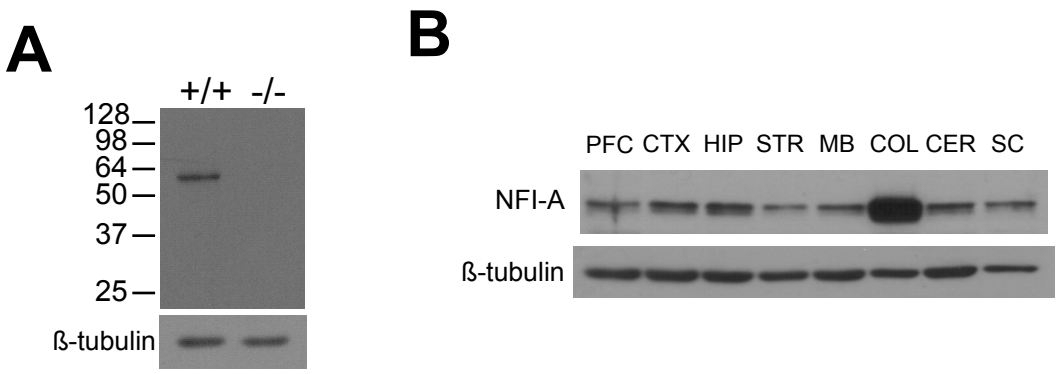
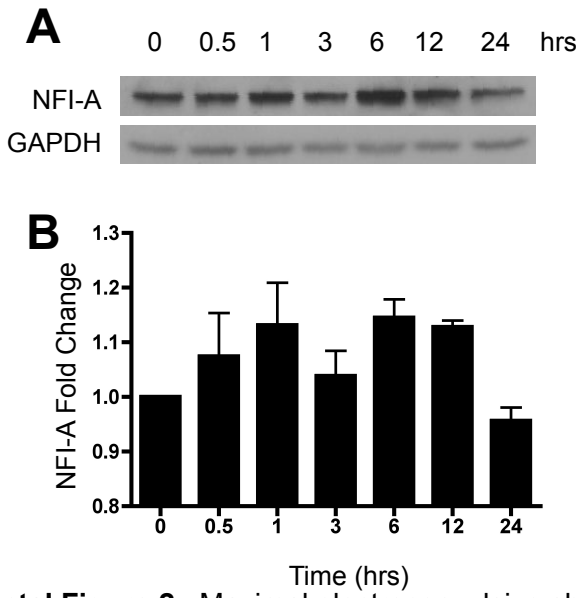
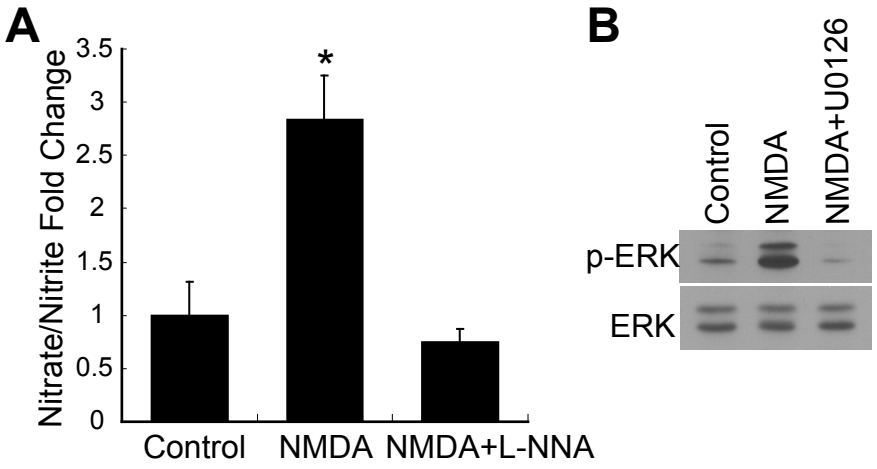


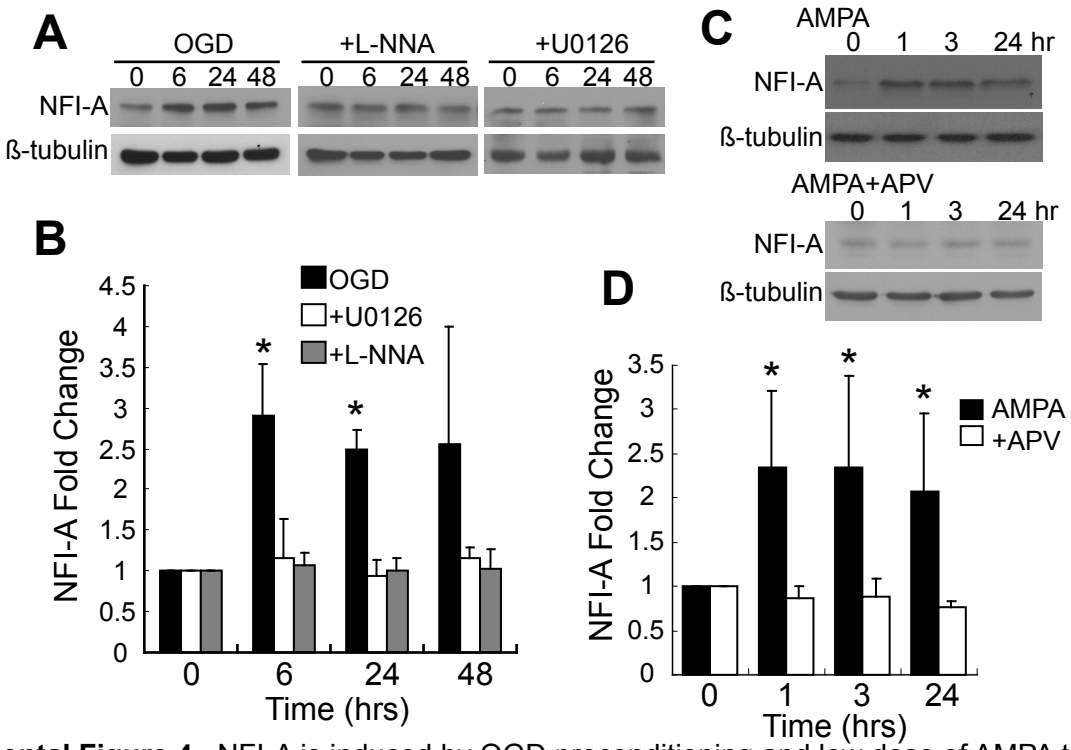
Supplemental Figure 1. (A) Immunoblot analysis of NFI-A polyclonal antibody. Note that the antibody recognizes one specific band at the molecule weight of NFI-A in a Nfia+/+ brain, but not in a Nfia-/- brain. (B) Regions of adult mouse brain express NFI-A. PFC: prefrontal cortex, CTX: cortex, HIP: hippocampus, STR: striatum, MB: midbrain, COL: colliculus, CER: cerebellum, SC: spinal cord.



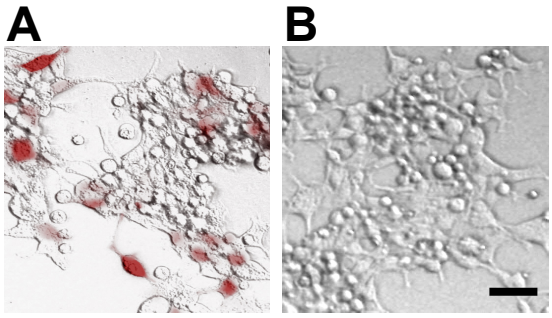
Supplemental Figure 2. Maximal electroconvulsive shock (MECS) transiently induces NFI-A with kinetics similar to in vitro NMDA stimulation. (A) Mouse hippocampi were dissected at indicated times following treatment with MECS and lysates subjected to western analysis. (B) Quantitation of western blots performed three biological replicates at each time point.



