FACTOR 2025
June 27, 2025

Sensory neurons regulate osteosarcoma disease progression

Sowmya Ramesh, PhD

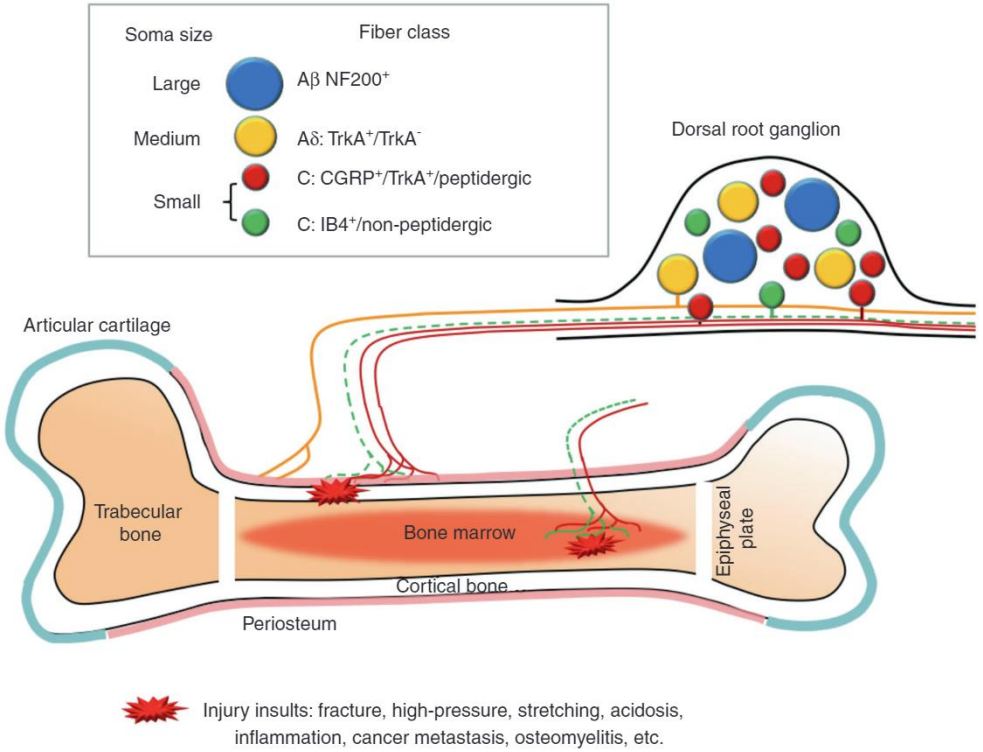
Qizhi Qin, Masnsen Cherief, Mary Archer, Lingke Zhong, Ankit Uniyal, Devadutta Balaji, Zhao Li, Xin Xing, Neelima Thottappillil, Mario Gomez-Salazar, Mingxin Xu, Manyu Zhu, Leslie Chang, Khadijah Mazhar, Monisha Mittal, Edward F McCarthy, Carol D Morris, Benjamin Levi, Yun Guan, Thomas L Clemens, Theodore J Price, Aaron W James

Department of Pathology, Johns Hopkins University; Baltimore, MD 21205
Department of Anesthesiology and Critical Care Medicine, Johns Hopkins University; Baltimore, MD 21205
Department of Neuroscience, Center for Advanced Pain Studies, University of Texas at Dallas; Dallas, TX 75080
Department of Surgery, University of Texas Southwestern; Dallas TX 75390
Department of Neurological Surgery, Johns Hopkins University; Baltimore, MD 21205
Department of Surgery, Orthopaedic Service, Memorial Sloan Kettering Cancer Center; NY, New York 10065
Department of Orthopaedics, University of Maryland School of Medicine; Baltimore, MD 21201
Research and Development Service, The Baltimore Veterans Administration Medical Center; Baltimore, MD 21201

Skeletal Innervating Neurons

- Majority of bone-innervating sensory fibers are unmyelinated, CGRP + /TrkA + peptidergic C-fibers (red)

- NGF binds to tropomyosin-receptor-kinase A (TrkA) on sensory nerve terminals to transmit nociceptive signals

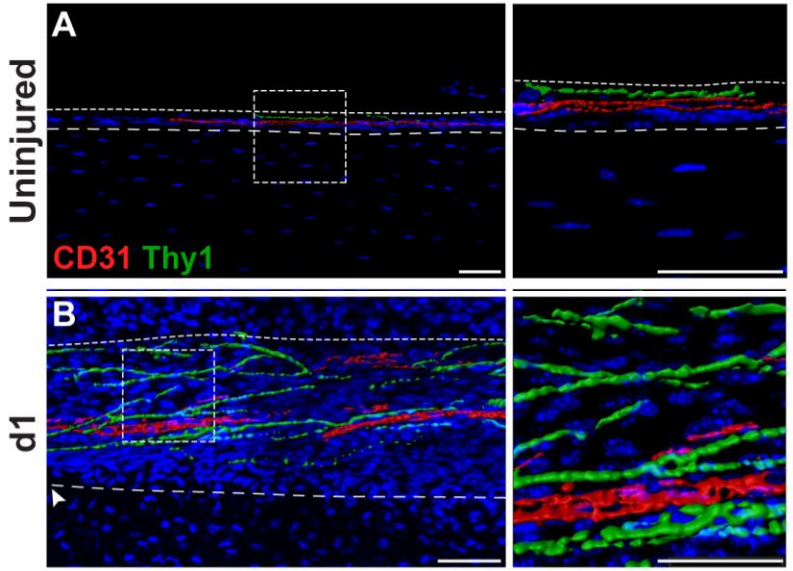


Nerve Growth Factor – NGF
TrkA tyrosine kinase receptor type 1

Zhen G, et. al. Bone Res. 2022 Jun 6;10(1):44.
Mantyh, Patrick W. *European journal of Neuroscience* 39.3 (2014): 508-519.
Pezet, Sophie, and Stephen B. McMahon. *Annu. Rev. Neurosci.* 29 (2006): 507-538.

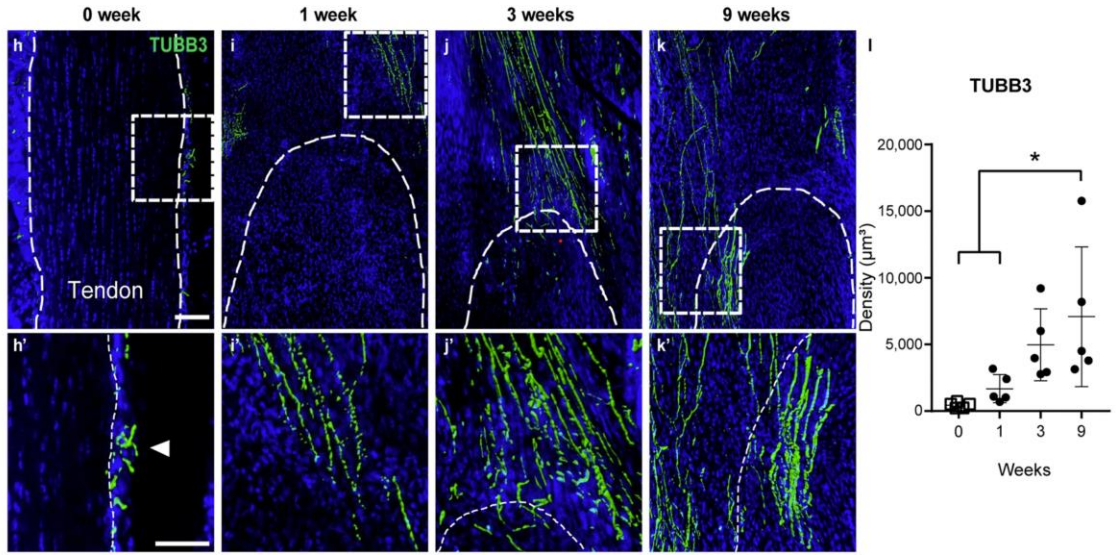
Bone is innervated by NGF–responsive TrkA sensory nerve fibers

Fracture repair requires TrkA signaling by skeletal sensory nerves



J Clin Invest. 2019;129(12):5137–5150

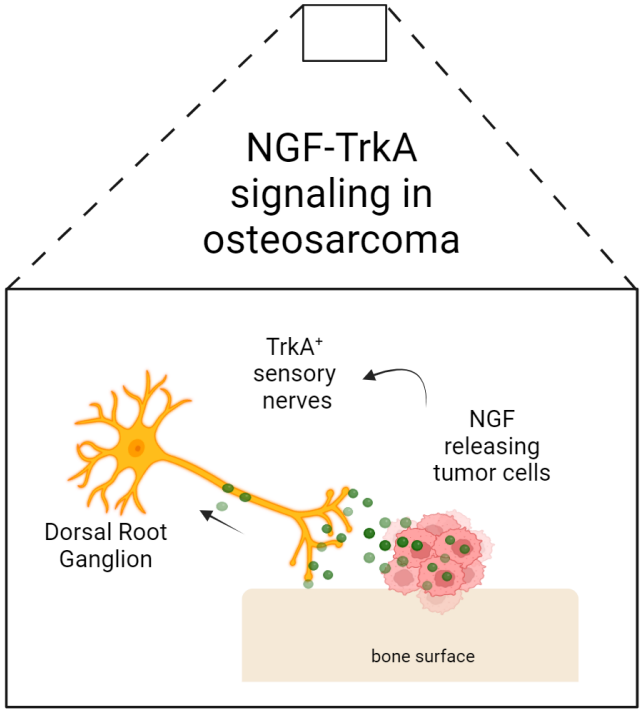
NGF-TrkA signaling dictates neural ingrowth and heterotrophic ossification progression



