

Inhibition of MNK1/2-eIF4E Signaling Alleviates Pain and Reduces Ectopic Bone Formation in Trauma-Induced Heterotopic Ossification

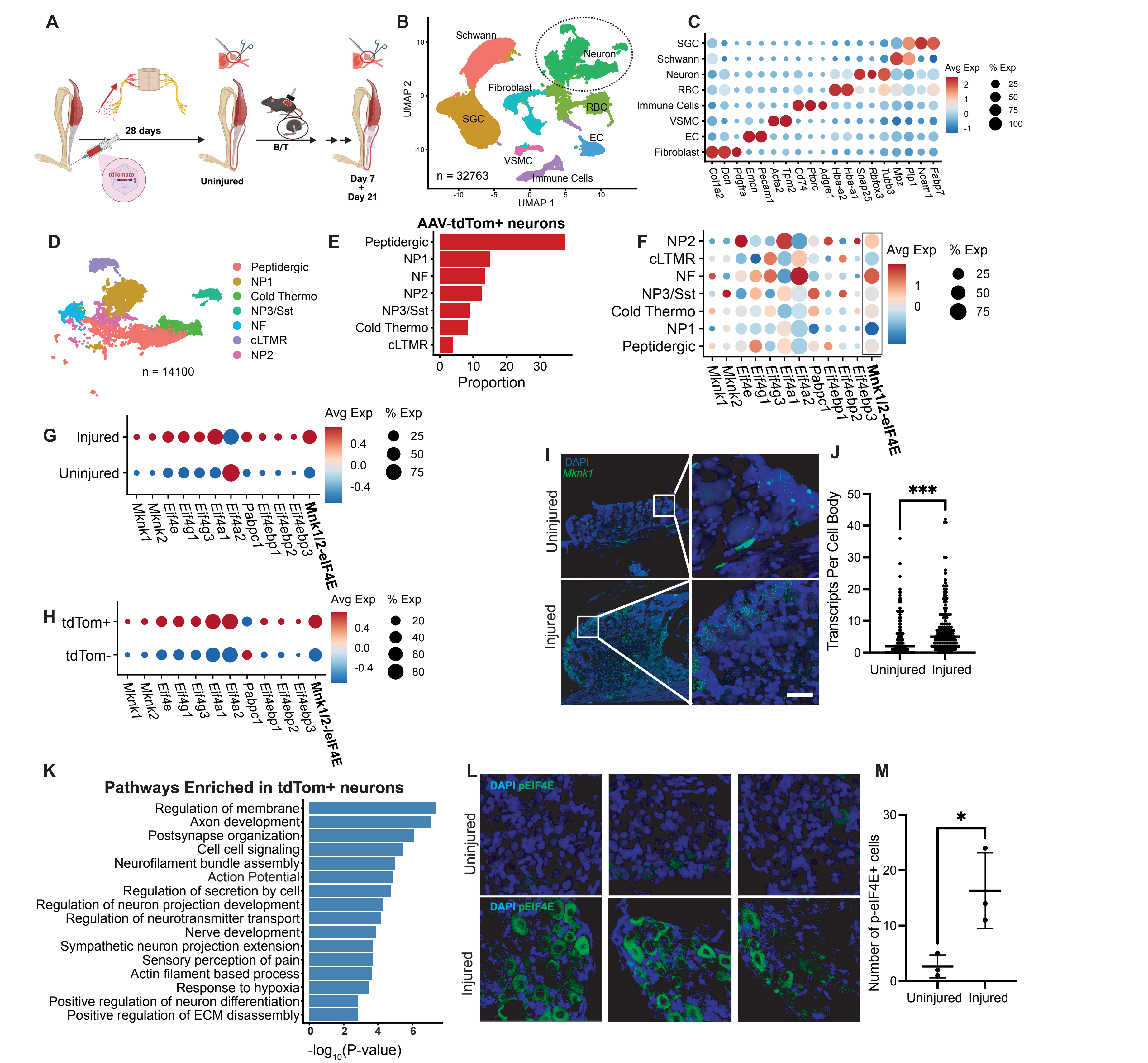
¹Monisha Mittal, ¹Madysen Hunter, ¹Neda Vishlaghi, ³Andi Wangzhou, ¹Sneha Korlakunta, ¹Badarigoda N. Somatilaka, ¹Chase Pagani, ¹Yuxiao Sun, ²Manyu Zhu, ²Masnsen Cherief, ²Zhao Li, ¹Heeseog Kang, ²Mingxin Xu, ²Aaron W. James, ³Theodore J. Price, and ¹Benjamin Levi
¹UT Southwestern Medical Center, Dallas, TX ; ²Johns Hopkins University, Baltimore, MD; ³University of Texas at Dallas, Richardson, TX