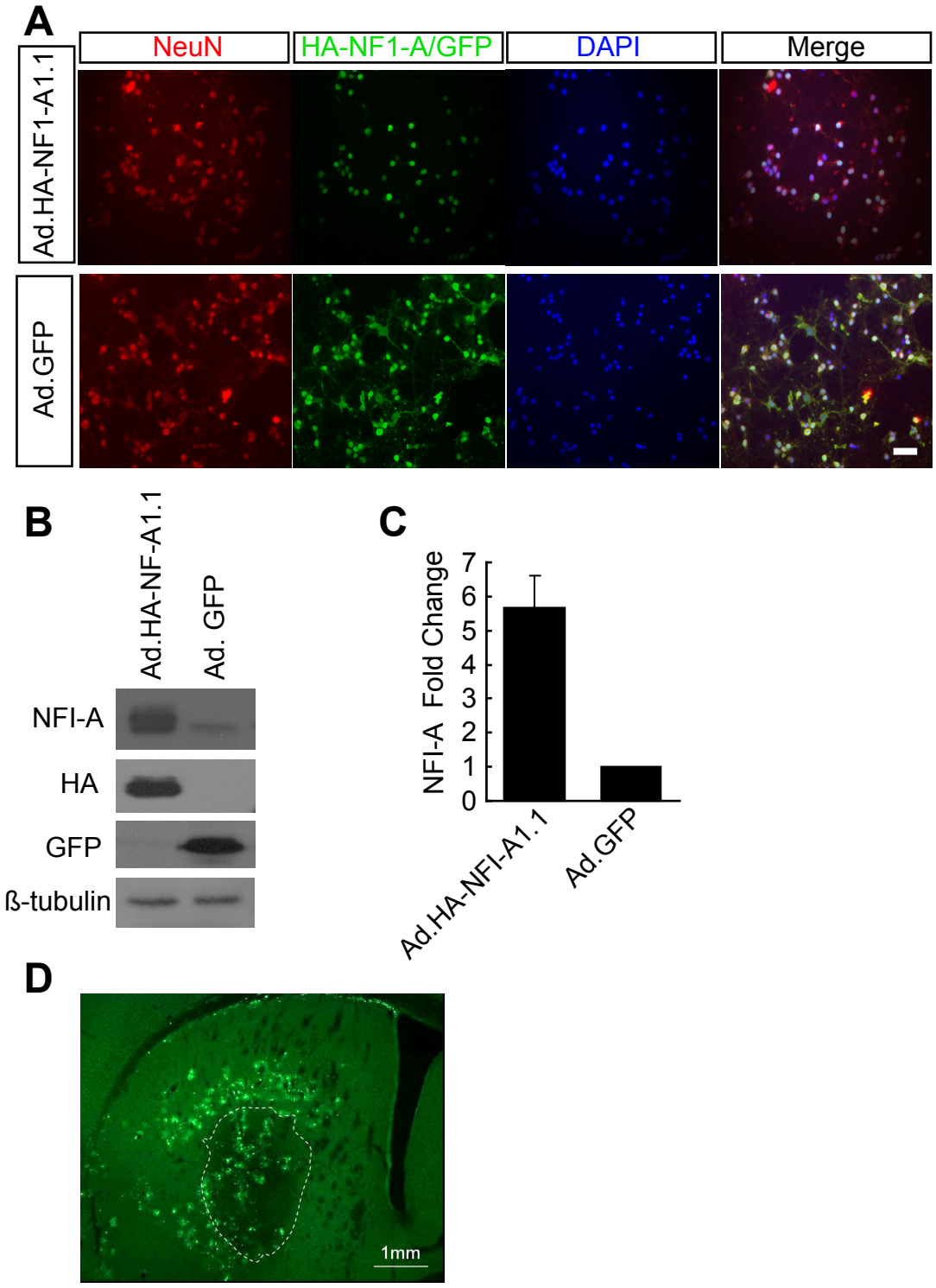
Supplemental Figure 3. Efficacy of NOS and MEK inhibition. (A) Nitric Oxide quantification assay (Active Motif, Inc) shows that the NOS inhibitor nitro-L-arginine (L-NNA, 100 mM) significantly inhibits (50 μM) NMDA induced NO production. Cortical cultures were harvested at 5 minutes after the NMDA treatment. L-NNA is incubated 1 hour before and during NMDA treatment. Significance was determined at * $p<0.01$ by one-way ANOVA followed by Tukey Kramer post hoc test. (B) Immunoblot analysis shows that the MEK inhibitor U0126 (50 mM) significantly blocks (50 mM) NMDA induced ERK phosphorylation. U0126 is incubated 1 hour before and during NMDA treatment. Cortical cultures were harvested at 5 minutes after the NMDA treatment.