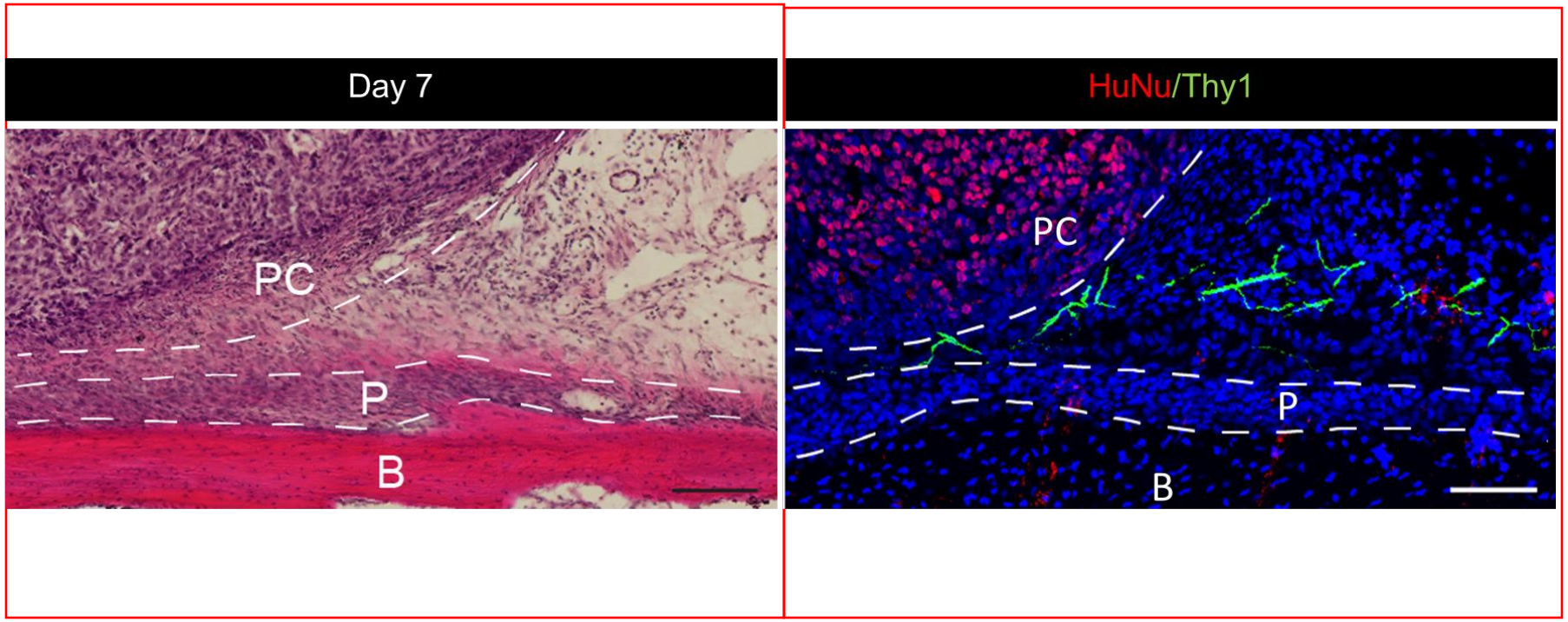
Nat Commun 12, 4939 (2021)

Hypothesis

Neuron-to-sarcoma signaling is a positive regulator of osteosarcoma disease progression

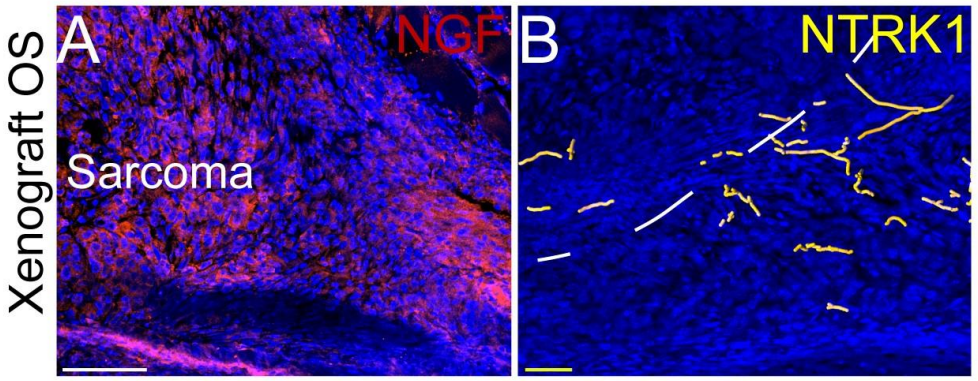


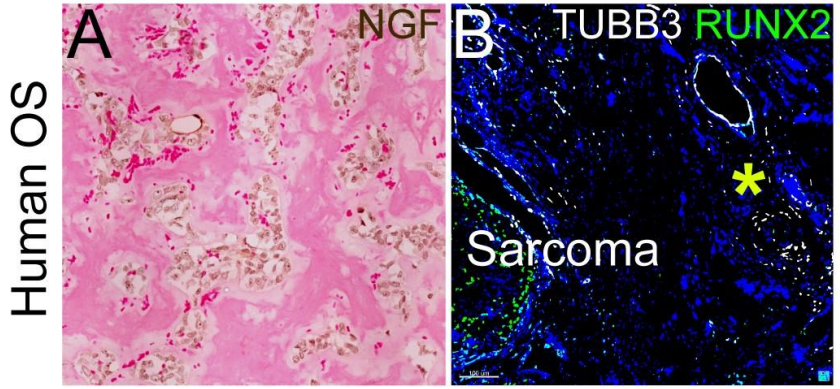
An orthotopic model of human osteosarcoma



Thy1-YFP; *Scid* mice
143B human osteosarcoma cells

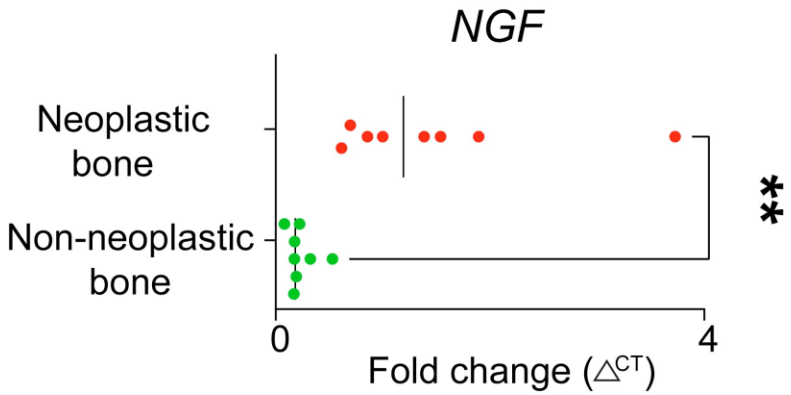
NGF-TrkA signaling dictates neural ingrowth in osteosarcoma



