

Fig. S1. Controls for overall NMP35 mRNA levels and FISH on DRG neurons. **A**, Despite the shift in axonal levels of NMP35 mRNA with injury-conditioning shown in Fig. 1B,C, by RT-PCR there is no corresponding apparent changes in the overall levels of NMP35 mRNA in the injury-conditioned compared to naïve DRG cultures. **B**, Representative FISH images for scrambled probe (controls) and no-probe controls consistently showed no signal in axons of injury-conditioned neurons in images matched to those in Fig. 1D for exposure, gain, and offset [Scale bars = 10 μ m].

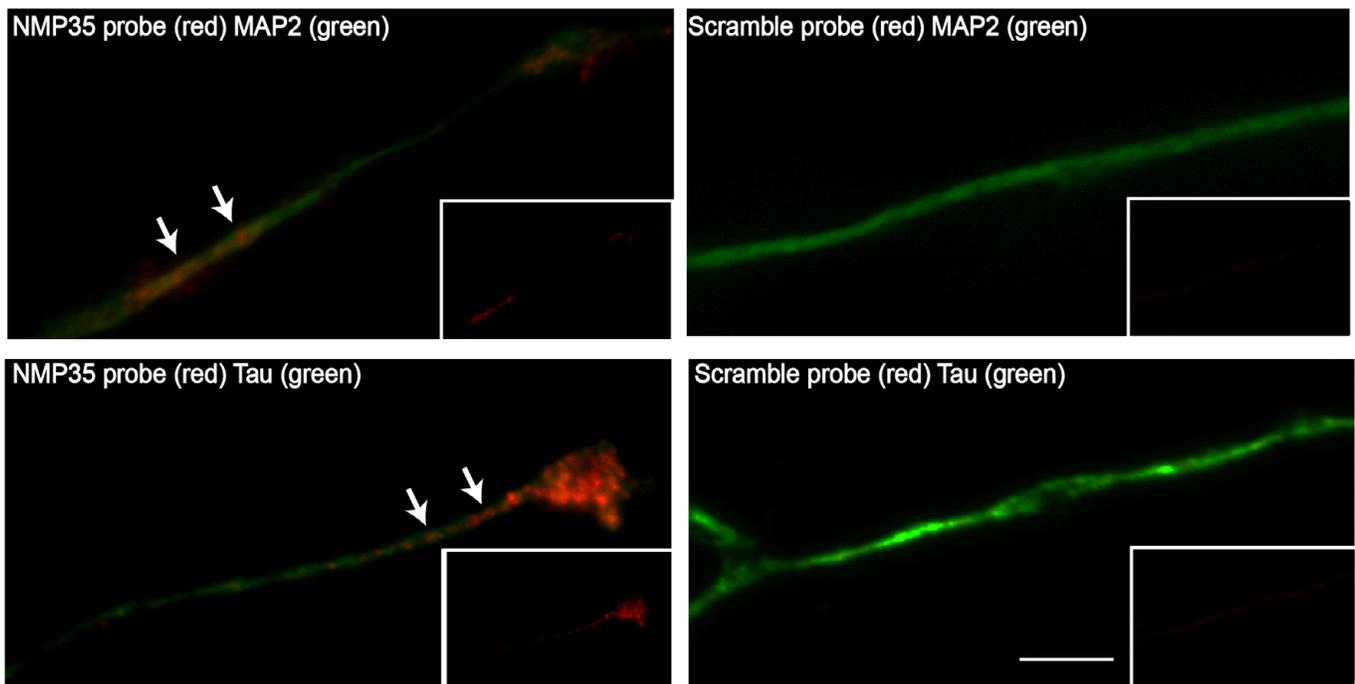


Fig. S2. Endogenous NMP35 mRNA localizes into both axons and dendrites. Cultured rat cortical neurons at 9 days in vitro were processed for FISH/IF for NMP35 mRNA and MAP2 or Tau protein as indicated. NMP35 mRNAs is seen in both axons (lower panels) and dendrites (upper panels, arrows), showing clear granular nature characteristic of transported mRNAs. Inset panels show only the NMP35 mRNA signal. Scramble probe do not show any signals in these processes exposure, gain and offset matched images [Scale bars = 5 μ m].