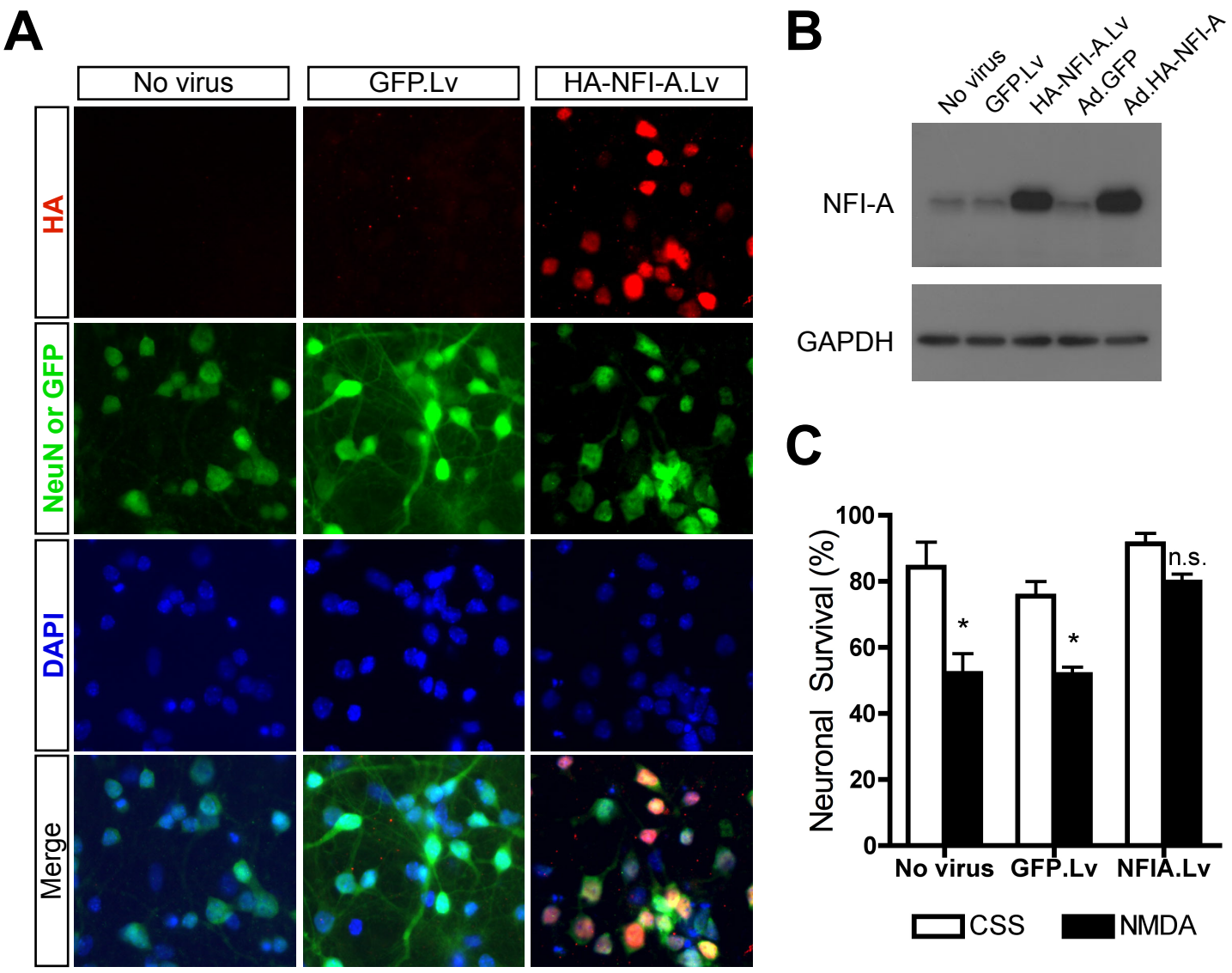
Supplemental Figure 4. NFI-A is induced by OGD preconditioning and low dose of AMPA treatment. (A) NFI-A induction profiles upon OGD preconditioning treatment (15 min) with or without the NOS inhibitor, L-NNA, or the MEK inhibitor, U0126, in cortical cultures. Cultures were harvested at 6, 24 and 48 hours post treatment. (B) Quantification of NFI-A induction levels from three independent experiments show that OGD preconditioning induced NFI-A expression is blocked by the MEK inhibitor U0126 or the NOS inhibitor L-NNA. Significance was determined at * $p<0.01$, one-way ANOVA followed by Tukey Kramer post hoc test, compared to NFI-A levels from preconditioned cortical cultures treated with U0126 or L-NNA. This experiment was replicated three times. (C) NFI-A induction profiles upon AMPA treatment (25 μM, 5 min) with or without NMDA receptor antagonist