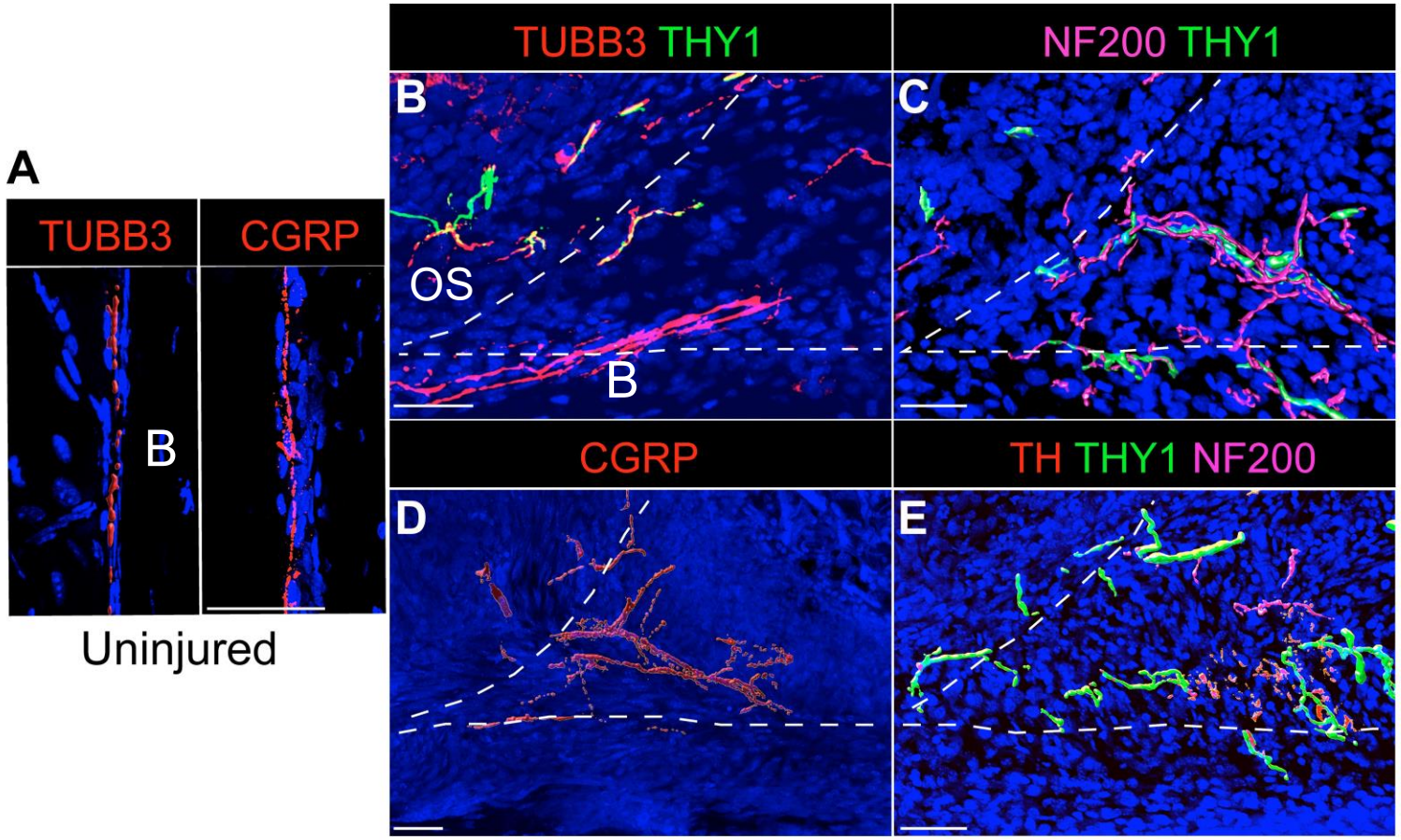


Human OS cell lines[1]

Primary human OS samples[2]

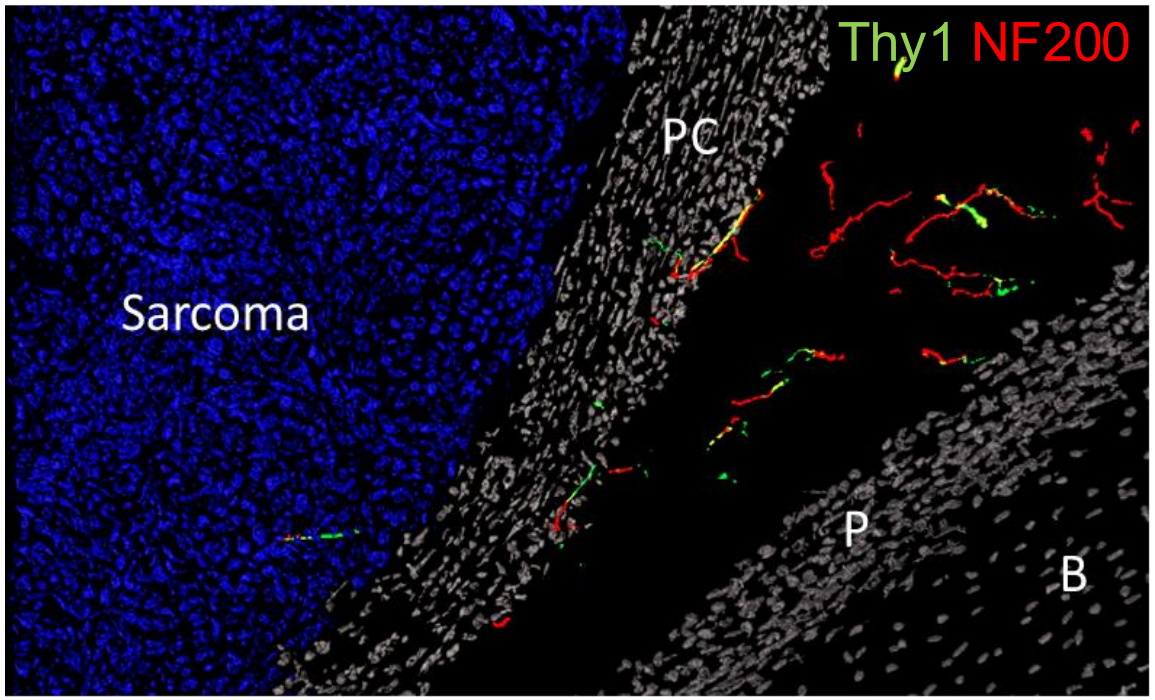


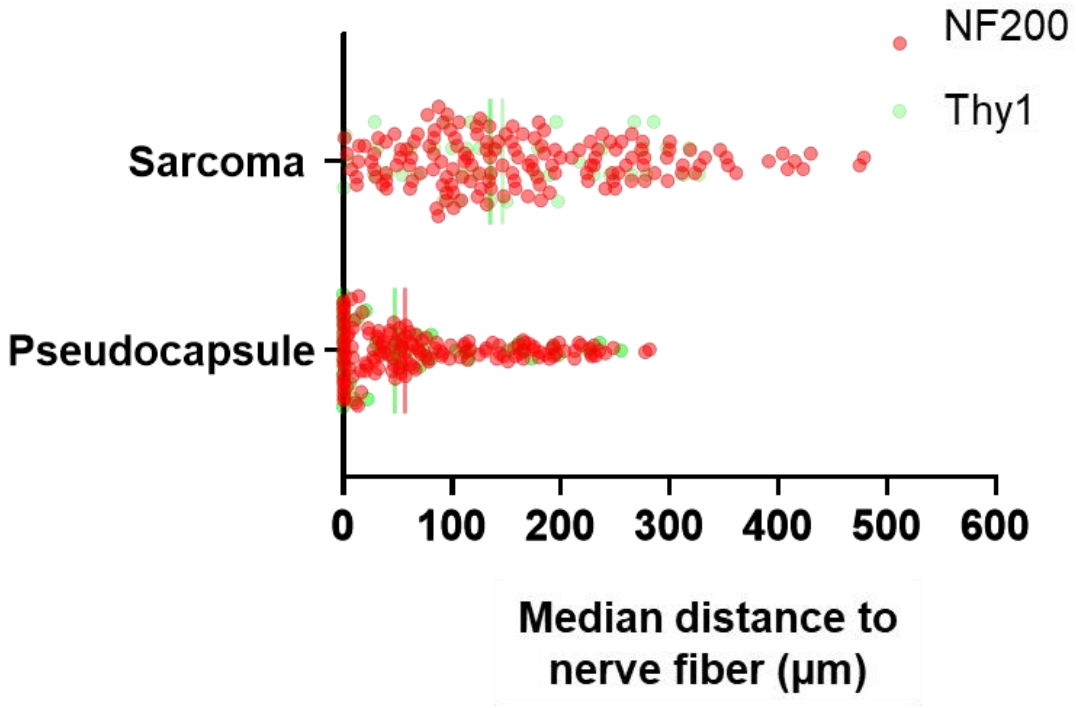
Pathological nerve sprouting in response to osteosarcoma



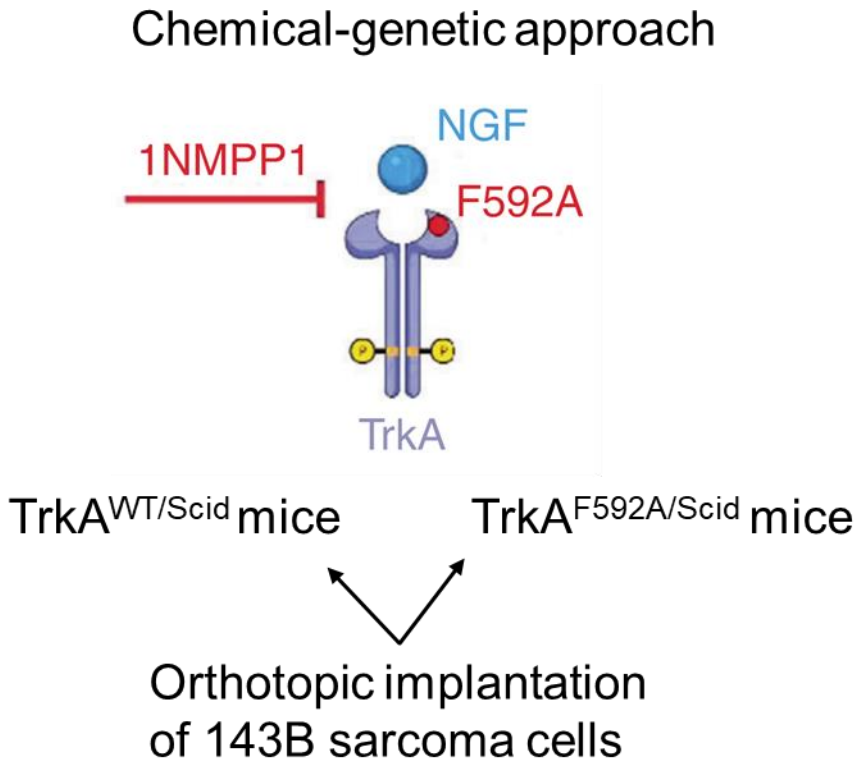
Pathological nerve sprouting in response to OS

