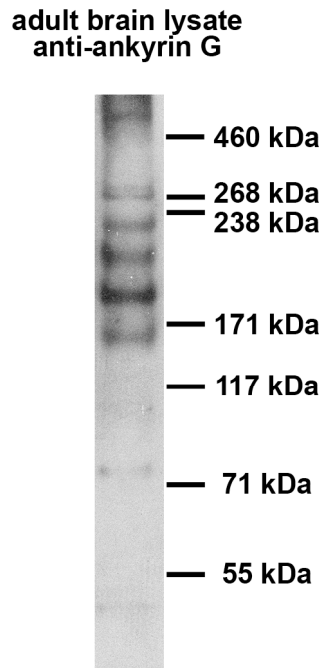
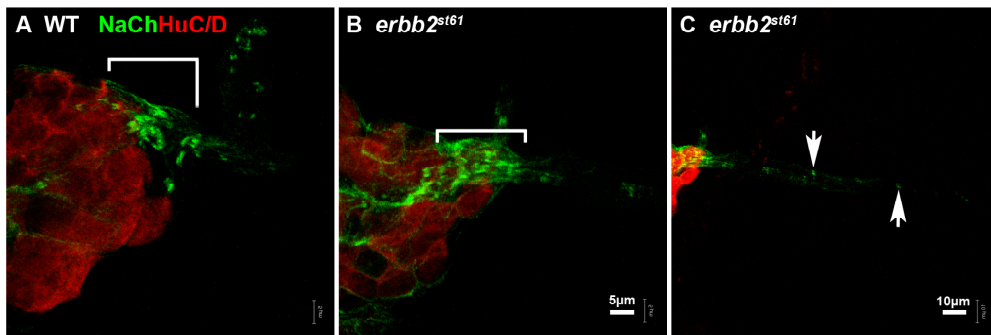


## Figure S1



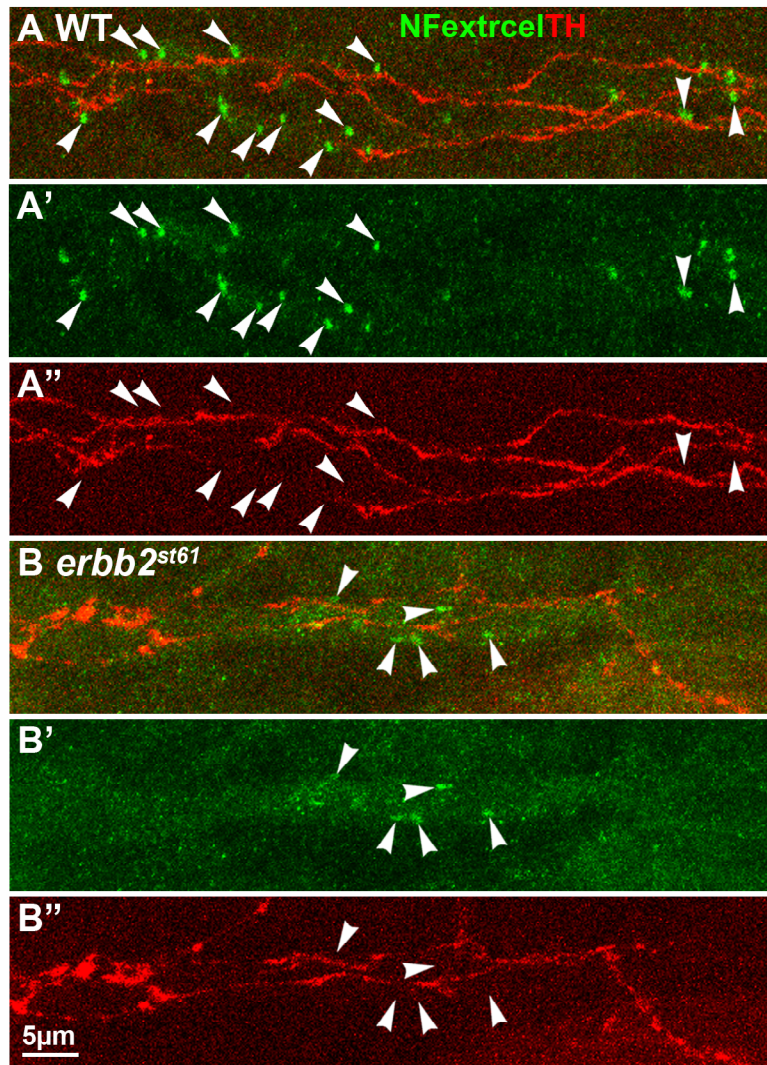
**Figure S1. Immunoblot with anti-ankyrin G antibodies.** In mammals, 480 and 270 kDa isoforms of ankyrin G are expressed in neural tissue, while lower molecular weight isoforms are expressed more broadly (Kordeli et al., 1995). An antibody raised against the spectrin-binding domain of zebrafish ankyrin G recognizes two putative ankyrin G isoforms near 270 kDa, one putative isoform >460 kDa, and a number of smaller putative isoforms in zebrafish brain lysate.