APV (250 μM) in cortical cultures. Cultures were harvested at 1, 3 and 24 hours post treatment. (D) Quantification of NFI-A induction levels from three independent experiments show that AMPA induced NFI-A expression is completely blocked by NMDA receptor antagonist APV. Significance was determined at * $p<0.05$, one-way ANOVA followed by Tukey Kramer post hoc test, compared to NFI-A levels from cortical cultures treated with AMPA and APV. This experiment was replicated three times.



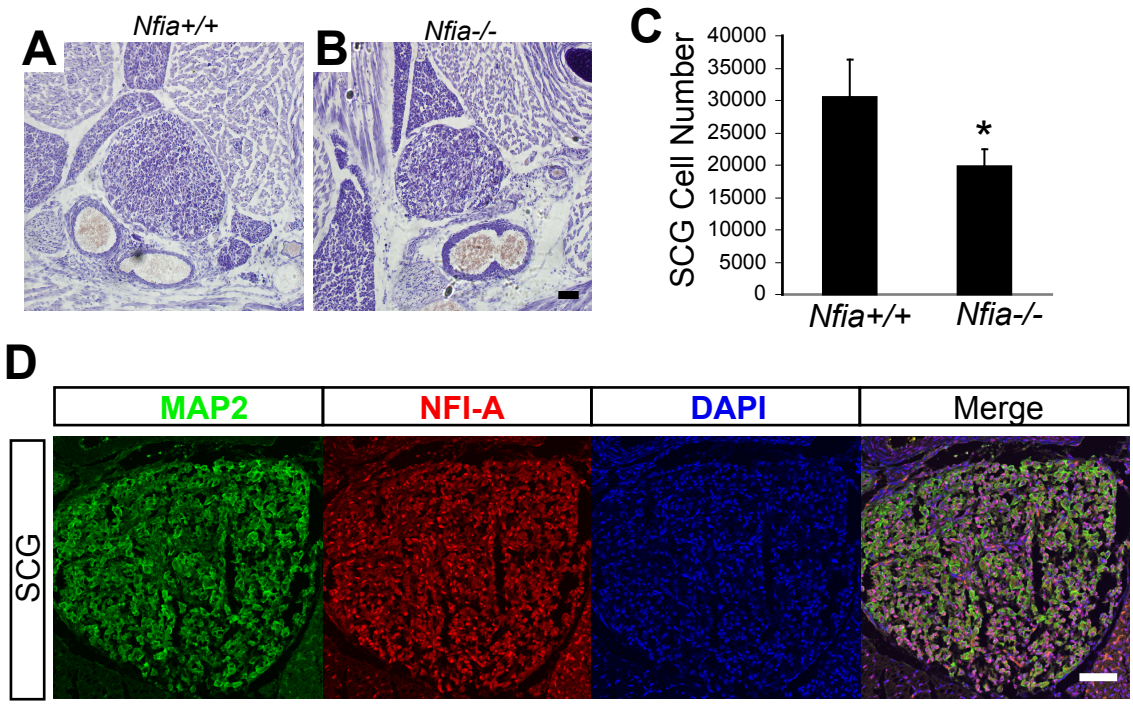
Supplemental Figure 5. DsRed siRNA molecules effectively knock-down exogenous DsRed protein expression. pCMV-DsRed-Express expression plasmid was co-transfected with SCR siRNA molecules (A) or DsRed siRNA molecules (B), n=3. 24 hours later, DsRed expression was monitored under fluorescence microscope. Neurons were identified by Hoffman modulation. Scale bar = 50 μm.