Spatial relationship between nerves and tumor



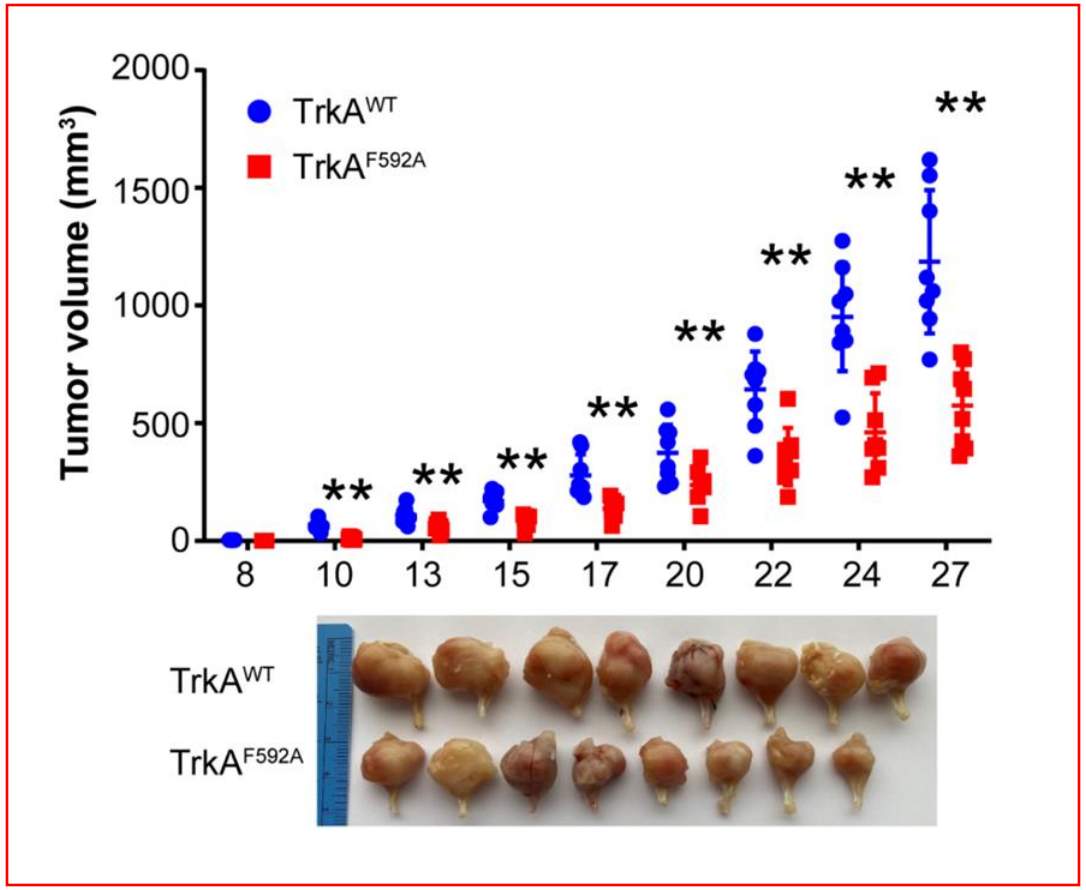


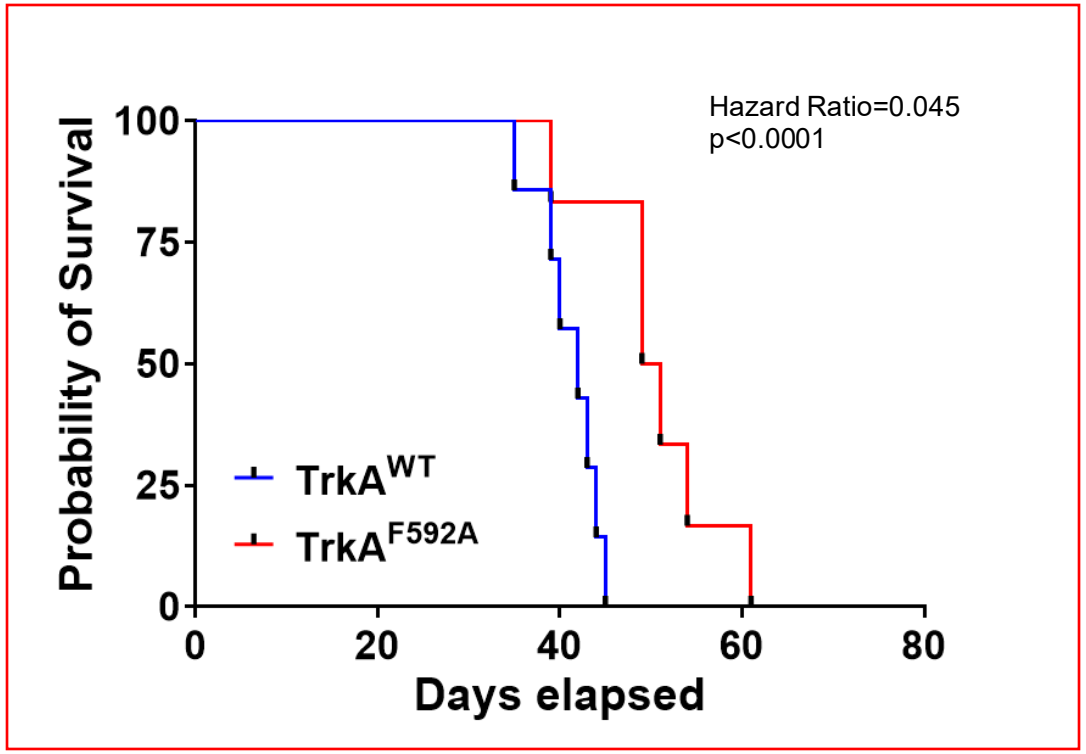
Disruption of TrkA signaling in osteosarcoma



Chen et al. A chemical-genetic approach to studying neurotrophin signaling. Neuron. 2005;46(1):13-21.