BACKGROUND

- Trauma-induced heterotopic ossification (HO) is a debilitating condition where ectopic bone forms in soft tissue following injury.
- HO can severely restrict joint movement, cause chronic pain, and hamper recovery in warfighters and civilian trauma patients alike.
- HO-related pain is multifactorial, involving inflammatory, mechanical, and neural mechanisms.
- NGF–TrkA signaling in sensory neurons is a key contributor to HO development and pain.
- MNK1/2 are key regulatory enzymes that drive inflammatory signaling and nociceptor sensitization by phosphorylating the eIF4E.

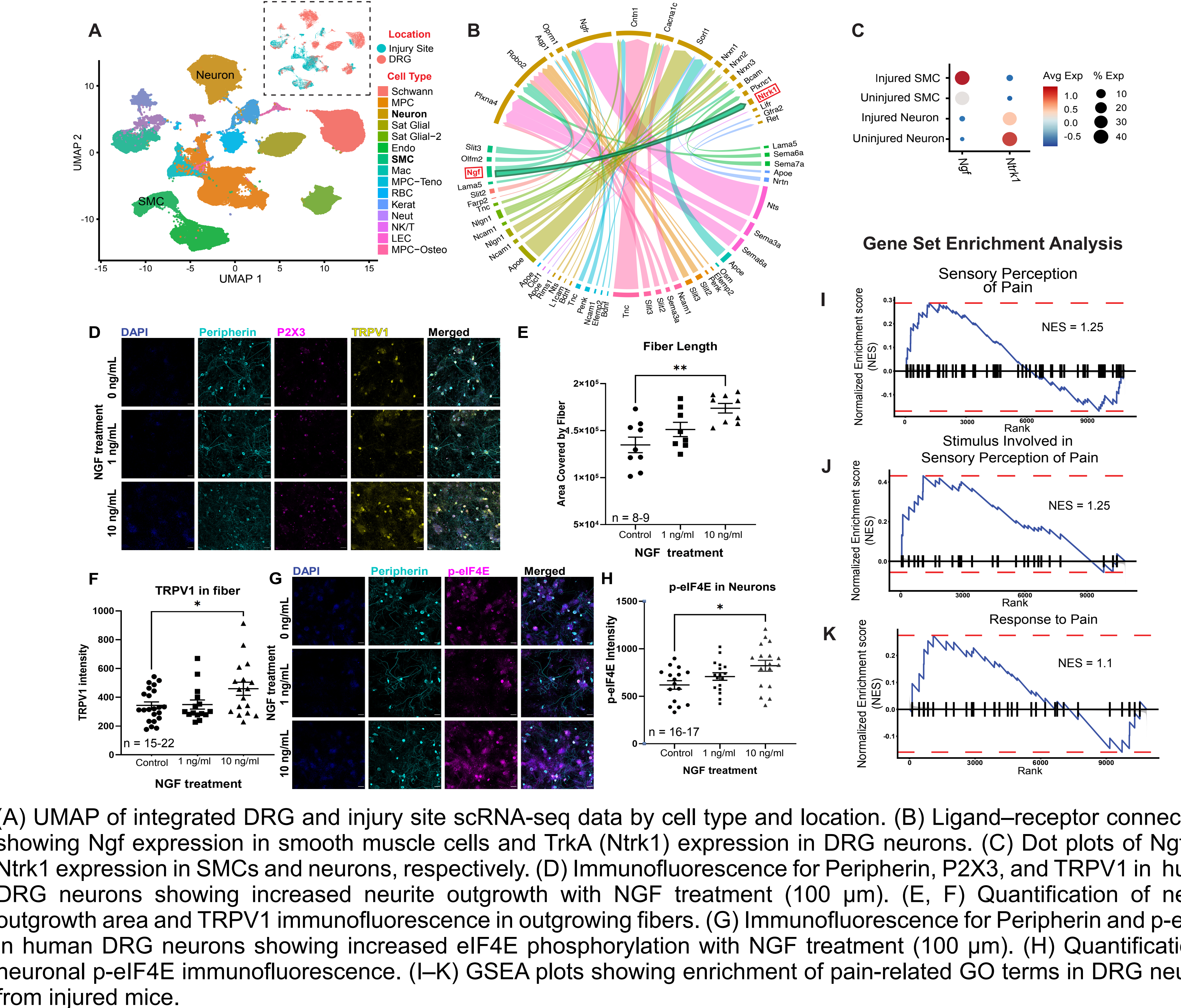
RESULTS

Injury induces the MNK1/2–eIF4E signaling axis in sensory neurons innervating the HO site