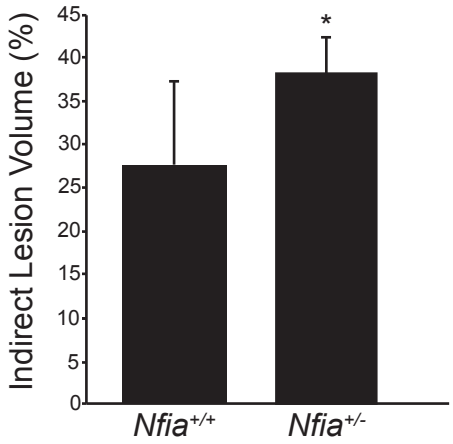
Supplemental Figure 6. Expression of NFI-A or GFP by adenovirus in cortical cultures. (A) Immunocytochemistry shows that neurons are transduced at a high-percentage rate (~ 80%) by both HA-NFI-A adenovirus and GFP adenovirus. Scale bar = 50 µm. (B) Immunoblot analysis shows over expression of both HA-NFI-A adenovirus and GFP adenovirus in cortical cultures. The NFI-A adenovirus expresses HA-tagged NFI-A, which is recognized by the anti-HA antibody. (C) Quantification of immunoblot data generated by laser densitometry showing an average of 5-6 fold over expression of HA-NFI-A by adenovirus transduction. (D) Low-power micrograph demonstrating in vivo expression of Ad.HA-NFI-A1.1 (anti-HA, green) following NMDA injection. Dashed line outlines the lesioned region of the striatum.



Supplemental Figure 7. Lentiviral expression of NFI-A slectively in neurons induces neuroprotection. (A) Immunostaining for GFP demonstrate selective expression of GFP.Lv in cells with neuronal morphology while co-localization of HA-NFI-A.Lv with NeuN+ neurons definitively demonstrates neuronal slectivity of the virus. (B) Western analysis with anti-NFI-A antisera demonstrates that HA-NFI-A.Lv expresses NFI-A at comparable levels to Ad.HA-NFI-A1.1. (C) HA-NFI-A.Lv imparts a significant neuroprotective phenotype to primary cortical neuron treated with 500 μM NMDA (black bars) while CSS treatment alone does not demonstrate significant decreases in viability (open bars). * indicates $P < 0.05$ two-way ANOVA with Bonferroni post-test, N=5.



Supplemental Figure 8. Nfia-/- SCG neurons are depleted in vivo. (A-B) Nissl staining of superior cervical ganglion (SCG) neurons in (A) Nfia+/+ or (B) Nfia-/- mice at P0.5. Scale bar = 100 μm. (C) Quantification of cell counts for Nfia+/+ or Nfia-/- SCG. n=4, Significance was determined at *p<0.05 by the Student's t-test. (D) Immunostaining for NFI-A in SCG sections shows that NFI-A is expressed in SCG neurons at P0.5. MAP2 staining is used as a neuronal marker. Scale bar = 100 μm.



Supplemental Figure 9. Indirect lesion volumes from Nfia+/+ and Nfia+/- mice following intrastriatal NMDA injection. Lesion volumes are expressed as a percentage of total striatal volume. * $P < 0.05$, Student's t-test, N=6).