Microglia-neuron interaction in chronic pain

Long-Jun Wu, PhD

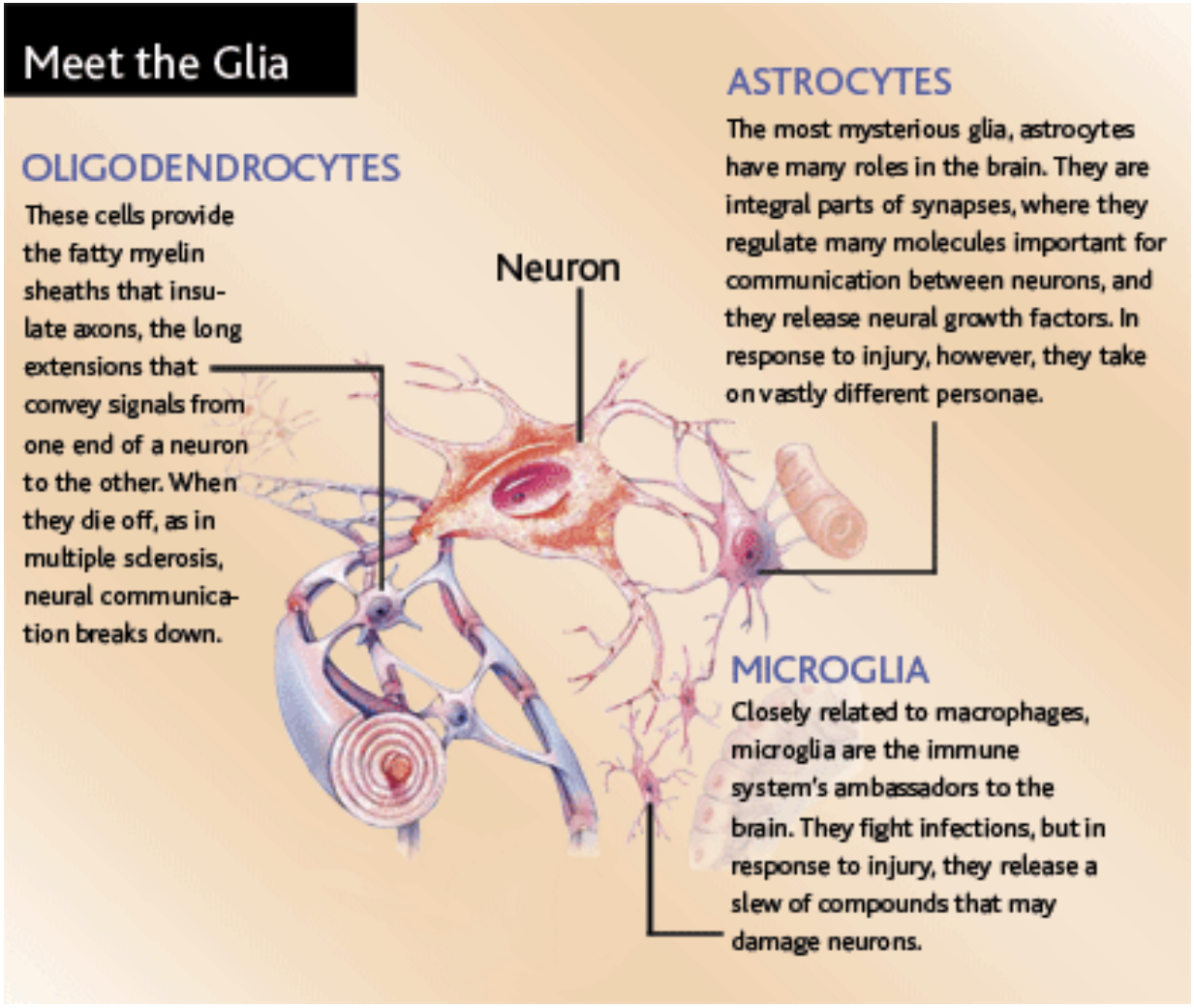
Professor and Consultant

Department of Neurology

Department of Neuroscience

Mayo Clinic

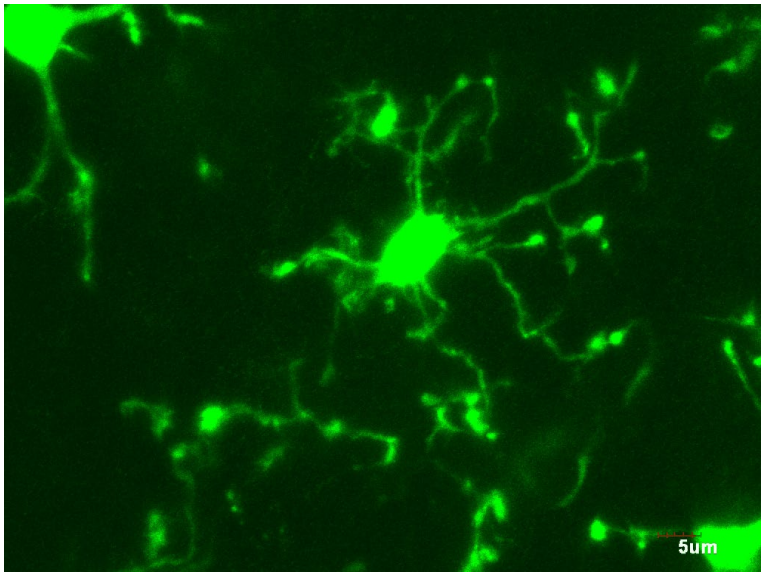
Microglia: principal immune cells in the brain