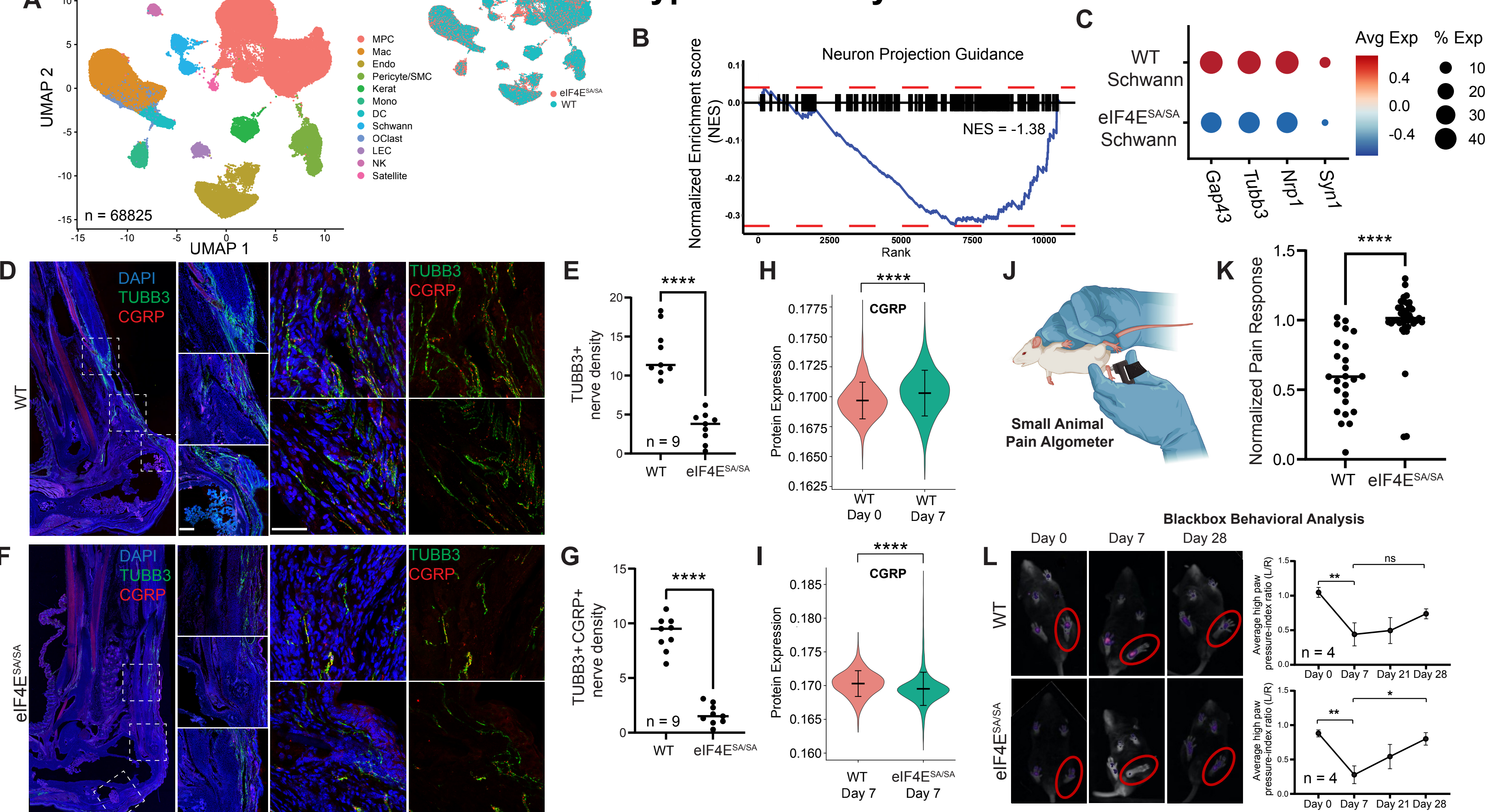


(A) Schematic of DRG collection 7 and 21 days post-B/T. (B) UMAP of all DRG cells with neuron cluster highlighted. (C) Dot plot of canonical marker genes across DRG cell clusters. (D) UMAP of neuron cluster showing neuronal subtypes. (E) Proportion of AAV-tdTom+ neurons by cluster. (F) Dot plot of MNK1/2–eIF4E signaling transcript expression across neuronal clusters. (G, H) Dot plots showing MNK1/2–eIF4E signaling transcript upregulation in neurons and tdTom+ neurons post-injury. (I, J) RNAScope validation of Mknk1 upregulation in injured vs. uninjured DRG neurons (n = 150 cell bodies/condition; 50 μ m). (K) GO term enrichment in tdTom+ neurons. (L, M) Representative immunofluorescence images and quantification of phosphorylated eIF4E (p-eIF4E) in DRGs 7 days post-B/T, showing increased expression after injury (n = 3; 50 μ m).

NGF–TrkA signaling activates the MNK1/2–eIF4E signaling axis and drives a persistent nociceptive program