## Figure S2



**Figure S2. Visualization of the PLLn ganglion in *erbB2* mutants.** The neuronal cell bodies of the PLL ganglion are labeled with anti-HuC/D (red). **(A)** In wild type siblings, numerous clusters of sodium channels (anti-panNa<sub>v</sub>Ch in green) can be seen at the base of the PLLn where it joins the ganglion (bracket). These clusters occur at the expected position of the PLLn axon initial segments. **(B)** The ganglion cell bodies (red) and AIS-like clusters (green, bracket) have a similar organization in *erbB2<sup>st61</sup>* mutants. **(C)** The same ganglion and PLLn as in (B) at lower magnification. Sodium channel clusters can be seen many microns away from the ganglion (arrows). Similar results have been seen in *erbB3<sup>st48</sup>* and *cls<sup>t3</sup>* mutants (data not shown).

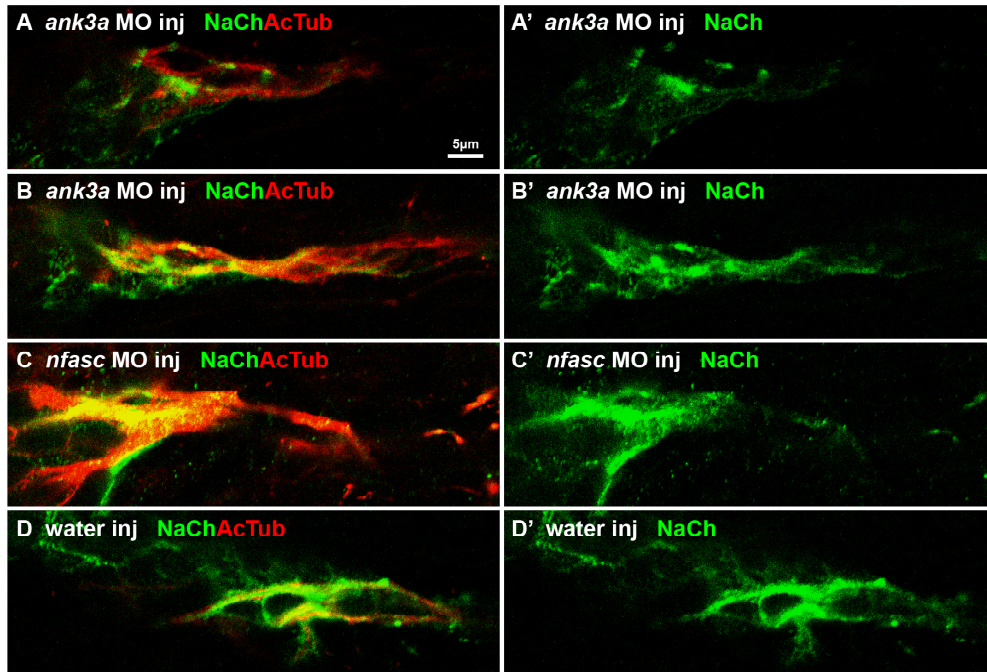
## Figure S3



### Figure S3. Neurofascin clusters are primarily found on afferent axons.

Larvae from *erbb2<sup>st61/+</sup>* intercrosses were labeled with anti-extracellular Neurofascin (green) and anti-Tyrosine Hydroxylase (TH, red), which labels efferent axons. In wild type siblings (**A**) and in *erbb2<sup>st61</sup>* mutants (**B**) most Neurofascin clusters do not overlap with TH-positive axons (e.g. white arrowheads). (**A'**, **B'**) Neurofascin labeling alone. (**A''**, **B''**) TH labeling alone.