Miller, *Science,* 2005

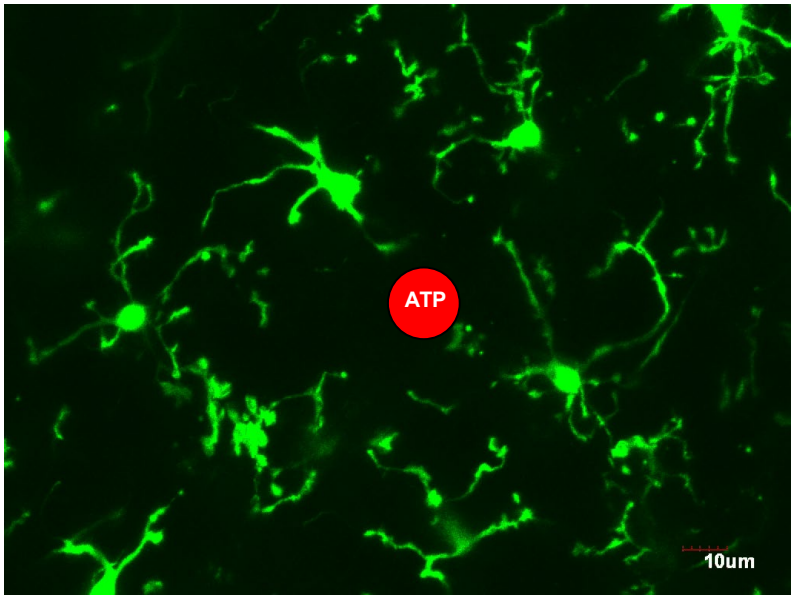
Microglia are highly dynamic

Microglia basal motility, 10 min



Microglia: CX3CR1-GFP

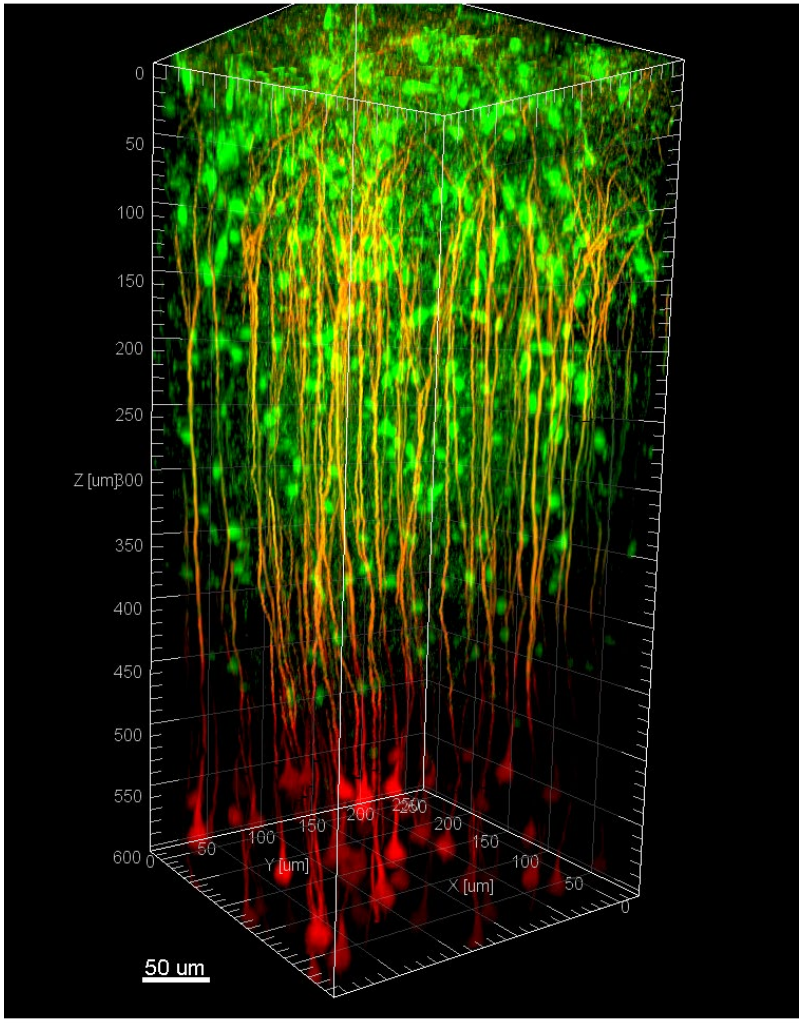
Microglia process chemotaxis, 30 min

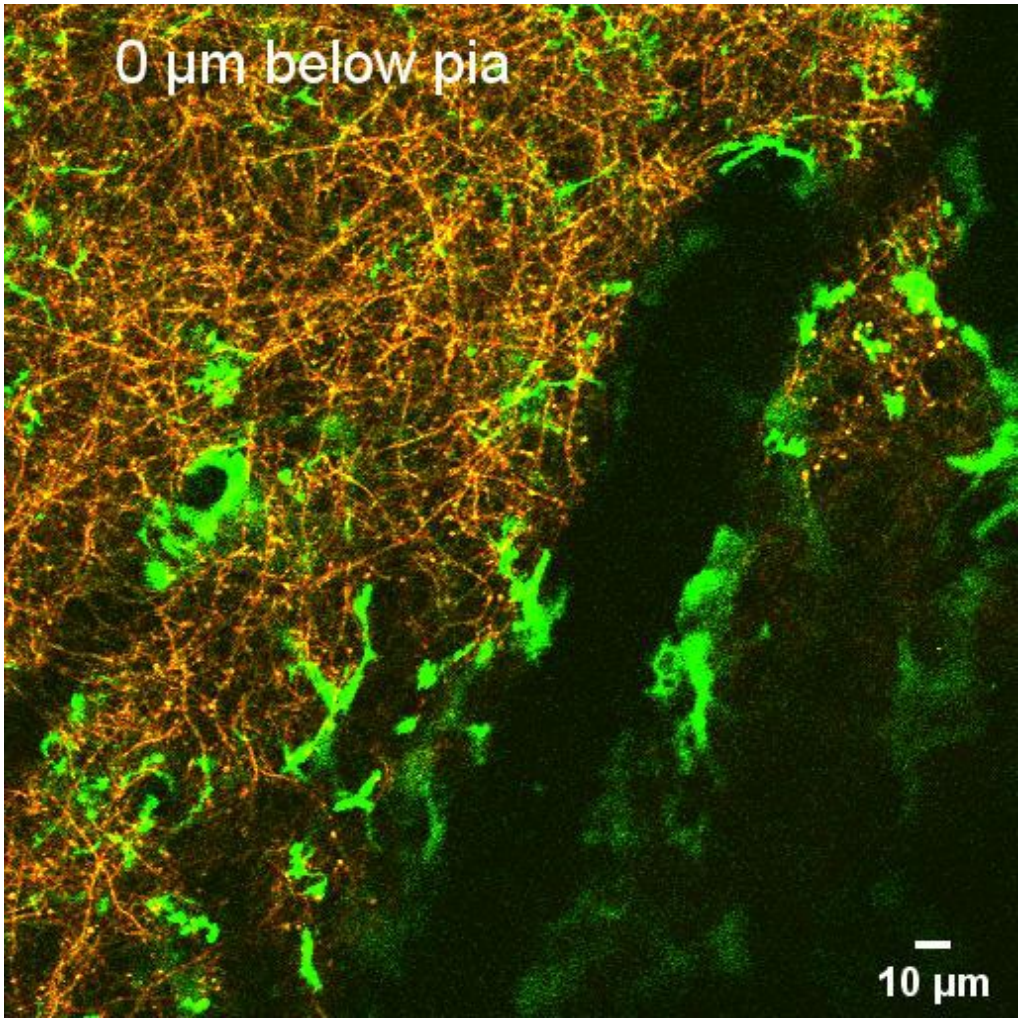


Wu et al., *Glia*, 2007

Two-photon imaging microglia-neuron interaction in vivo

Neuron: Thy1-YFP **Microglia: CX3CR1-GFP**

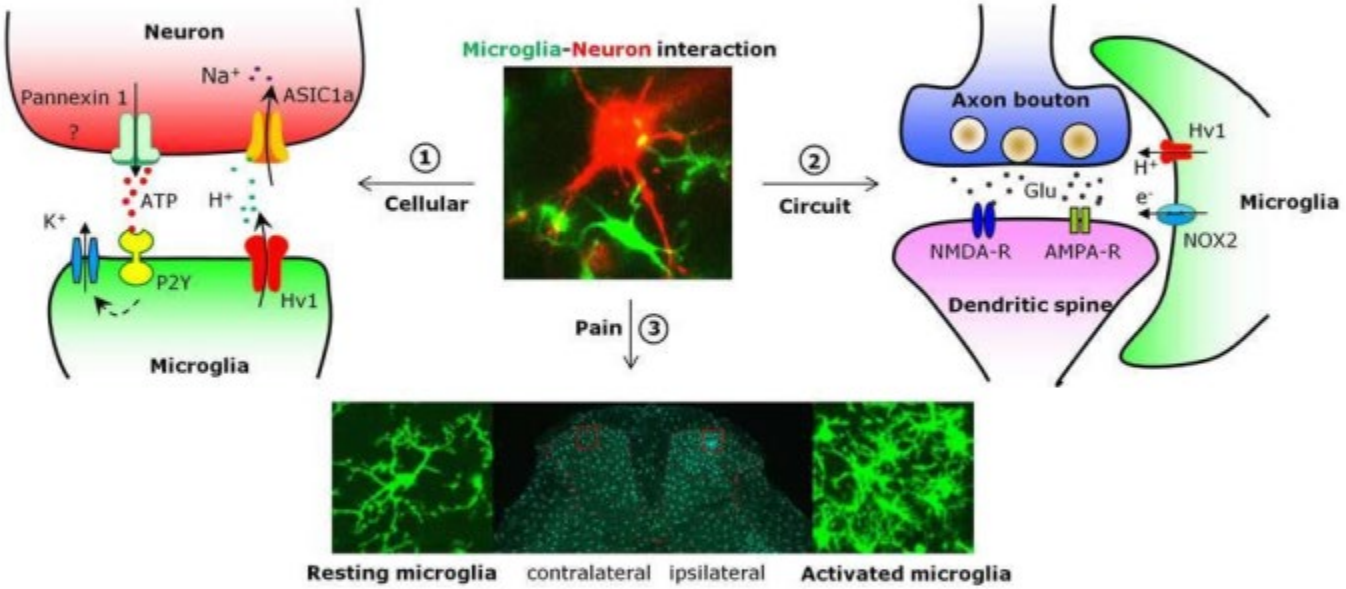




MAYO CLINIC

Neuroimmune Interaction in Health and Disease

HOME RESEARCH MEMBERS PUBLICATIONS LAB NEWS LAB PHOTOS



Major Research Topics

(1) The molecular signaling of microglia-neuron communication

(2) Microglia in synaptic function and neuronal circuits

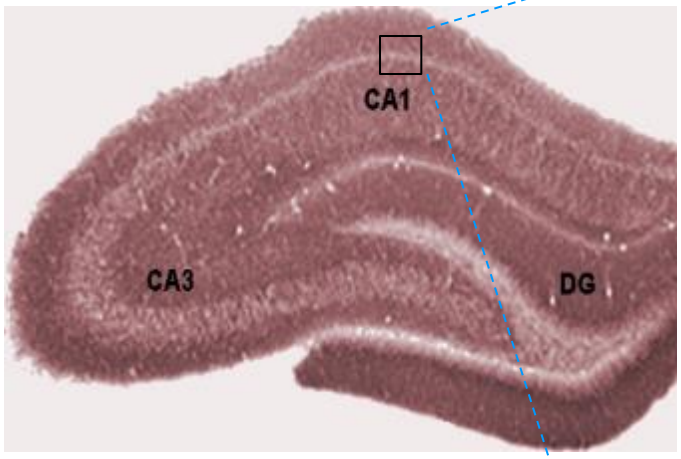
(3) Microglia in pain, epilepsy, stroke, and autoimmune diseases

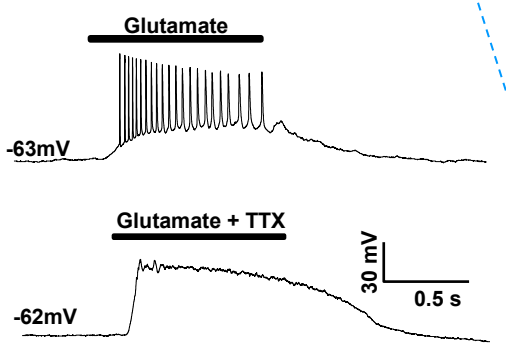
http://neuroimmunelab.mayo.edu/

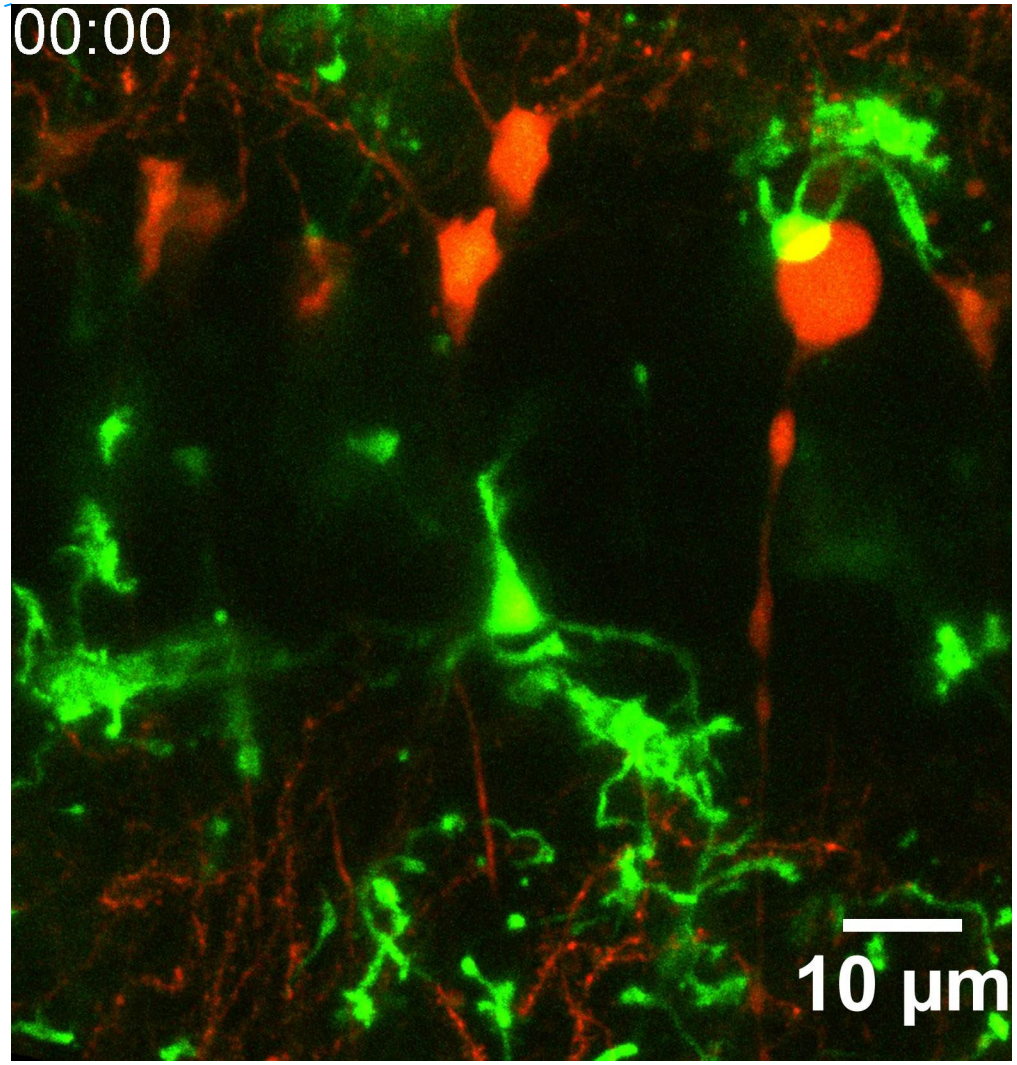
Overview

- How microglia sense neuronal activity?
- What is the function of microglia in chronic pain?