Disruption of TrkA signaling abrogates OS growth, prolongs overall survival





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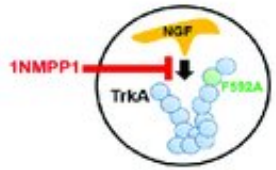
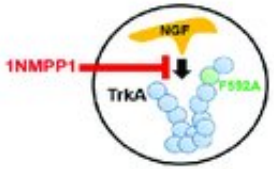
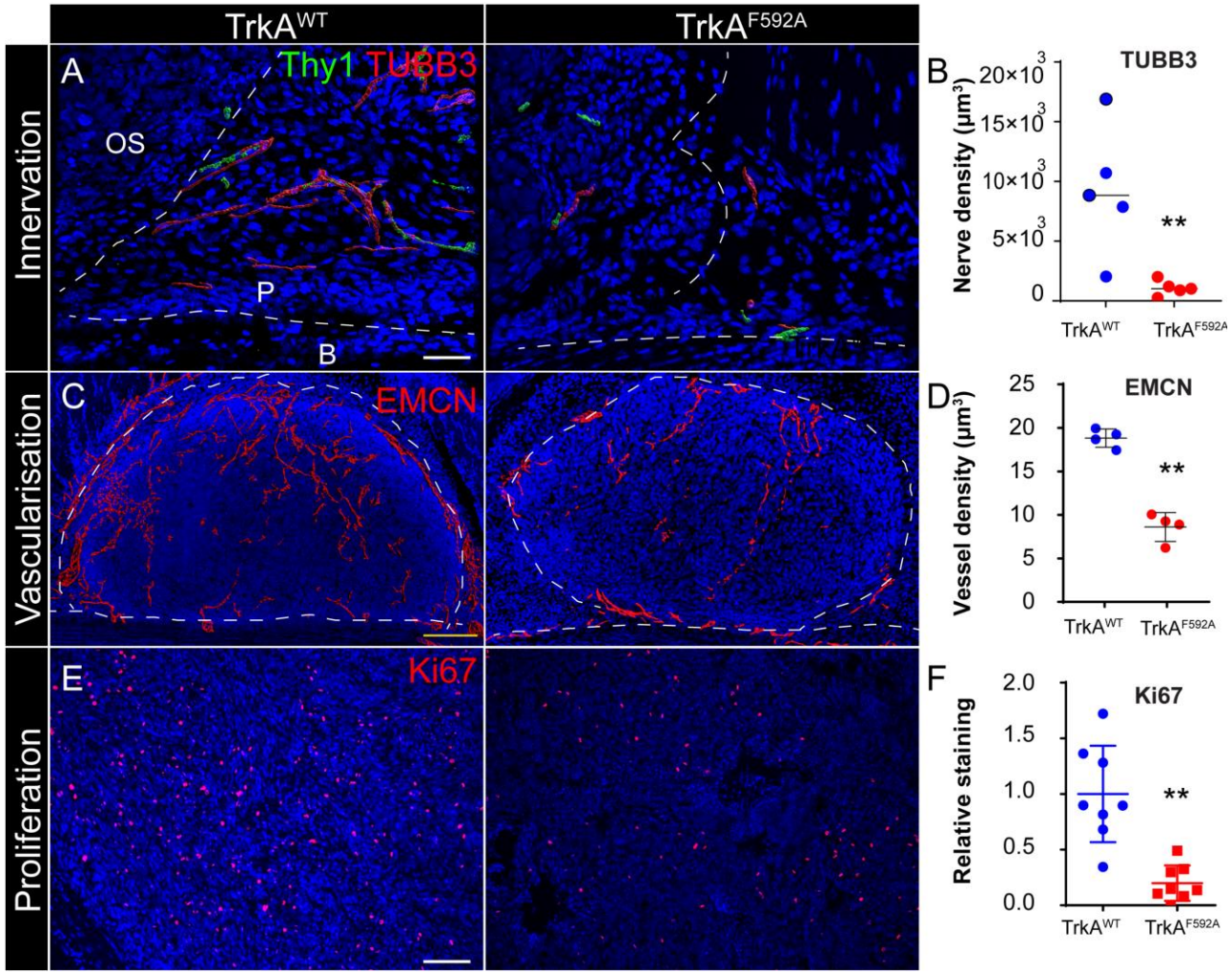


Neuron. 2005 Apr 7;46(1):13-21

Disruption of TrkA signaling attenuates reinnervation, vascularization, proliferation





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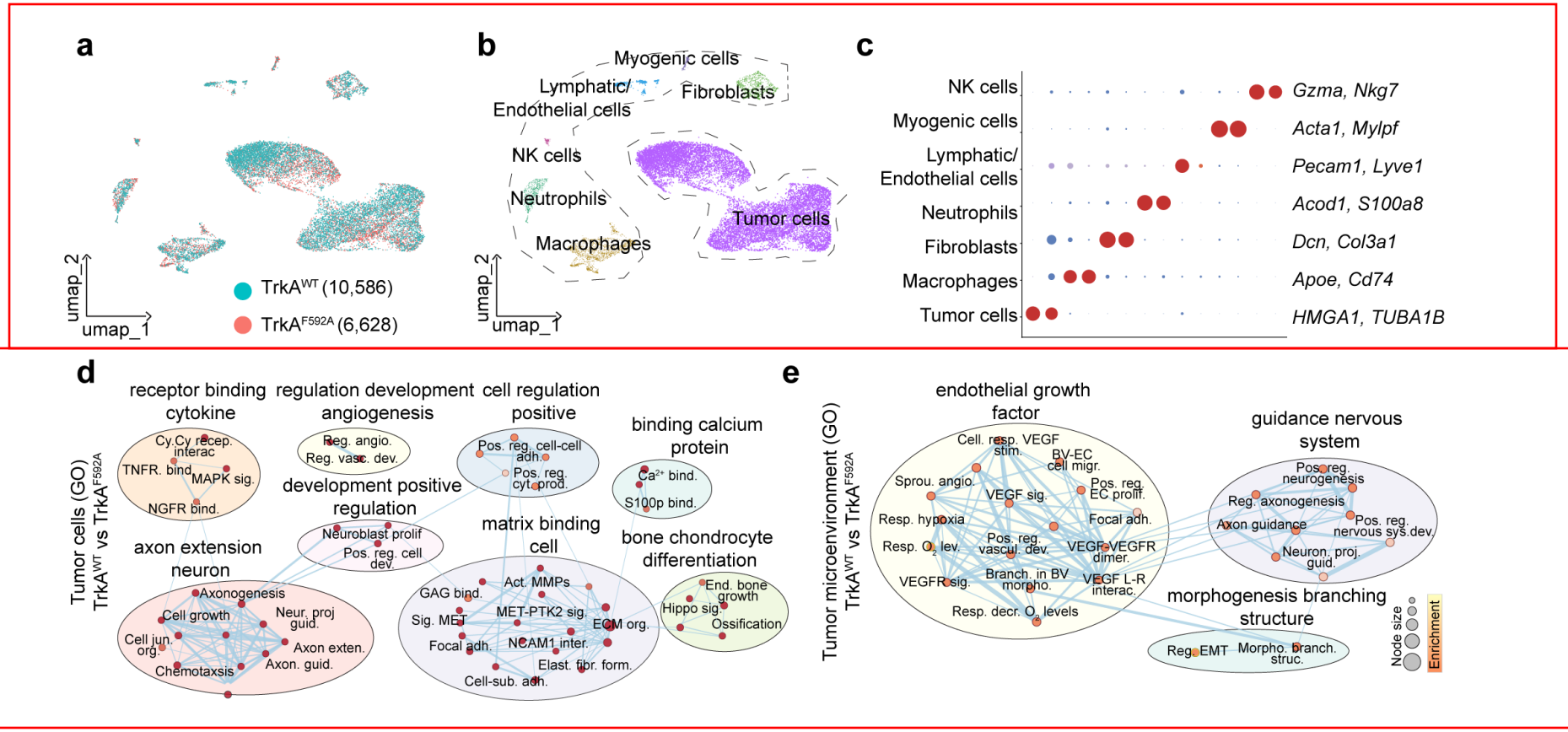
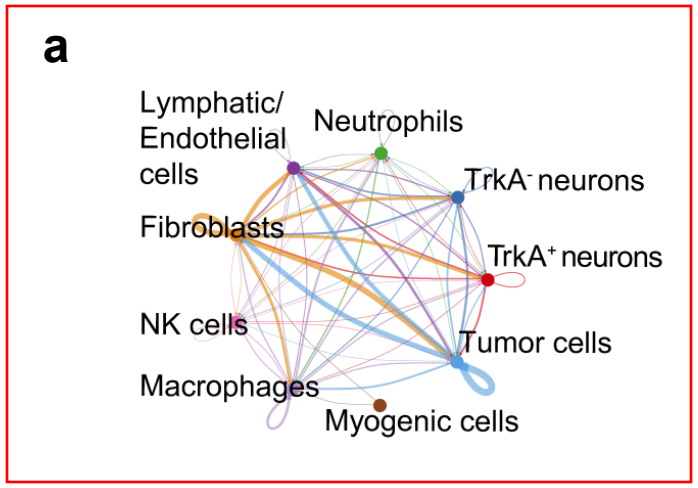
54% ↓