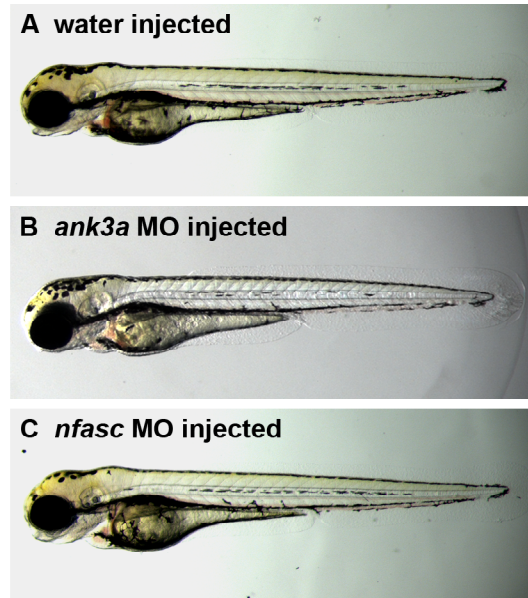
**Figure S4**



**Figure S4. Sodium channel clustering at the AIS is variably reduced in *ank3a* MO injected embryos. (A-B)** In *ank3a* MO injected embryos, sodium channel clustering at the AIS varied from strongly reduced (A) to mildly reduced (B). Sodium channels are labeled green and acetylated tubulin is labeled red. **(C)** Sodium channel clustering at the AIS of *nfasc* MO injected embryos was unaffected. **(D)** Sodium channel clustering at the AIS in a water injected control embryo. All embryos are 3 dpf.