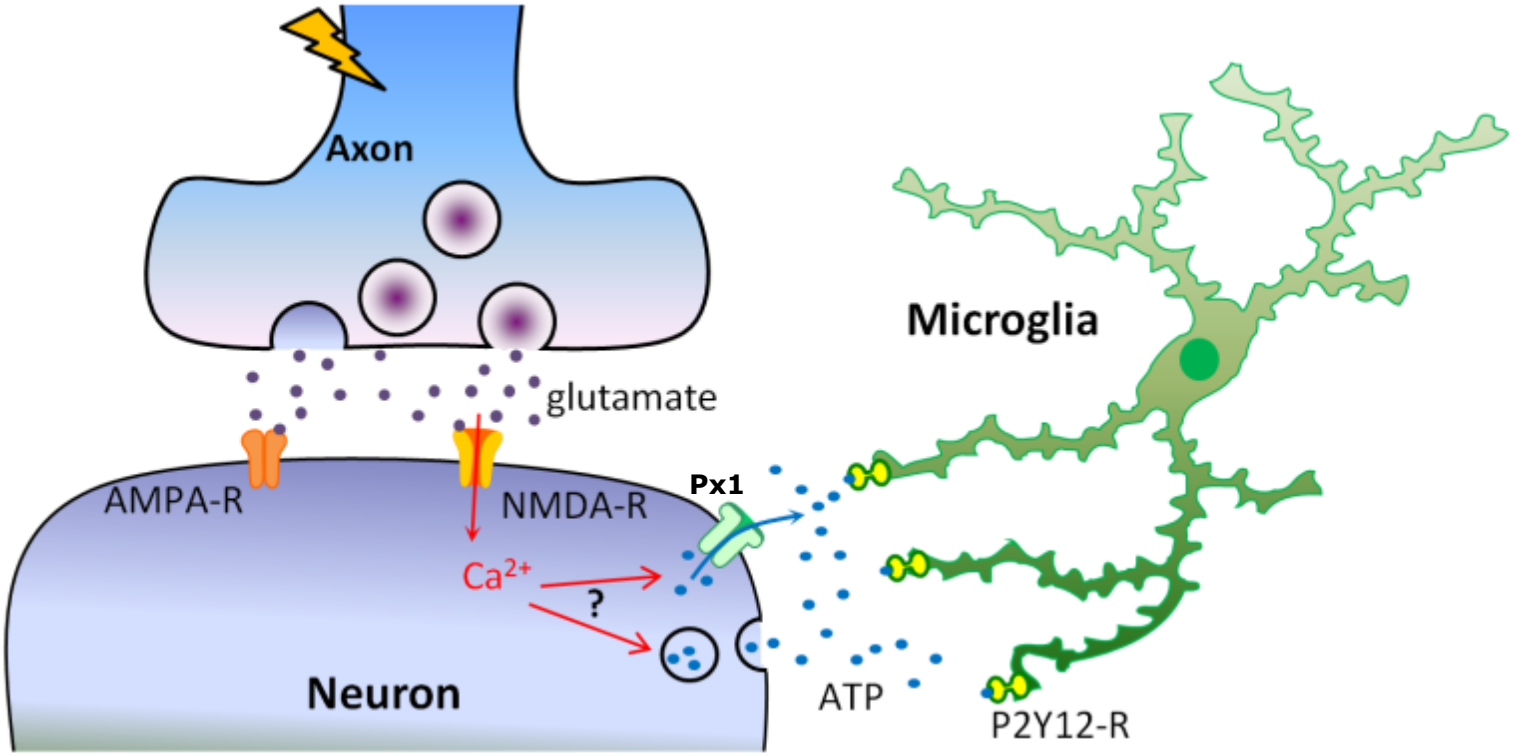
Microglial process extension to hyperactive neurons







Microglial process extension to hyperactive neurons

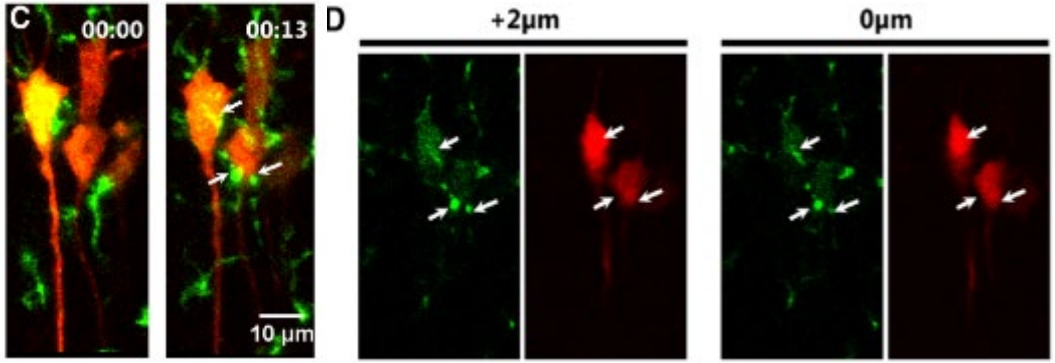


Eyo et al., *J Neurosci*, 2014

The Journal of Neuroscience, August 6, 2014 • 34(32):10528–10540

Neuronal Hyperactivity Recruits Microglial Processes via Neuronal NMDA Receptors and Microglial P2Y12 Receptors after Status Epilepticus

Ukpong B. Eyo, Jiyun Peng, Przemyslaw Swiatkowski, Aparna Mukherjee, Ashley Bispo, and Long-Jun Wu
Department of Cell Biology and Neuroscience, Rutgers University, Piscataway, New Jersey 08854



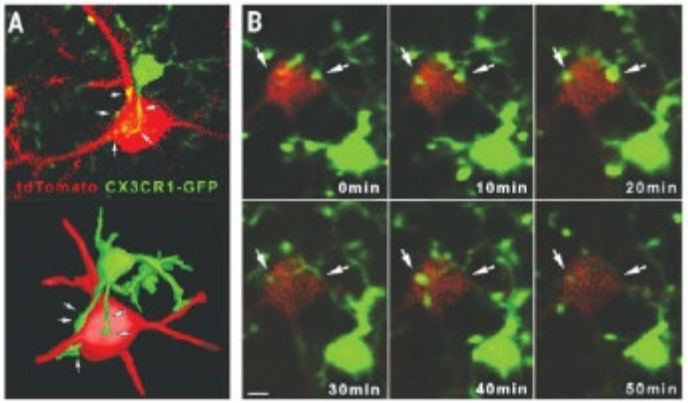
Science **367**, 528–537 (2020) 31 January 2020

RESEARCH ARTICLE

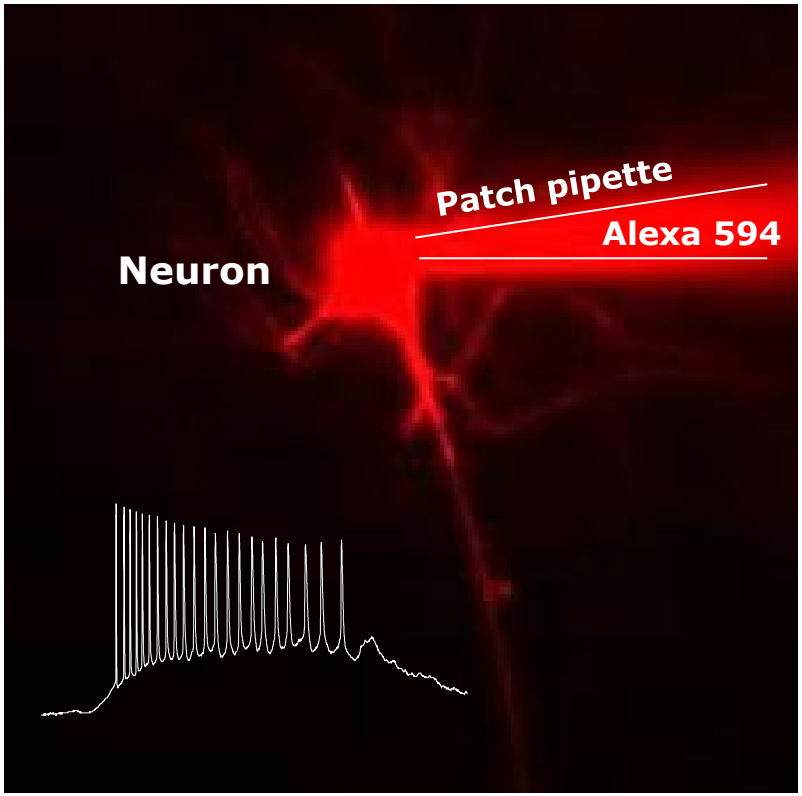
CELLULAR NEUROSCIENCE

Microglia monitor and protect neuronal function through specialized somatic purinergic junctions

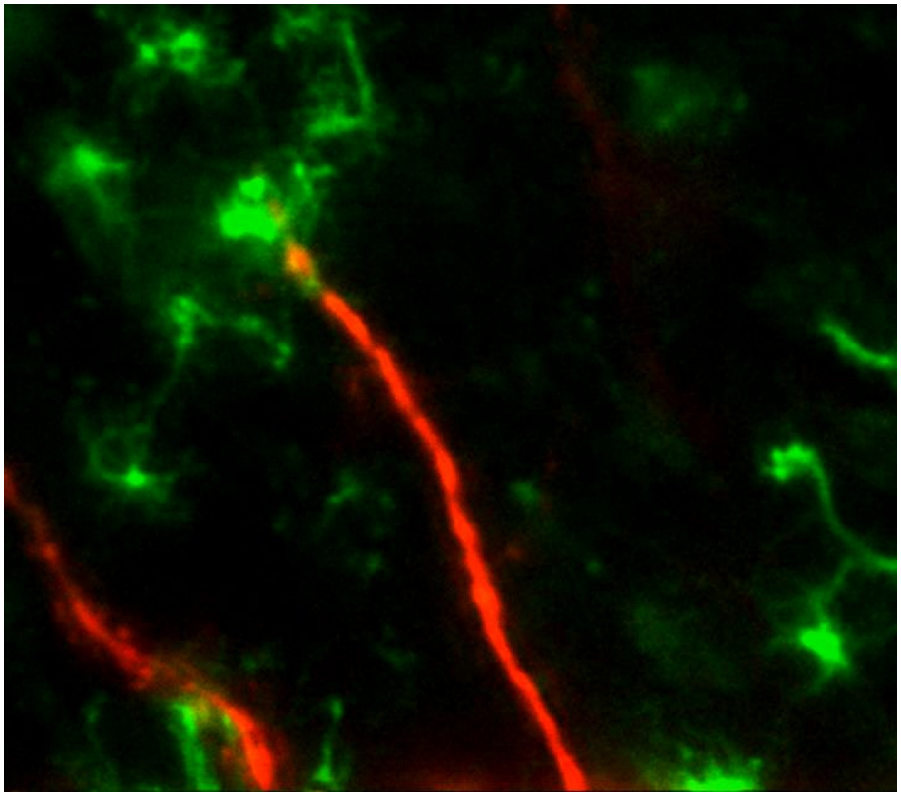
Csaba Cserép[1]*, Balázs Pósfai[1,2]*, Nikolett Lénárt[1], Rebeka Fekete[1,2], Zsófia I. László[2,3], Zsolt Lele[3], Barbara Orsolits[1], Gábor Molnár[4], Steffanie Heindl[5], Anett D. Schwarcz[1], Katinka Ujvári[1], Zsuzsanna Környei[1], Krisztina Tóth[1,2], Eszter Szabadits[1], Beáta Sperlágh[6], Mária Baranyi[6], László Csiba[7], Tibor Hortobágyi[8,9,10], Zsófia Maglóczky[11], Bernadett Martinecz[1], Gábor Szabó[12], Ferenc Erdélyi[12], Róbert Szipőcs[13], Michael M. Tamkun[14], Benno Gesierich[5], Marco Duering[5,15], István Katona[3], Arthur Liesz[5,15], Gábor Tamás[4], Ádám Dénes[1]†