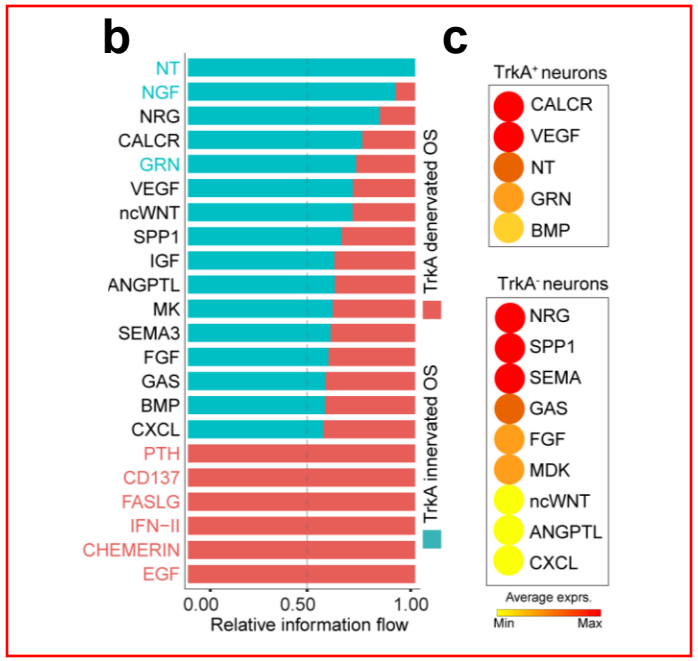
80% ↓

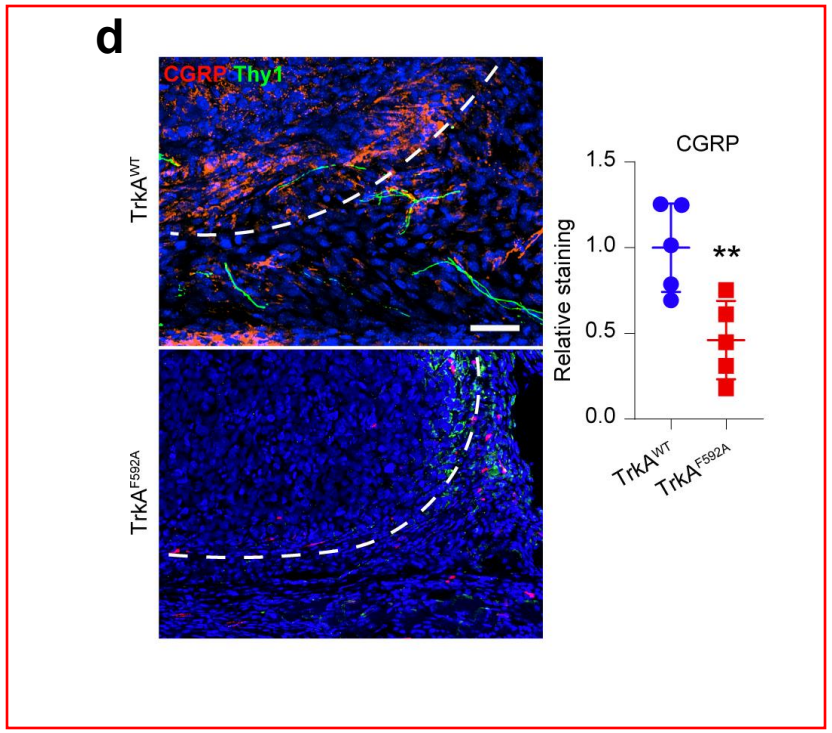
Single-cell RNA-sequencing identifies shifts in human OS cell signaling after denervation



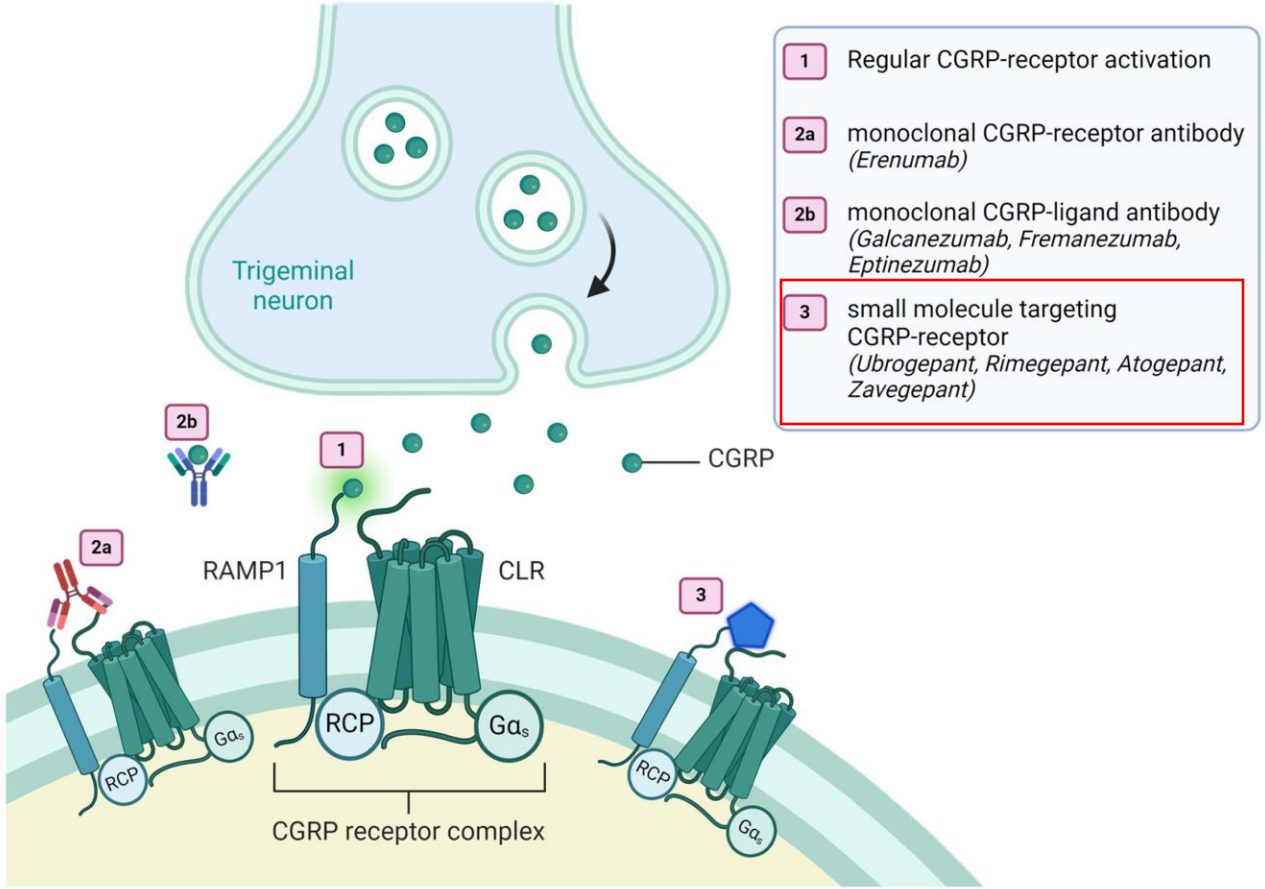
Sequencing identifies CGRP signaling via peripheral neurons as a driver of tumor growth

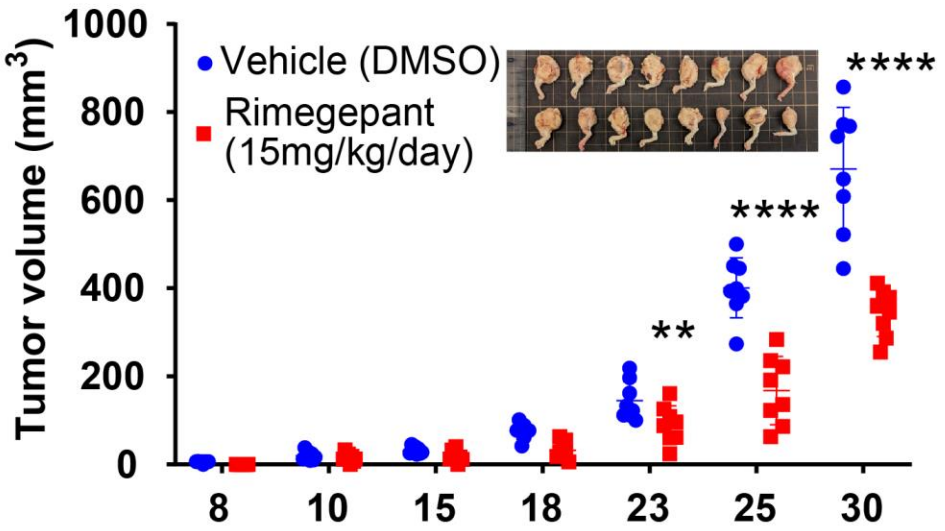






FDA-Approved CGRP Inhibitor Impairs Tumor Growth in OS





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Use of FDA-approved lipid nanoparticles

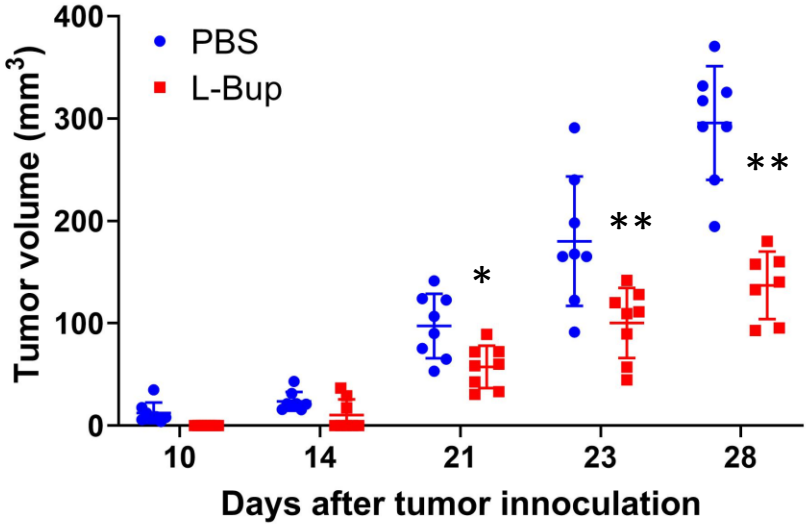
- Exparel™ (L-Bup) is a multivesicular liposomal formulation of bupivacaine currently approved by the FDA for local administration to provide postsurgical analgesia