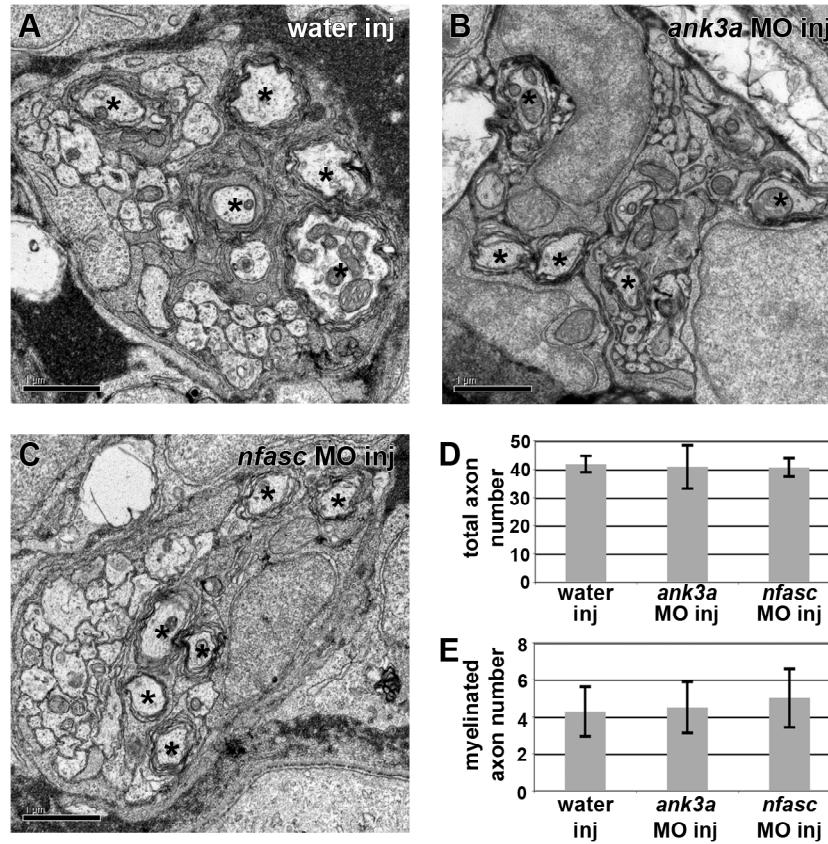


## Figure S5



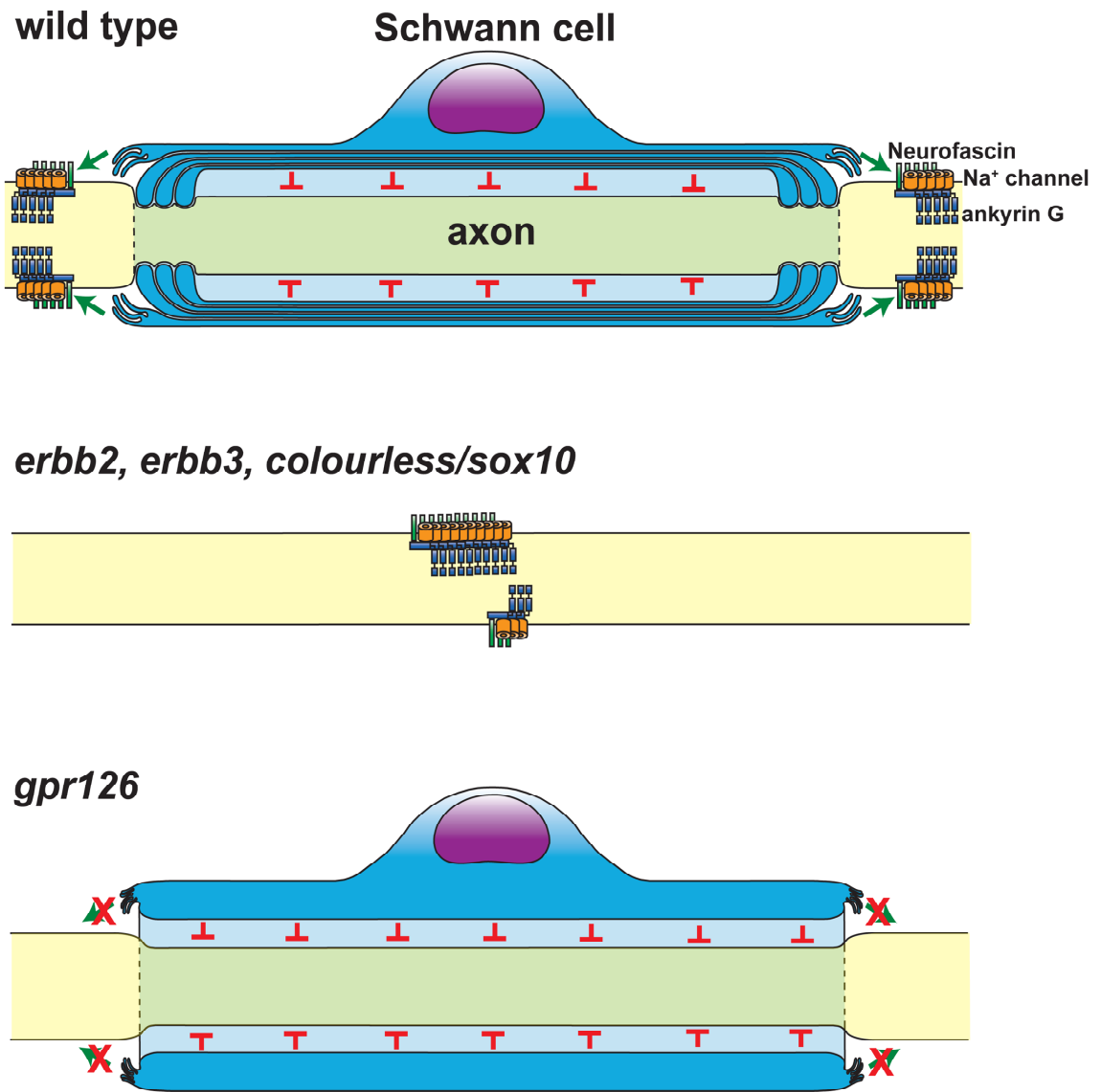
**Figure S5. Normal morphology of embryos injected with *ank3a* MO or *neurofascin* MO.** Embryos injected with water (**A**), *ank3a* MO (**B**), and *neurofascin* MO (**C**) shown here at 80 hpf.

**Figure S6**



**Figure S6. Axon number and myelination are unaffected in the PLL nerves of *ank3a* MO injected and *nfasc* MO injected embryos. (A-C)** Electron micrographs of the PLLn of water injected (A), *ank3a* MO injected (B), and *nfasc* MO injected (C) embryos at 3 dpf. Asterisks indicate myelinated axons. Scale bars represent 1  $\mu$ m. **(D)** Quantification of total axon number in water and morpholino injected embryos. **(E)** Quantification of the total number of myelinated axons in water and morpholino injected embryos. Error bars represent  $\pm$  one standard deviation.

Figure S7



**Figure S7. Schwann cells inhibit axon-intrinsic clustering of sodium channels.** Schematic representation of some molecules involved in sodium channel clusters and mutants analyzed. In peripheral axons, sodium channel clustering at the nodes of Ranvier requires cues from Schwann cells (top, arrows) that act through Neurofascin. In *erbb2*, *erbb3* and *cls/sox10* mutant peripheral nerves, there are no Schwann cells, but clusters form through an

axon-intrinsic mechanism (middle). In *gpr126* mutants, Schwann cells arrest at the promyelinating stage and form very few clusters (bottom). Our interpretation of these data is that Schwann cells inhibit the axon-intrinsic mechanism outside of the nodes and AIS (red Ts). The arrest of myelination in the *gpr126* mutant blocks Schwann cell derived clustering cues (Xs, bottom), but the Schwann cell inhibition of axon-intrinsic clustering is still active.