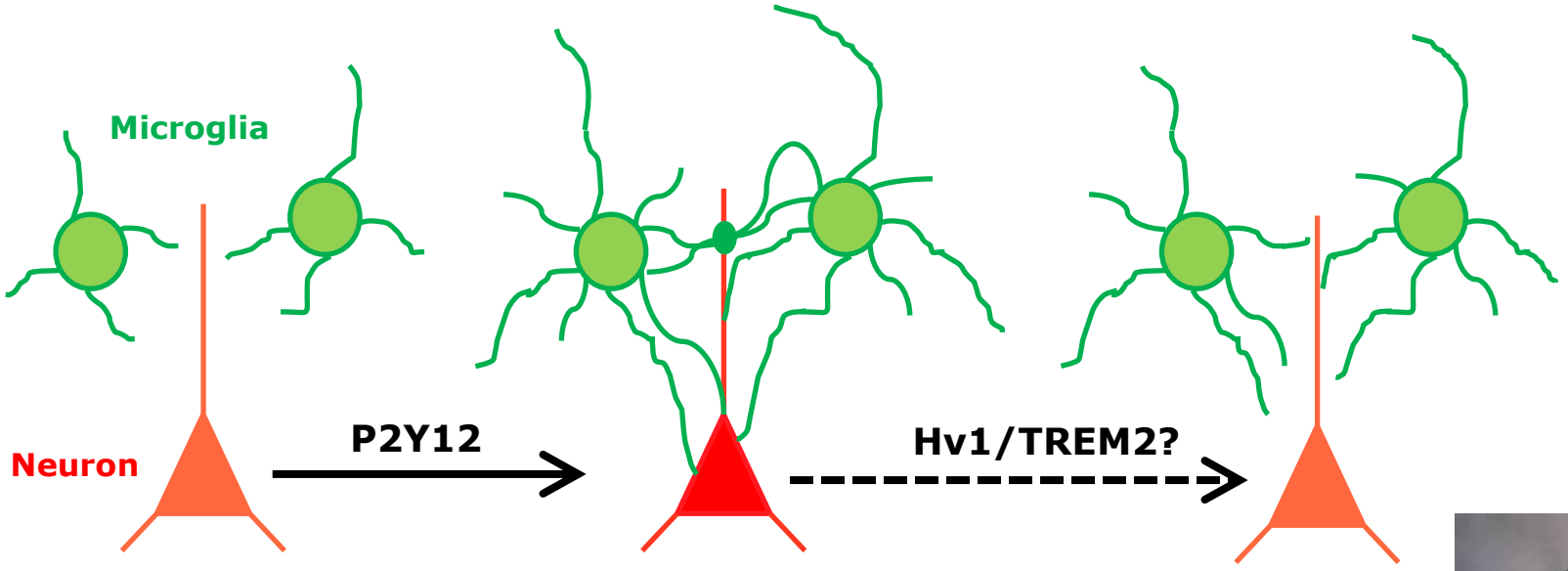
Neuronal firing triggers microglial process convergence



Microglia: GFP Neuron: Alexa 594



Microglia sense ATP from hyperactive neurons



Neuronal activities induce microglial process extension/convergence/phagocytosis

Microglia dampen neuronal activities

Dr. Ukpong Eyo

Now at U Virginia

Eyo et al., *J Neurosci*, 2014

Eyo et al., *J Neurosci*, 2015

Tian et al., *J Neurosci*, 2017

Eyo et al., *Cell Rep*, 2018

Mo et al., *J Neurosci*, 2019

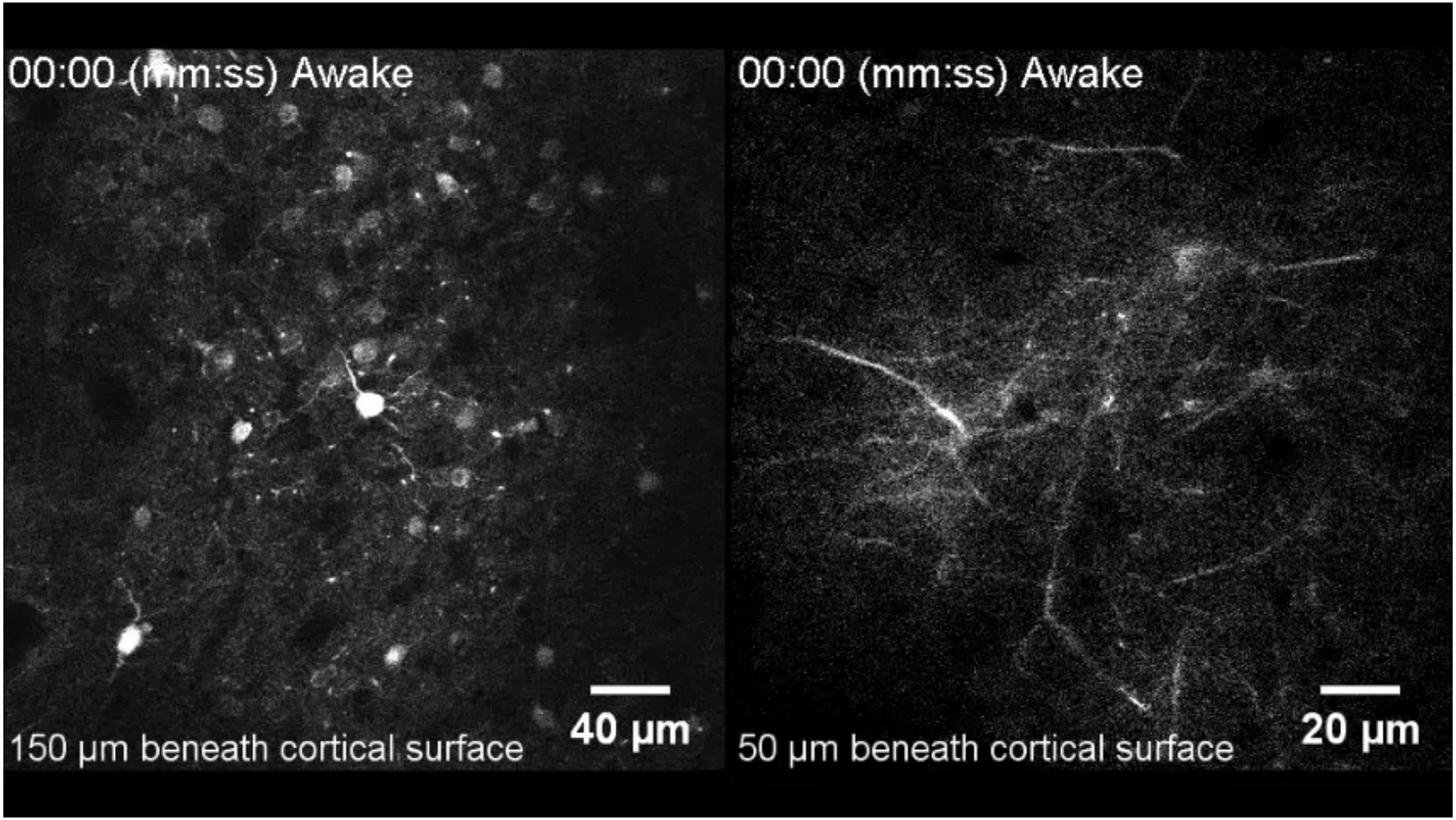
Eyo and Wu, *Prog Neurobiol*, 2019

A critical question in the microglia field:

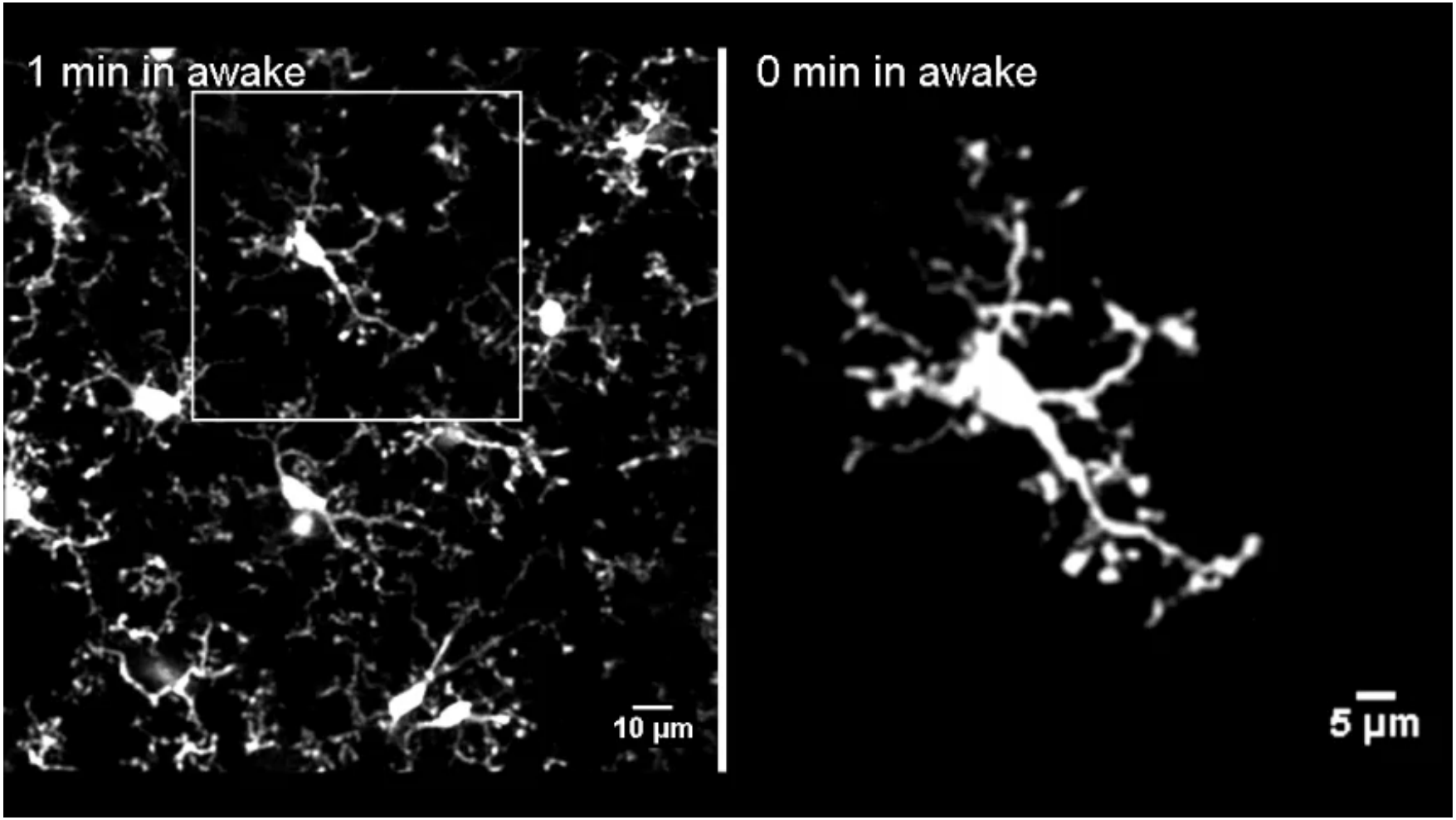
Most microglia in vivo imaging was done under anesthesia.