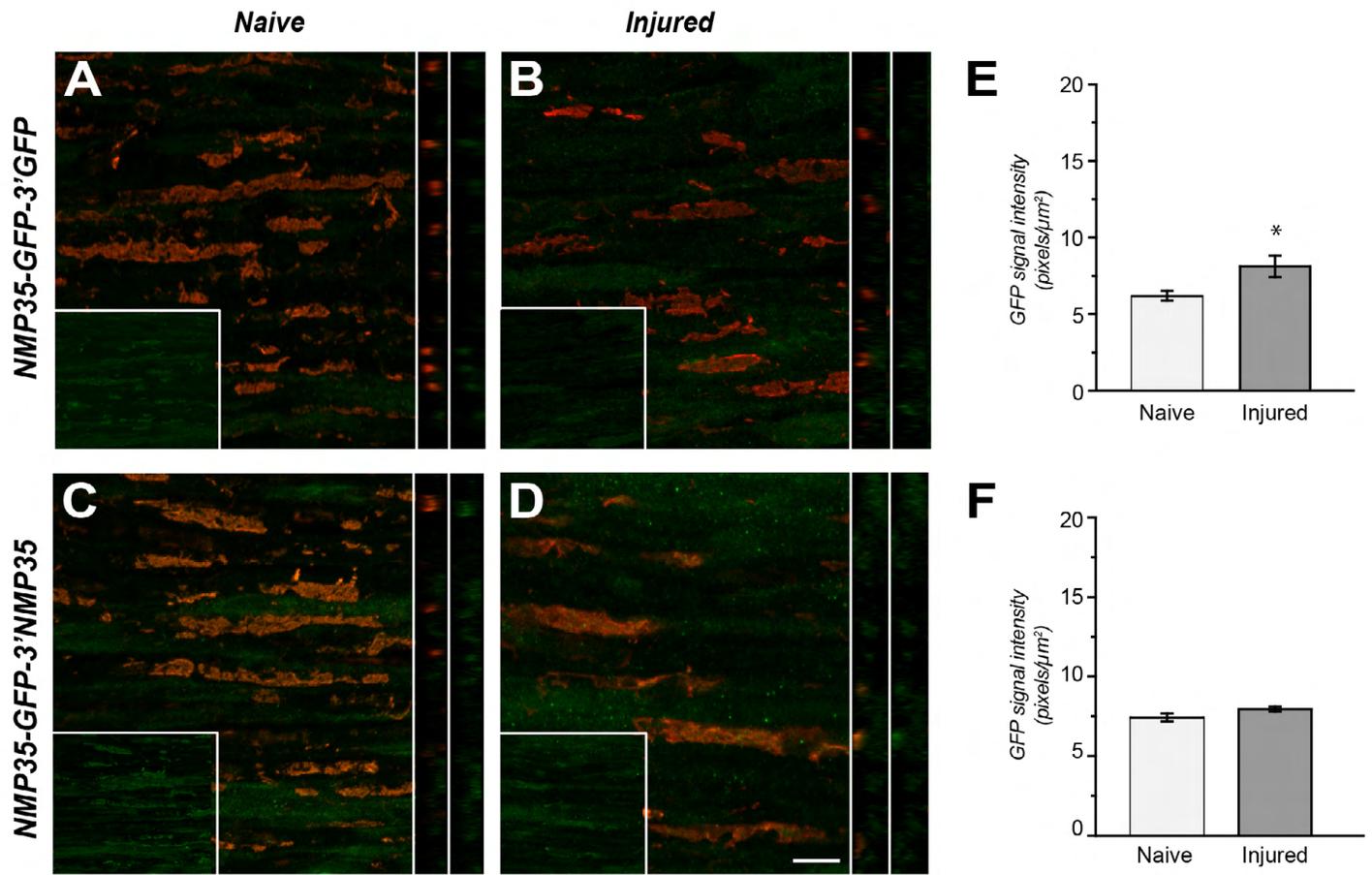


Fig. S3. 3'UTR of NMP35 mRNA is needed for localization into sensory sciatic nerve. L4-5 DRGs were transduced with LV-NMP35-GFP-3'GFP and LV-NMP35-GFP-3'NMP35. Nerve segments taken near the injection site show GFP signals (green) in the Schwann cells for both preparations (A-D). E-F show quantification of the GFP protein intensity that overlapped with NF H signals for sciatic (n \geq 30 from at least 3 separate experiments; * = p \leq 0.05 by student's T test) [Scale bars = 10 μm].