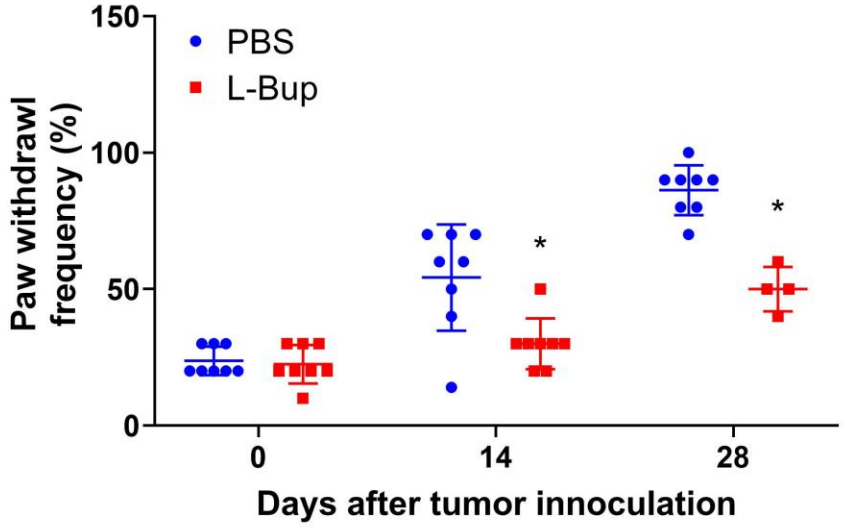
- Exparel™ has been provided to more than 10 M patients since its approval in 2012 for local analgesia

- L-Bup at a particular dose inhibits neurite growth in addition to its analgesic effects

Maya Kaduri et. al. *Sci. Adv.* 2021; 7
Ilfeld BM et. al. *Anesthesiology.* 2021 Feb 1;134(2):283-344.

L-Bup treatment negatively regulates tumor growth and relieves pain





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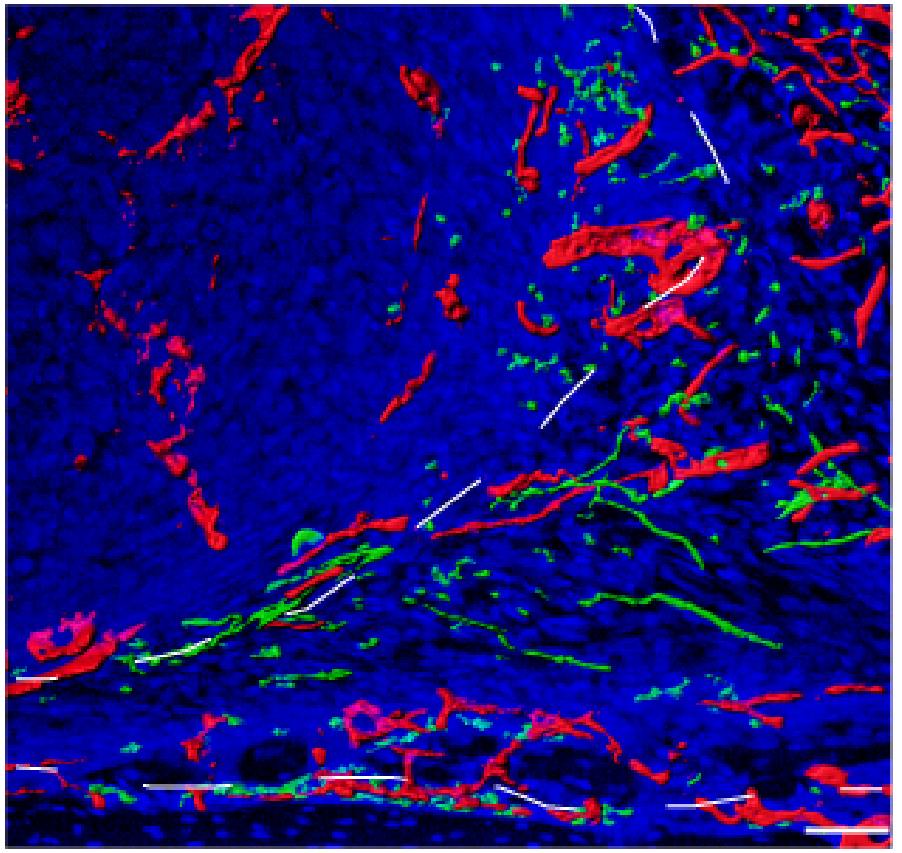
30% ↓

Take-home messages

- Sensory nerve sprouting is a consistent and pathological feature of osteosarcoma.
- Disruption of TrkA signaling reduces tumor growth and impairs neural ingrowth.
- CGRP acts as a downstream neuropeptide mediator, and its inhibition suppresses tumor growth.
- Local nerve silencing with liposomal bupivacaine (LBup) effectively reduces osteosarcoma growth, highlighting a clinically accessible strategy to target tumor-innervating nerves.



bioRxiv [Preprint]. 2024 Jun 25:2024.06.20.599869. doi: 10.1101/2024.06.20.599869.

TrkA $^{+}$ sensory neurons regulate osteosarcoma proliferation and vascularization to promote disease progression

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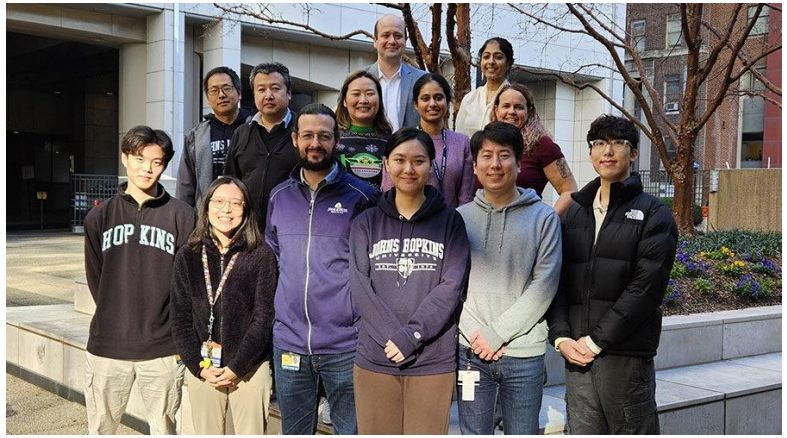
PMID: 38979210 PMCID: PMC11230162 DOI: 10.1101/2024.06.20.599869

Acknowledgments



THE JAMES LABORATORY @ JOHNS HOPKINS

@AaronWJames_Lab
@sowmya_rames

Funding Sources

OREF Orthopaedic Research and Education Foundation

NIH National Institute of Arthritis and Musculoskeletal and Skin Diseases

Department of Defense

NIH National Institute of Dental and Craniofacial Research

MSCRF Maryland Stem Cell Research Fund

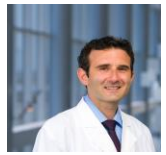
MTF Musculoskeletal Transplant Foundation