How about microglia dynamics in awake mice?

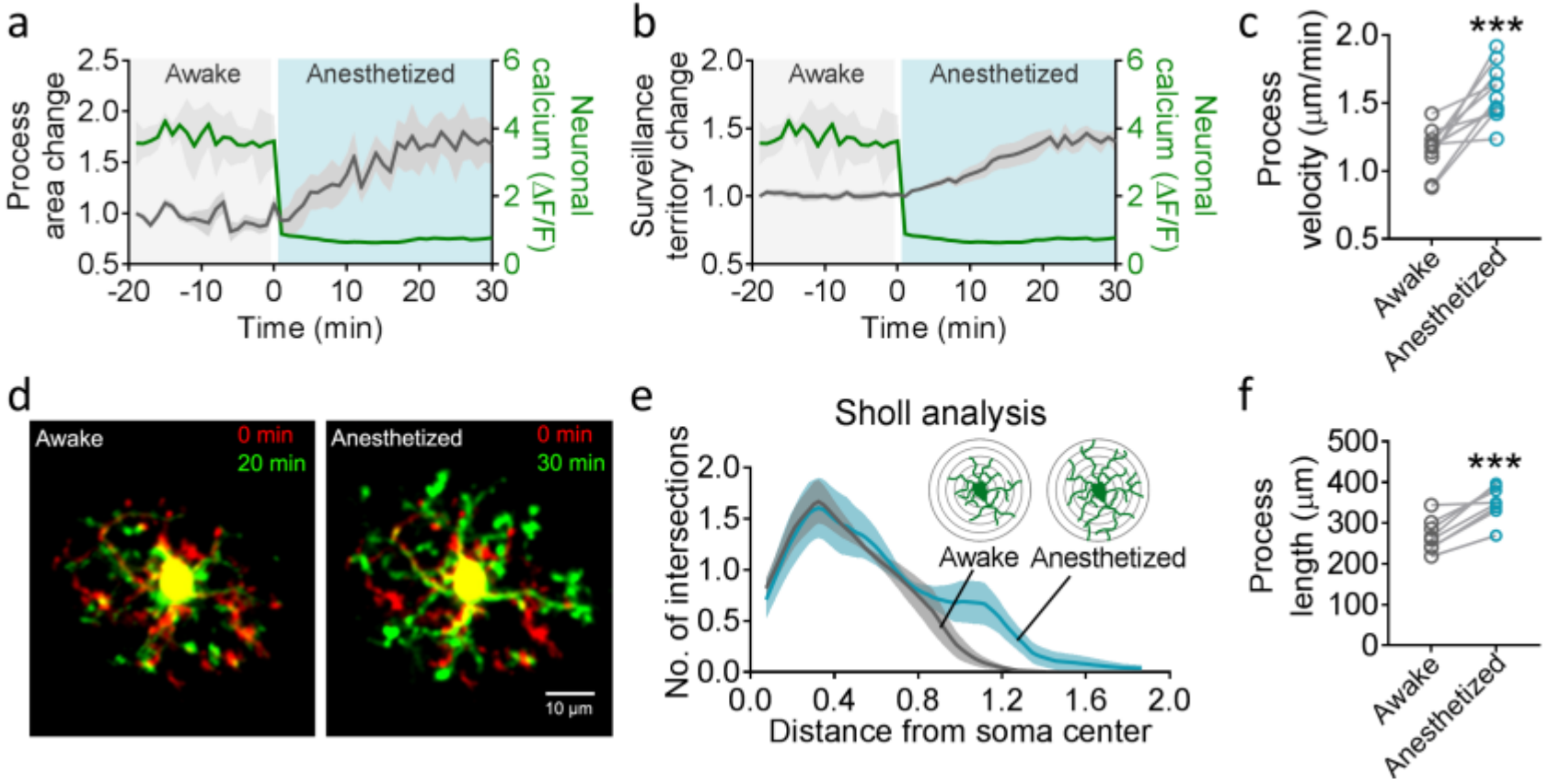
Neuronal activity in awake and anesthetized mice



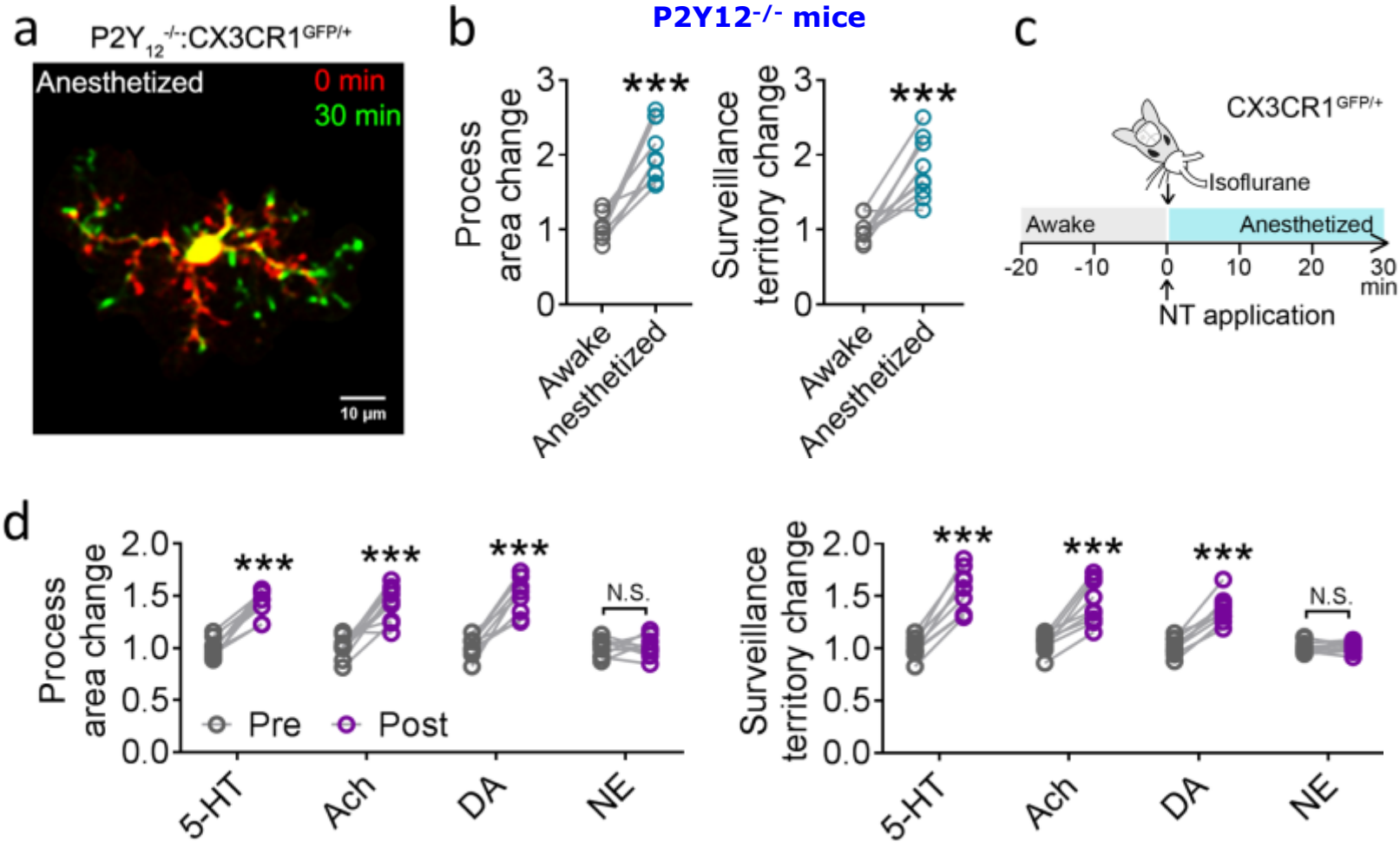
Microglial process dynamics in awake and anesthetized mice