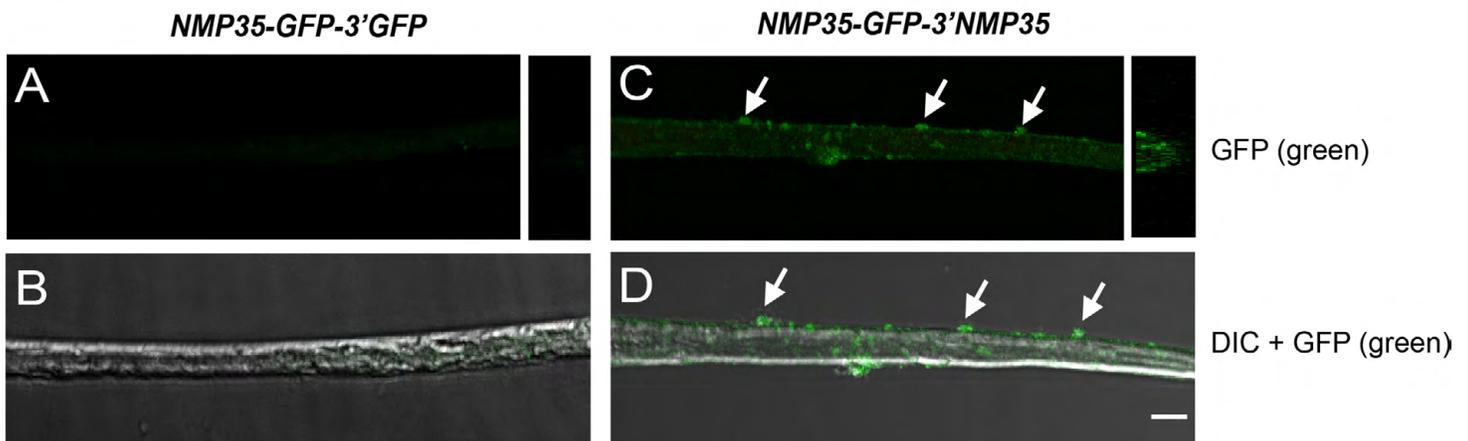


Fig. S4. 3'UTR of NMP35 mRNA localization in the teased sciatic nerve. Teased nerve preparations also confirmed the axonal localization of GFP signals in the LV-NMP35-GFP-3'NMP35 (C,D) transduced animals and lack of signals in the LV-NMP35-GFP-3'GFP transduced animals (A-B). A and C show XYZ reconstructions of GFP signals, with YZ projections in the inset panels; B and D show XYZ projections of GFP signals merged with DIC images. The NMP35-GFP signals are concentrated near the membrane of the axon for the NMP35-GFP-3'NMP35 transduced samples (arrows) [Scale bars = 10 μm].