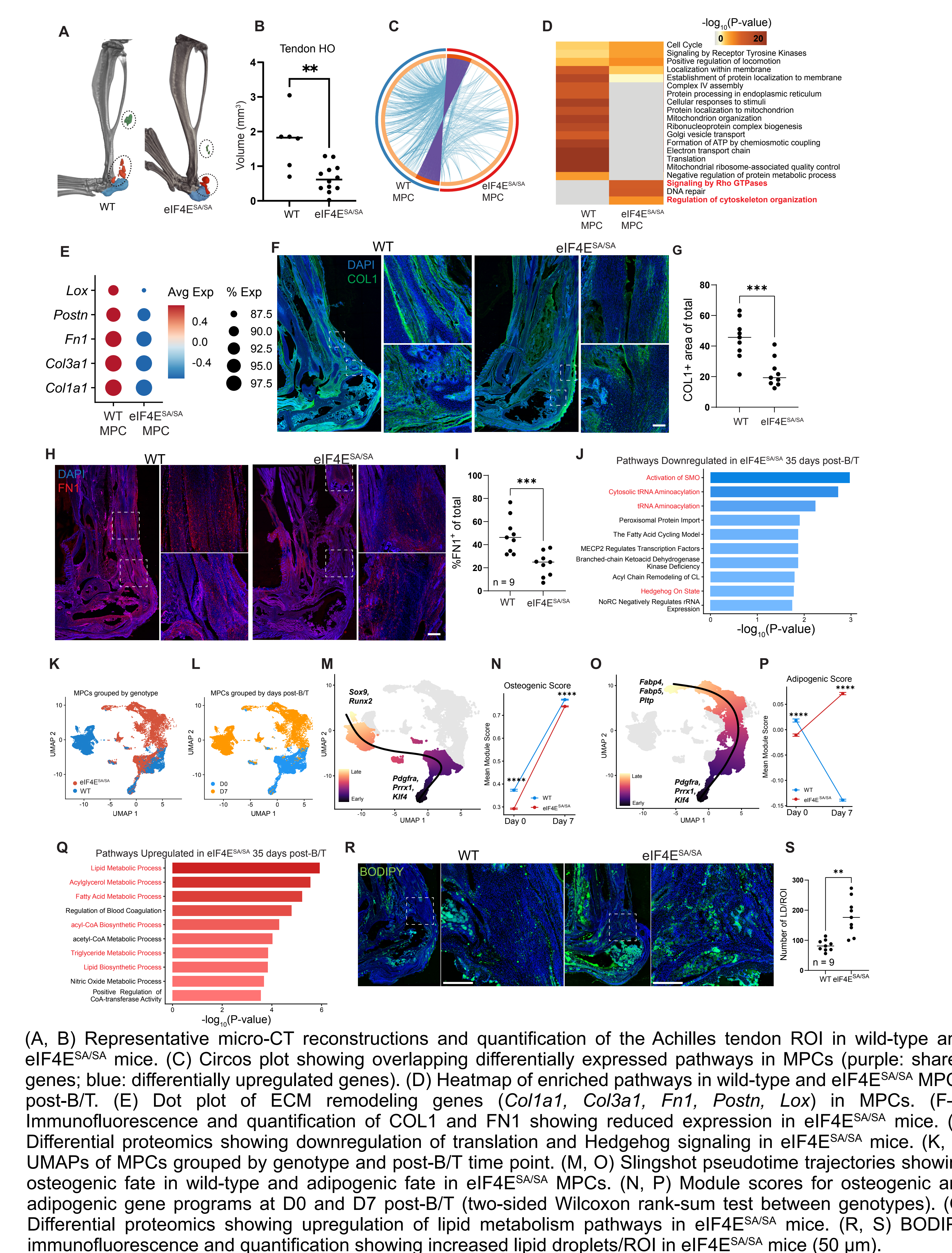
MNK1/2–eIF4E signaling axis inhibition reduces neural programs, axonal density and pain hypersensitivity



(A) UMAP of injury-site scRNA-seq from wild-type and eIF4E^{SA/SA} mice. (B) GSEA showing downregulation of neuron projection development in eIF4E^{SA/SA} Schwann cells. (C) Dot plot of cytoskeletal (*Gap43*, *Tubb3*), axon guidance (*Nrp1*), and synaptic (*Syn1*) genes in Schwann cells. (D–G) Immunofluorescence and quantification of β III-tubulin (TUBB3) and CGRP at the injury site 9 weeks post-B/T, showing reduced axonal staining and nerve density in eIF4E^{SA/SA} mice (200, 50 μ m). (H, I) Violin plots of scTranslator-predicted CGRP protein expression in uninjured vs. injured wild-type mice and eIF4E^{SA/SA} vs. wild-type mice at 7 days post-B/T. (J) Schematic of Small Animal Algometer testing for pressure hypersensitivity. (K) Pain thresholds 9 weeks post-B/T showing reduced hypersensitivity in eIF4E^{SA/SA} mice. (L) Blackbox behavioral analysis showing increased pain at day 7 in both groups and faster recovery in eIF4E^{SA/SA} mice by day 28 (2-way ANOVA).

RESULTS

Loss of MNK1/2–eIF4E signaling reduces ectopic bone formation, suppresses reparative programs, and osteogenic lineage commitment



CONCLUSIONS

- HO induces MNK1/2–eIF4E signaling in sensory neurons innervating the injury site
- NGF–TrkA activation of MNK1/2–eIF4E sustains nociceptive signaling.
- MNK1/2–eIF4E inhibition reduces axonal density and pain hypersensitivity.
- MNK1/2–eIF4E inhibition suppresses ectopic bone formation, reparative programs, and shifts osteogenic toward adipogenic lineage commitment.