Increased microglial process dynamics after anesthesia

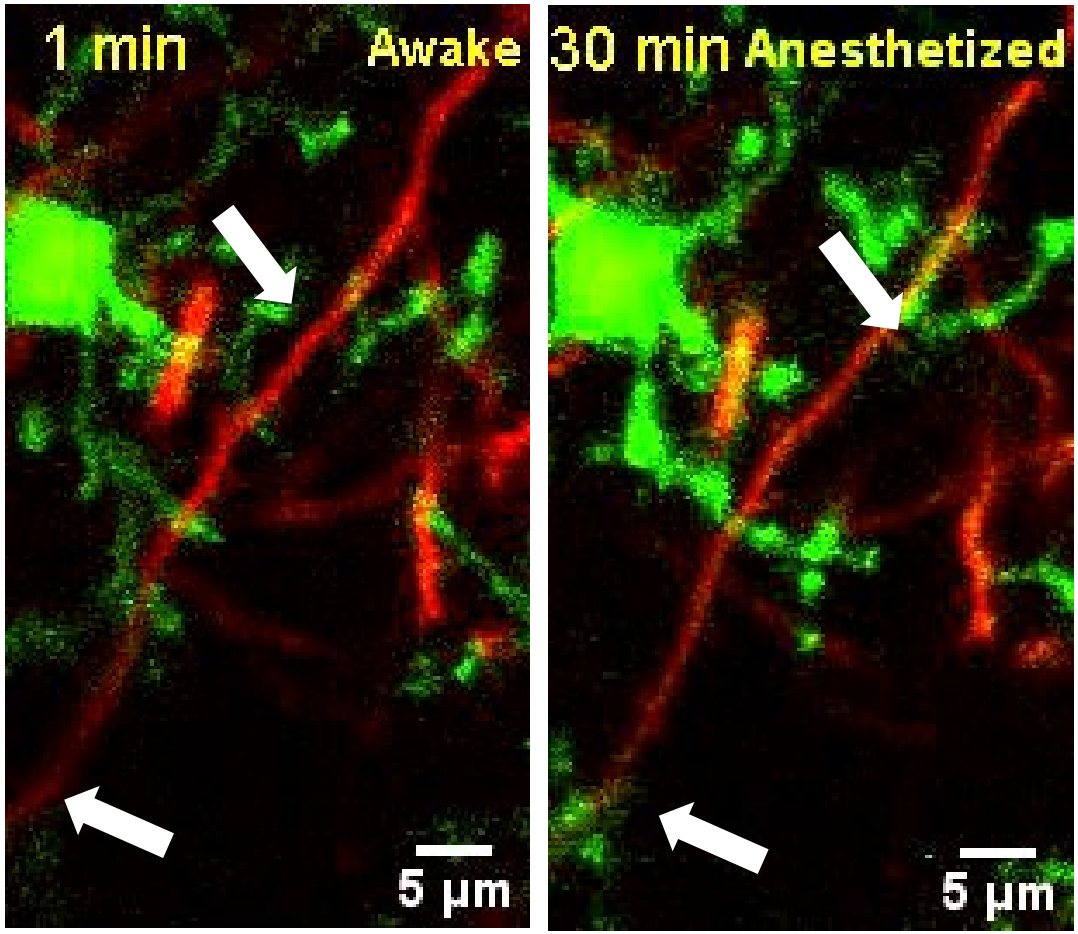


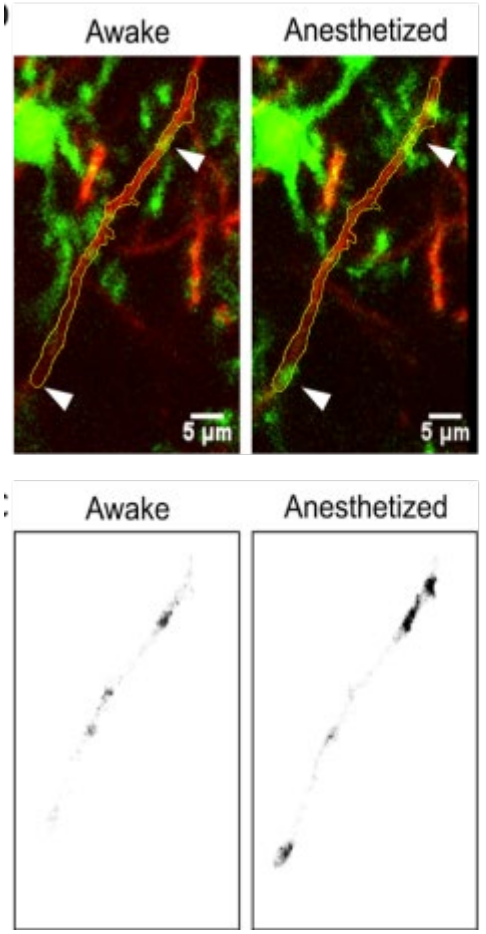
What is the molecular mechanisms?

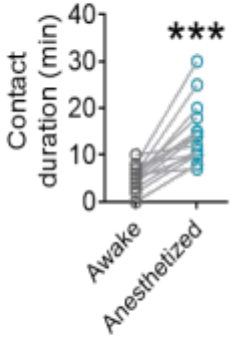


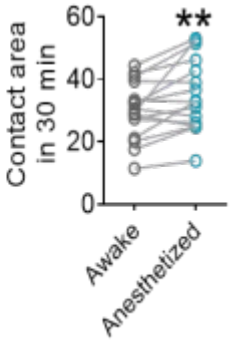
Norepinephrine controls microglial process dynamics

The functional significance?





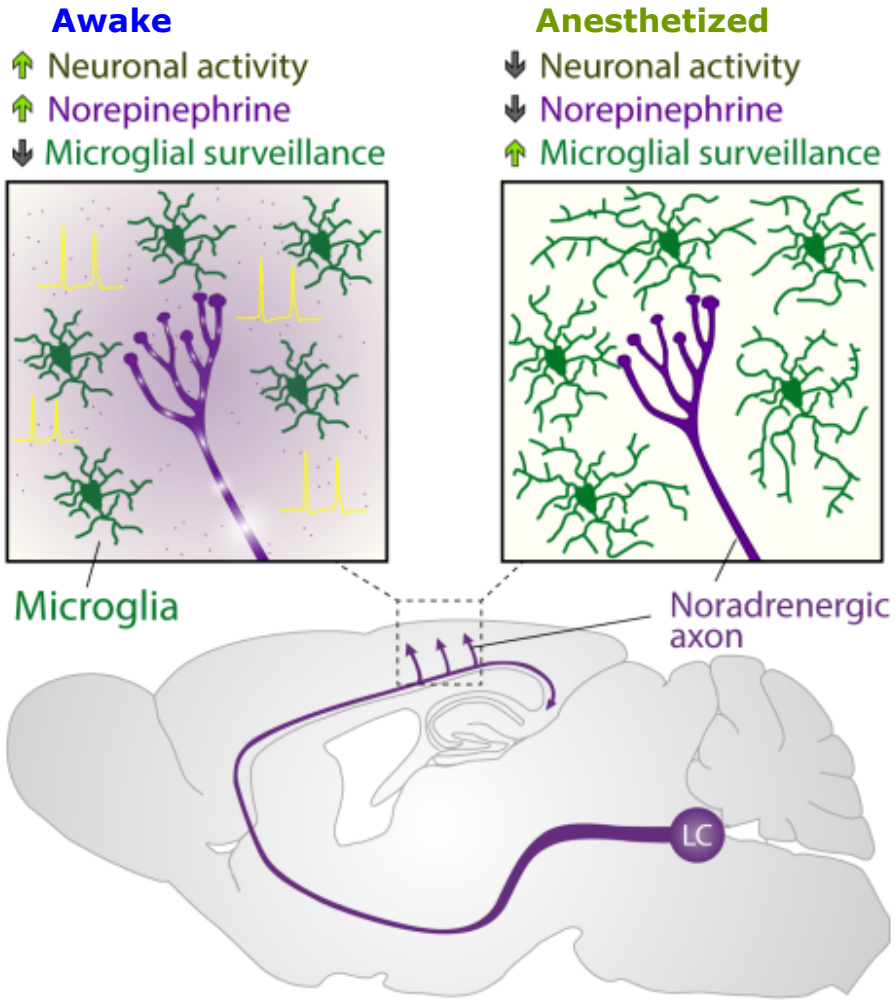




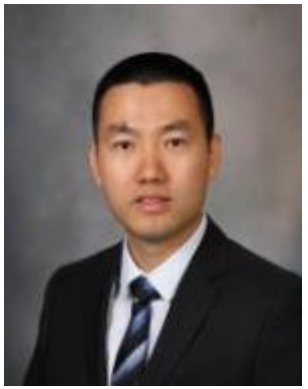
CX3CR1-GFP:Thy1-YFP

Increased contact with neuronal dendrites after anesthesia

Neuronal activity inhibits microglial process dynamics via norepinephrine signaling in awake mice



Liu et al., *Nat Neurosci*, 2019



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nature neuroscience

ARTICLES

https://doi.org/10.1038/s41593-019-0511-3

Neuronal network activity controls microglial process surveillance in awake mice via norepinephrine signaling

Yong U. Liu[1], Yanlu Ying[1], Yujiao Li[1], Ukpong B. Eyo[1,2], Tingjun Chen[1], Jiaying Zheng[1], Anthony D. Umpierre[1], Jia Zhu[1], Dale B. Bosco[1], Hailong Dong[3] and Long-Jun Wu[1,4,5*]

NATURE NEUROSCIENCE | VOL 22 | NOVEMBER 2019 | 1771–1781 | www.nature.com/natureneuroscience

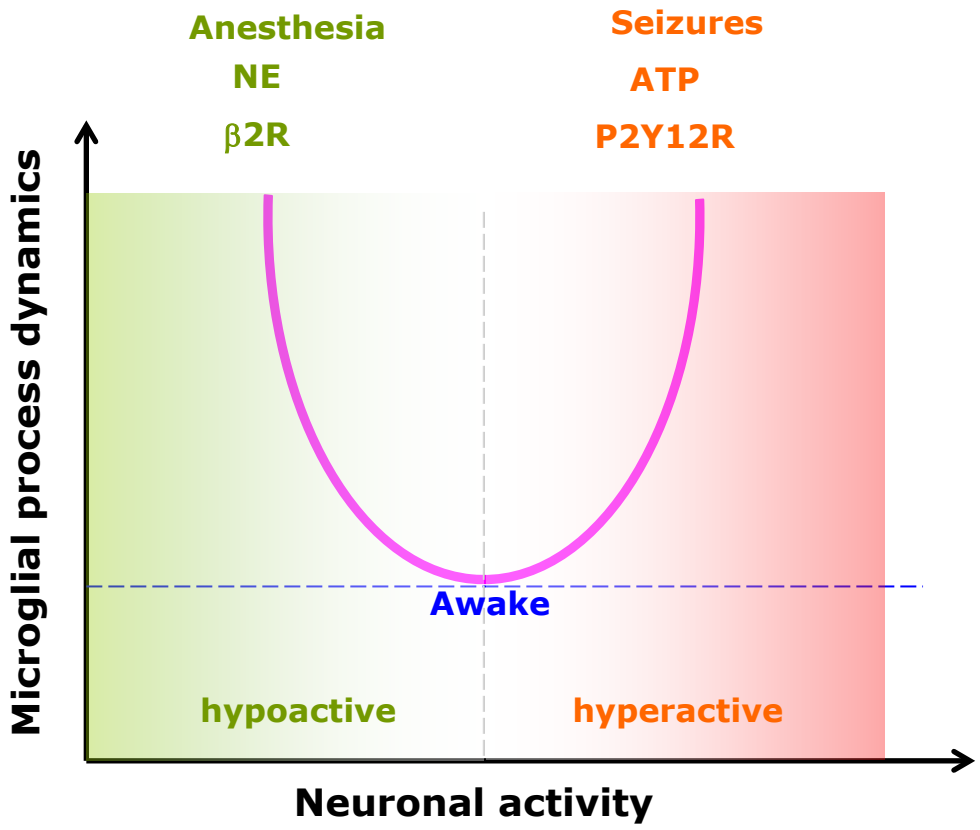
Noradrenergic signaling in the wakeful state inhibits microglial surveillance and synaptic plasticity in the mouse visual cortex

Rianne D. Stowell[1,2,7], Grayson O. Sipe[3,7], Ryan P. Dawes[1,2], Hanna N. Batchelor[1], Katheryn A. Lordy[1], Brendan S. Whitelaw[1,2], Mark B. Stoessel[1,2], Jean M. Bidlack[4], Edward Brown[5], Mriganka Sur[3] and Ania K. Majewska[1,6*]