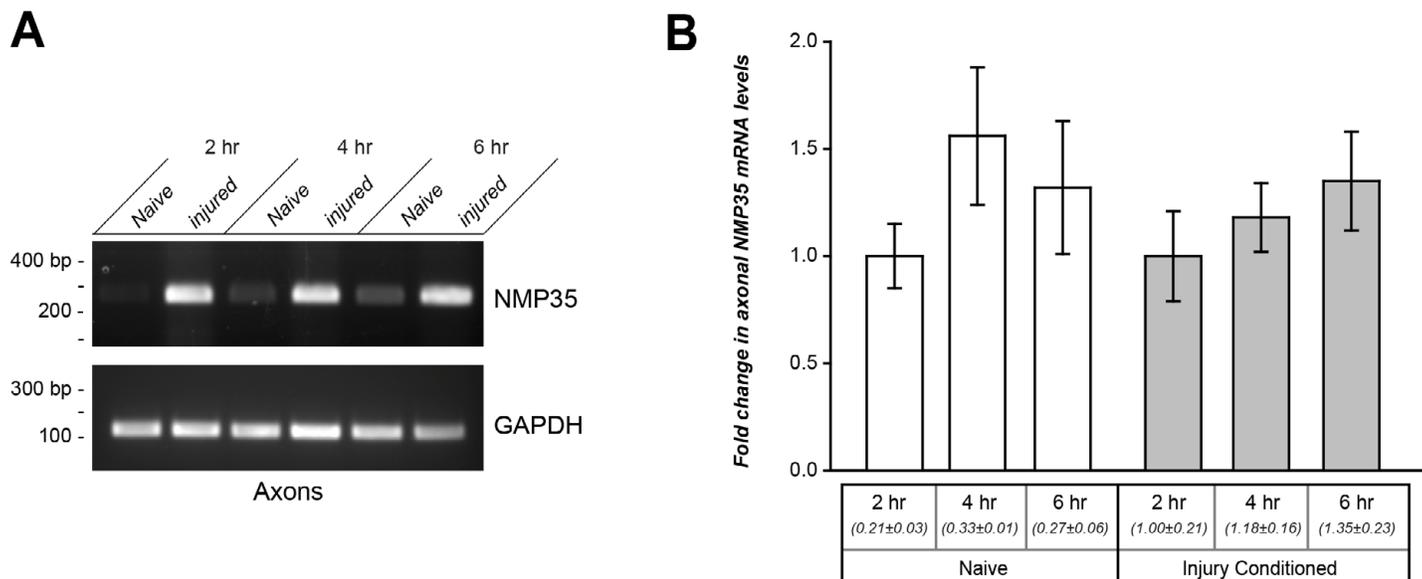
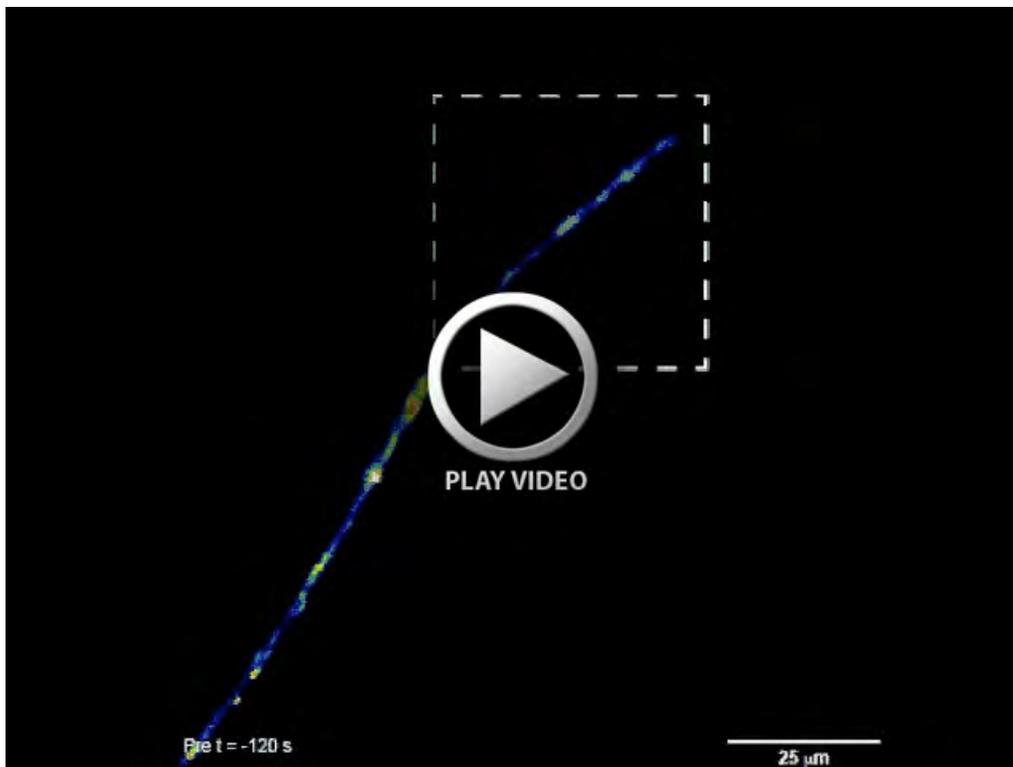


Fig. S5. Stability of NMP35 mRNA in axons. Degeneration of severed axons was delayed by treatment with CsA and NMP35 mRNA levels were measured in axons from overnight cultures of naïve and 7 day injury conditioned DRG neurons. **A** shows standard RT-PCR for NMP35 and GAPDH mRNAs with increased levels of NMP35 mRNA in the axons of injury conditioned compared to naïve neurons. **B** shows results from RTqPCR over three separate experiments where the NMP35 mRNA was normalized to 12S rRNA and expressed as fold change relative 2 hour time point \pm SEM. The signals vary between time points but there is no significant difference between the 2, 4 and 6 hour values for the naïve and injury conditioned sequences. RTqPCR analyses also showed significant more NMP35 mRNA in the axons from injury conditioned than naïve neurons at each time point ($p \leq 0.01$ at 2 hours and $p \leq 0.001$ at 4 and 6 hours by student's T test).



Movie 1. Axonal translation of GFP^{myr3'}NMP35 in DRG neurons. Representative video of FRAP sequence for DRG neurons transfected with GFP^{myr3'}NMP35 is shown. Pre-bleach (2 min.), bleach (1 min.) and post-bleach (30 min.) intervals are shown consecutively as outlined in the methods. The boxed area represents ROI subjected to photobleaching. GFP signals are shown as a spectrum as indicated (see Fig. 3A).



Movie 2. Protein synthesis inhibition prevents translation of GFP^{myr3'}NMP35 in DRG axons. Representative FRAP sequence for GFP^{myr3'}NMP35 transfected DRG neurons as in Suppl. Video 1 where cultures were pretreated with the protein synthesis inhibitor anisomycin (see Fig. 3B).