American Cancer Society

Alex's Lemonade Stand Foundation for Childhood Cancer

Collaborators

Prof. Thomas L. Clemens
Univ. Maryland, Baltimore

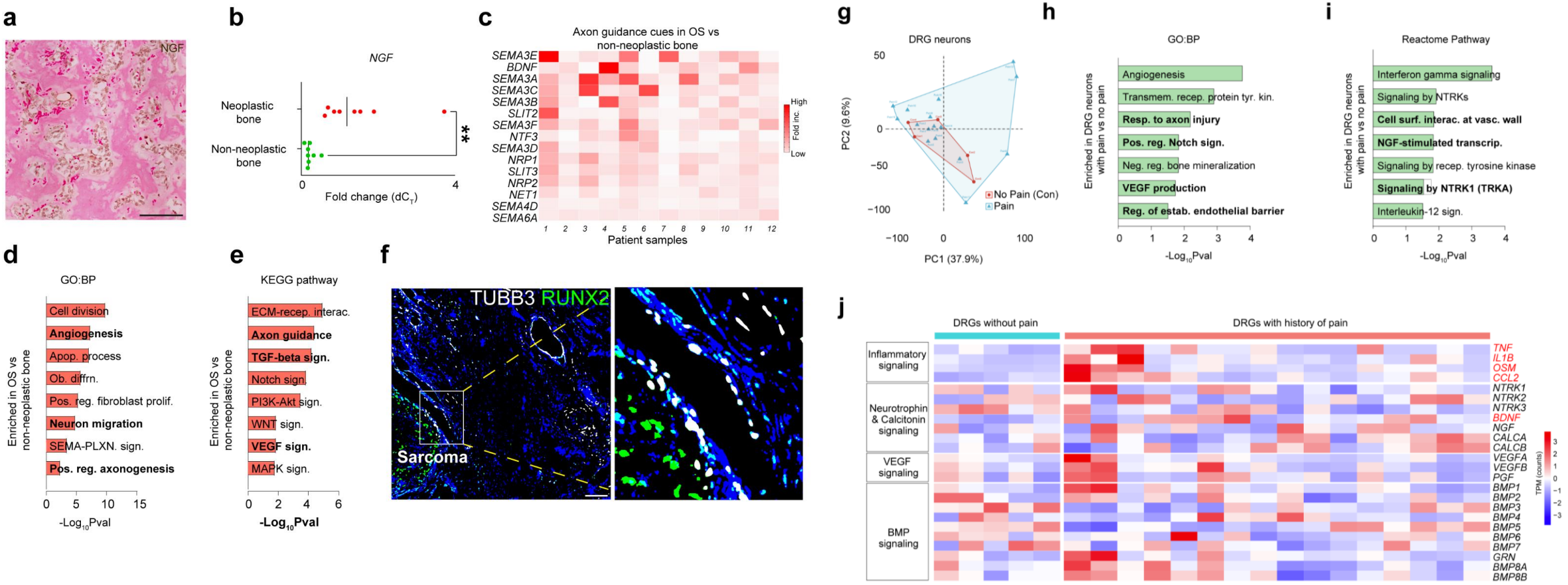


Dr. Benjamin Levi
UT Southwestern, Dallas

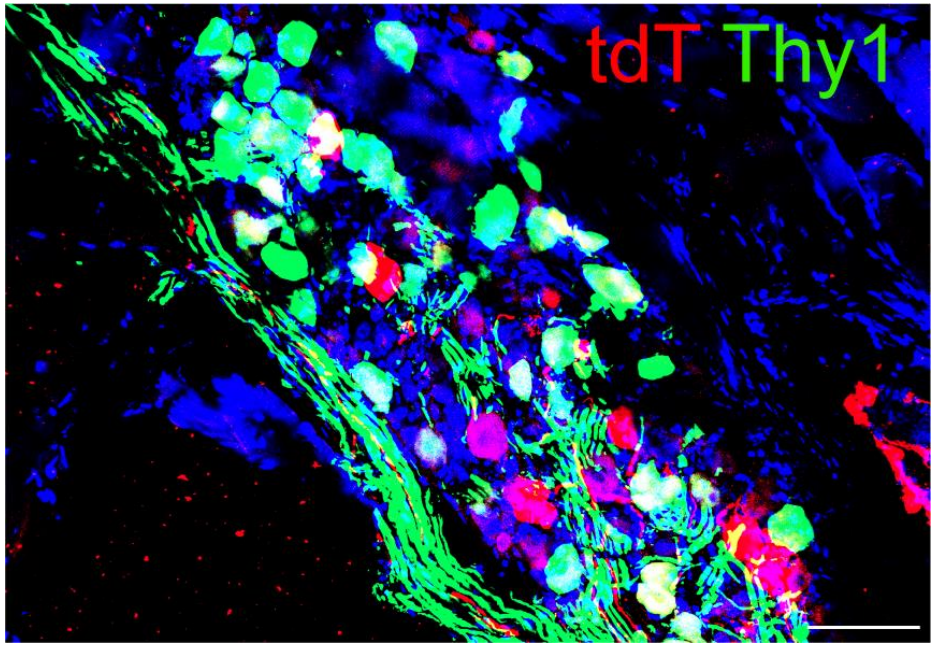
Dr. Yun Guan
Johns Hopkins, Baltimore

Dr. Theodore Price
UT Dallas

NGF-TrkA signaling and innervation within human osteosarcoma biology

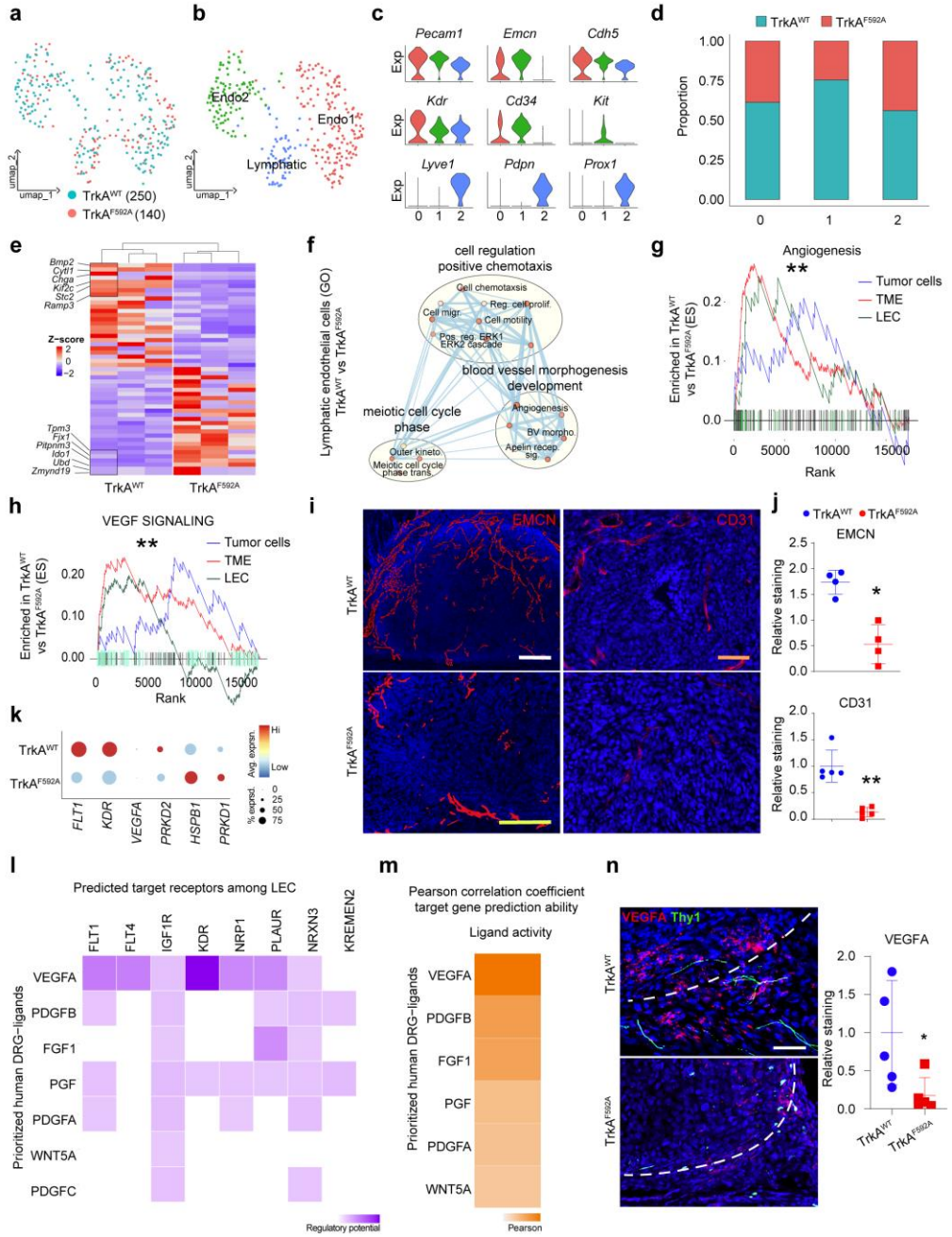


Retrograde neuro-tracer to label tumor-infiltrating peripheral neurons



Lumbar level 5 - DRG

4 weeks after AAV- tdT injection into the tumor implants
Thy1 CD90
DRG Dorsal root ganglion


a
b
c
d
Endo2
Endo1
Lymphatic
umap_1
umap_2
TrkA^WT (250)
TrkA^F592A (140)
Pecam1
Emcn
Cdh5
Kdr
Cd34
Kit
Lyve1
Pdpn
Prox1
Exp
TrkA^WT
TrkA^F592A
Proportion
e
Bmp2
Cyth1
Chga
Kif2c
Stc2
Ramp3
Z-score
Tpm3
Fjx1
Pltpnim3
Ido1
Ubd
Zmynd19
f
cell regulation positive chemotaxis
Cell chemotaxis
Reg. cell prolif.
Cell migr.
Cell motility
Pos. reg. ERK1 ERK2 cascade
blood vessel morphogenesis development
meiotic cell cycle phase
Angiogenesis
BV morpho.
Apelin recep. sig.
Outer kineto.
Meiotic cell cycle phase trans.
Lymphatic endothelial cells (GO) TrkA^WT vs TrkA^F592A
g
Angiogenesis
Tumor cells
TME
LEC
Enriched in TrkA^WT vs TrkA^F592A (ES)
Rank
h
VEGF SIGNALING
i
EMCN
CD31
j
EMCN
CD31
Relative staining
k
FLT1
KDR
VEGFA
PRKD2
HSPB1
PRKD1
Avg. exprsn.
% expsd.
l
Predicted target receptors among LEC
FLT1
FLT4
IGF1R
KDR
NRP1
PLAUR
NRXN3
KREMEN2
Prioritized human DRG−ligands
VEGFA
PDGFB
FGF1
PGF
PDGFA
WNT5A
PDGFC
Regulatory potential
m
Pearson correlation coefficient target gene prediction ability
Ligand activity
Pearson
n
VEGFA Thy1
VEGFA

Direct effect of drug on human OS cells