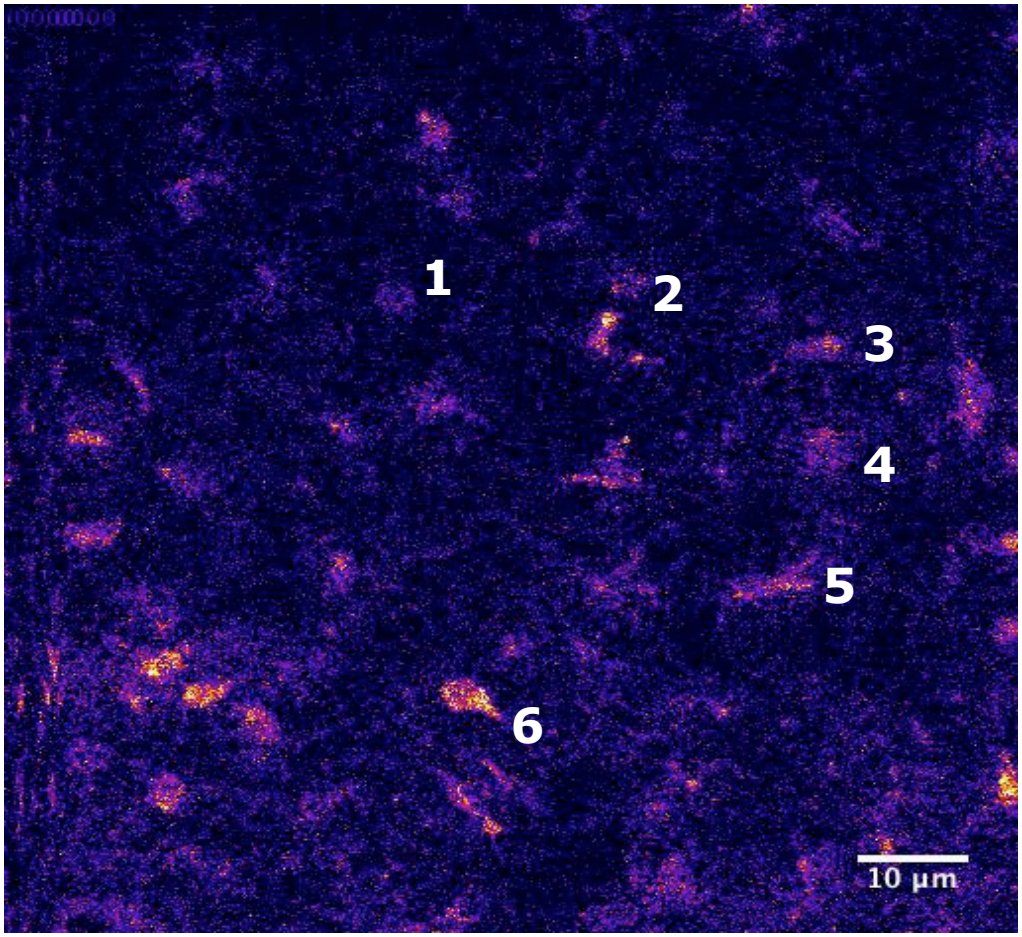
NATURE NEUROSCIENCE | VOL 22 | NOVEMBER 2019 | 1782–1792 | www.nature.com/natureneuroscience

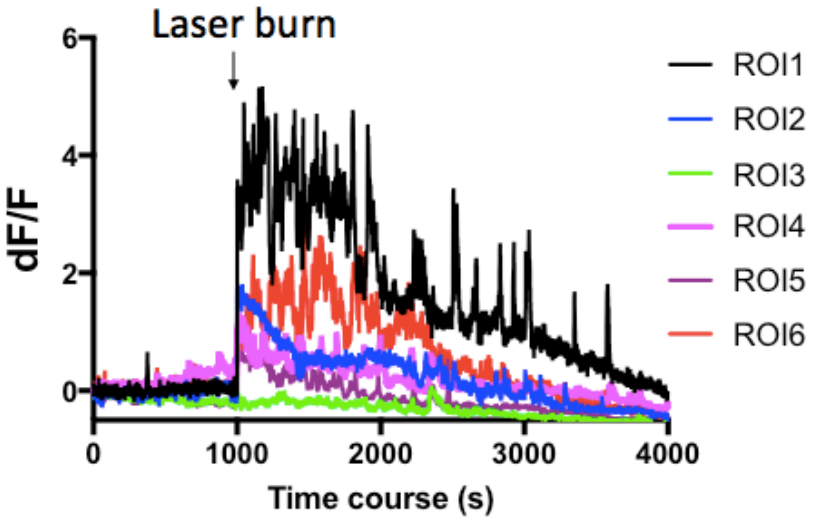
Summary

Biphasic neuronal activity increases microglial process dynamics



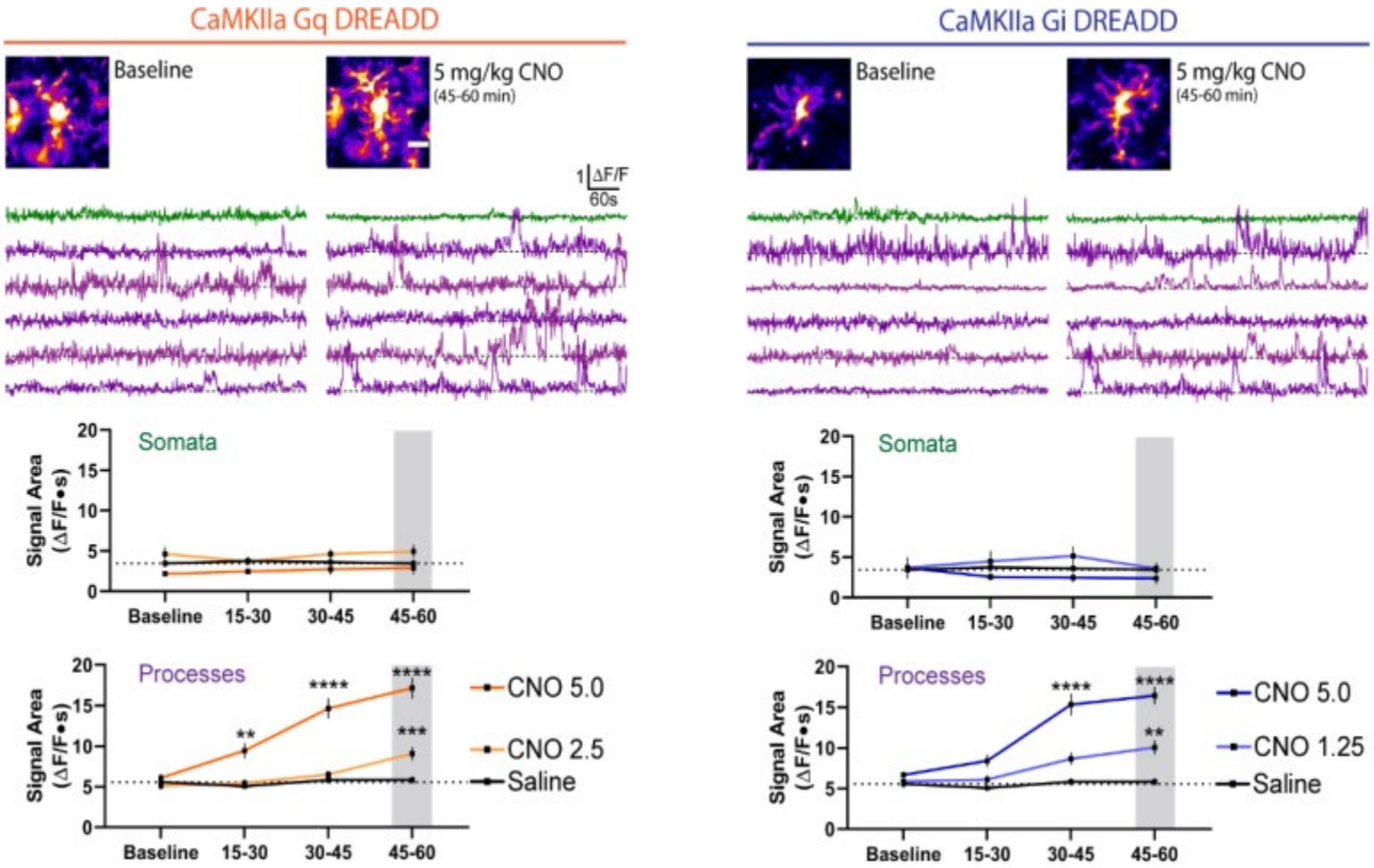
Microglial Ca^{2+} imaging in vivo





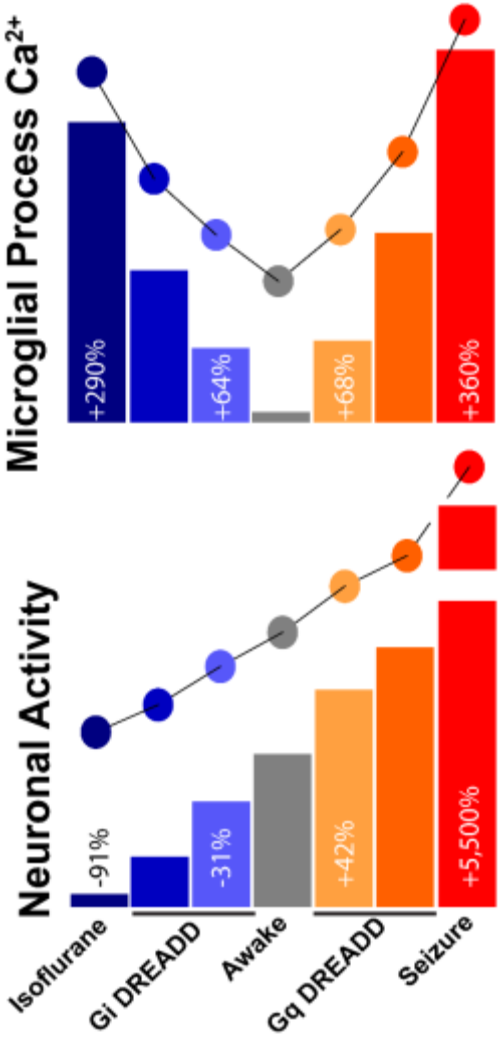
CX3CR1-CreER:ROSA-CAG-GCaMP6s mice

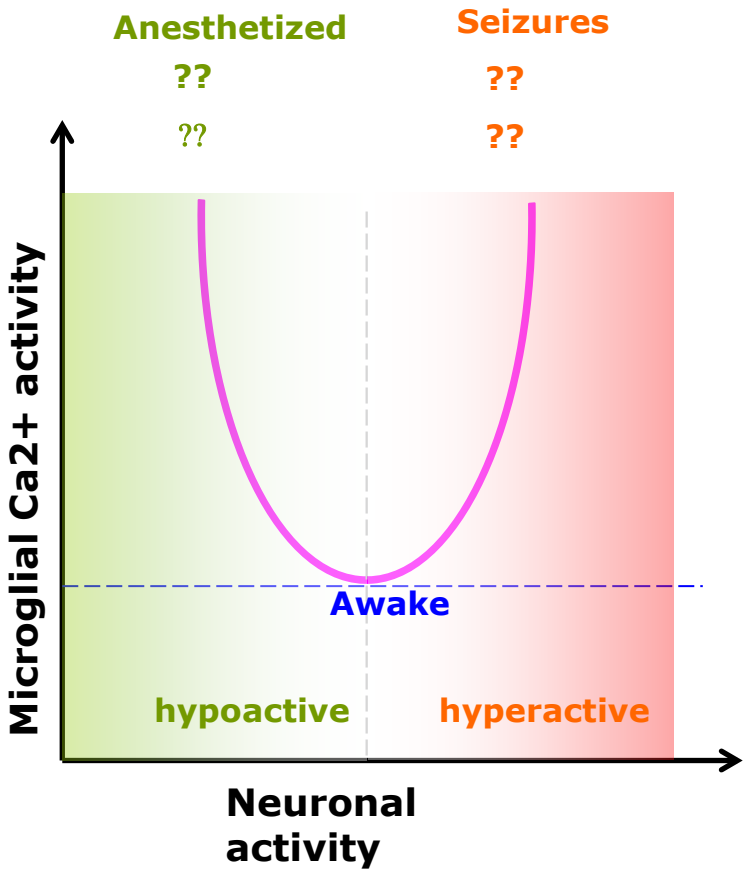
Microglial Ca^{2+} in response to biphasic neuronal activity



Umpierre et al., *eLife,* 2020

Biphasic neuronal activity increases microglial Ca^{2+} activity





Dr. Anthony Umpierre

Umpierre et al, *eLife,* 2020

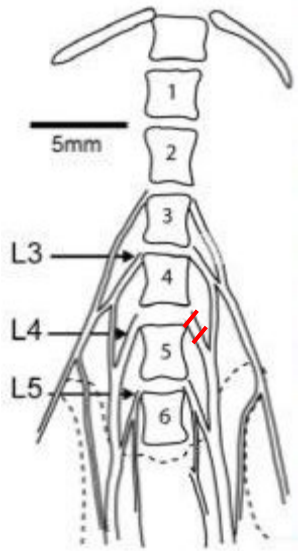
Overview

- How microglia sense neuronal activity?
- **What is the function of microglia in chronic pain?**

Microgliosis and neuropathic pain

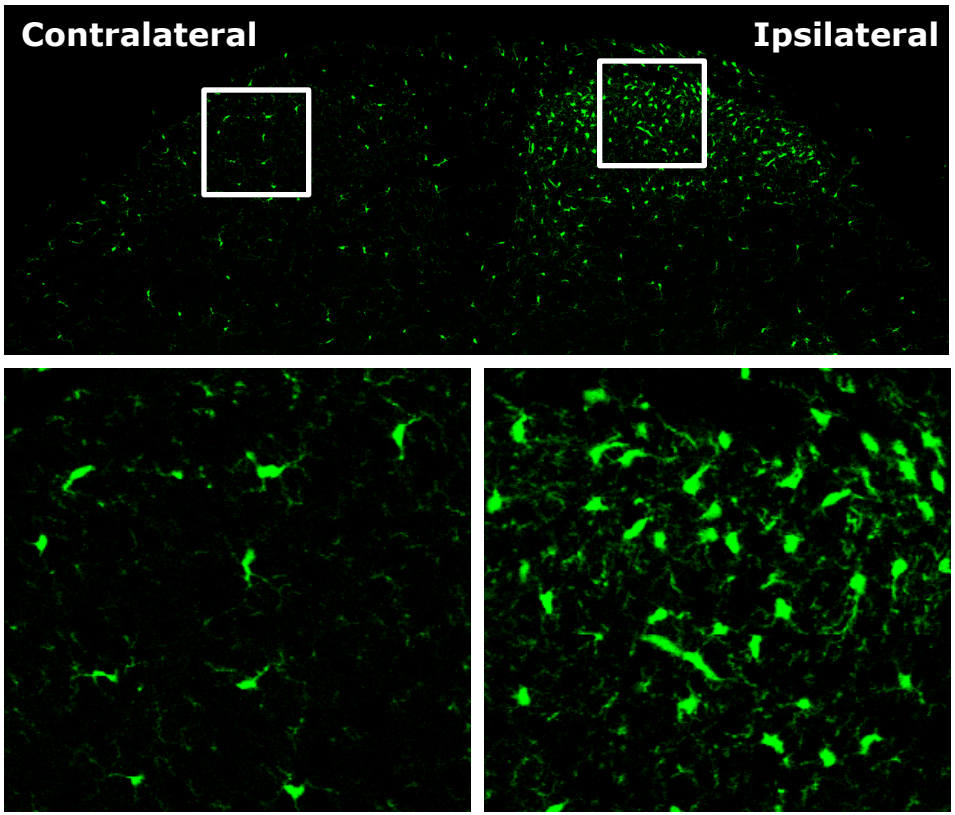
a

L4 spinal nerve transection (SNT)

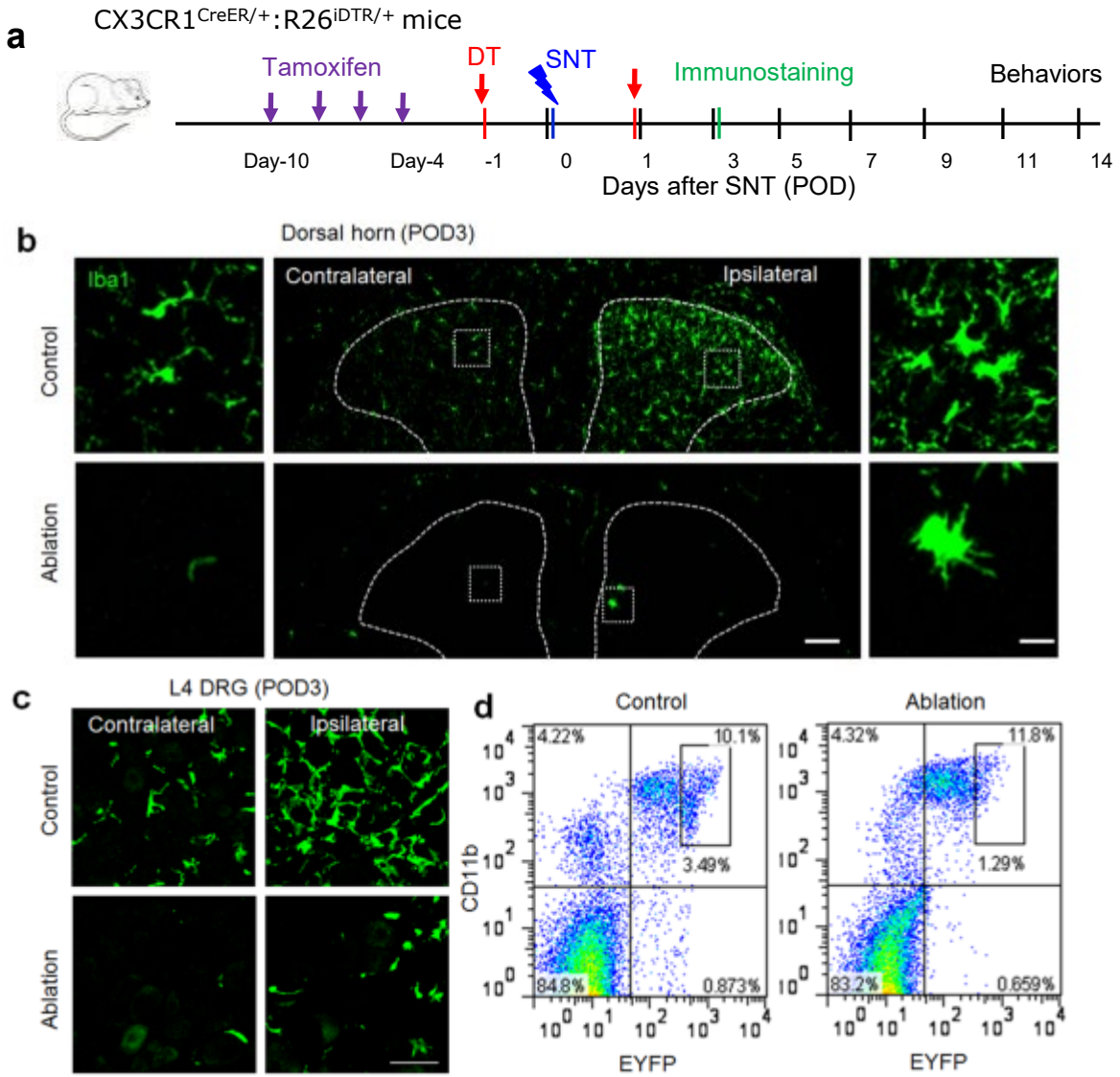


b

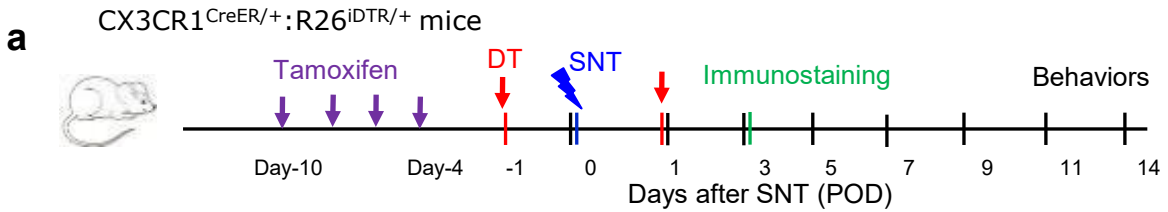
Day 7 after SNT

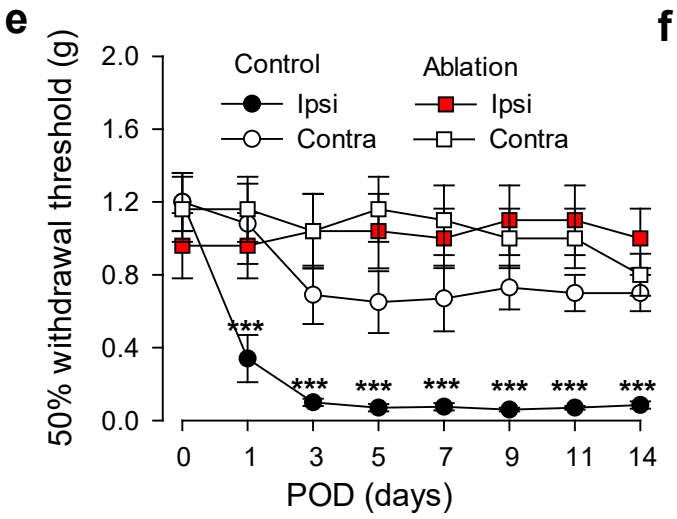


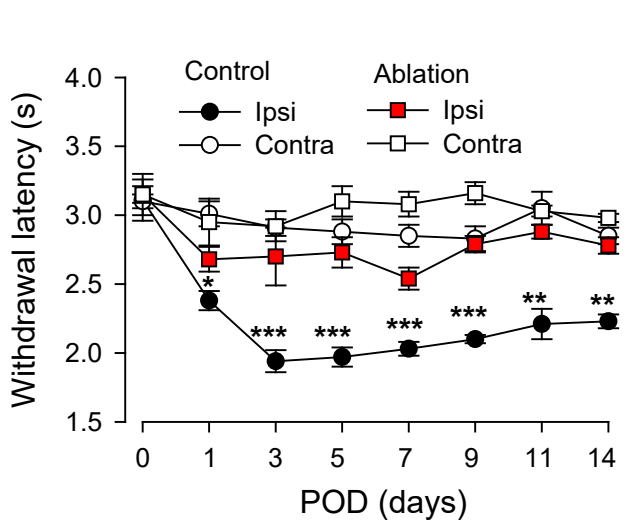
Microglia/monocyte ablation



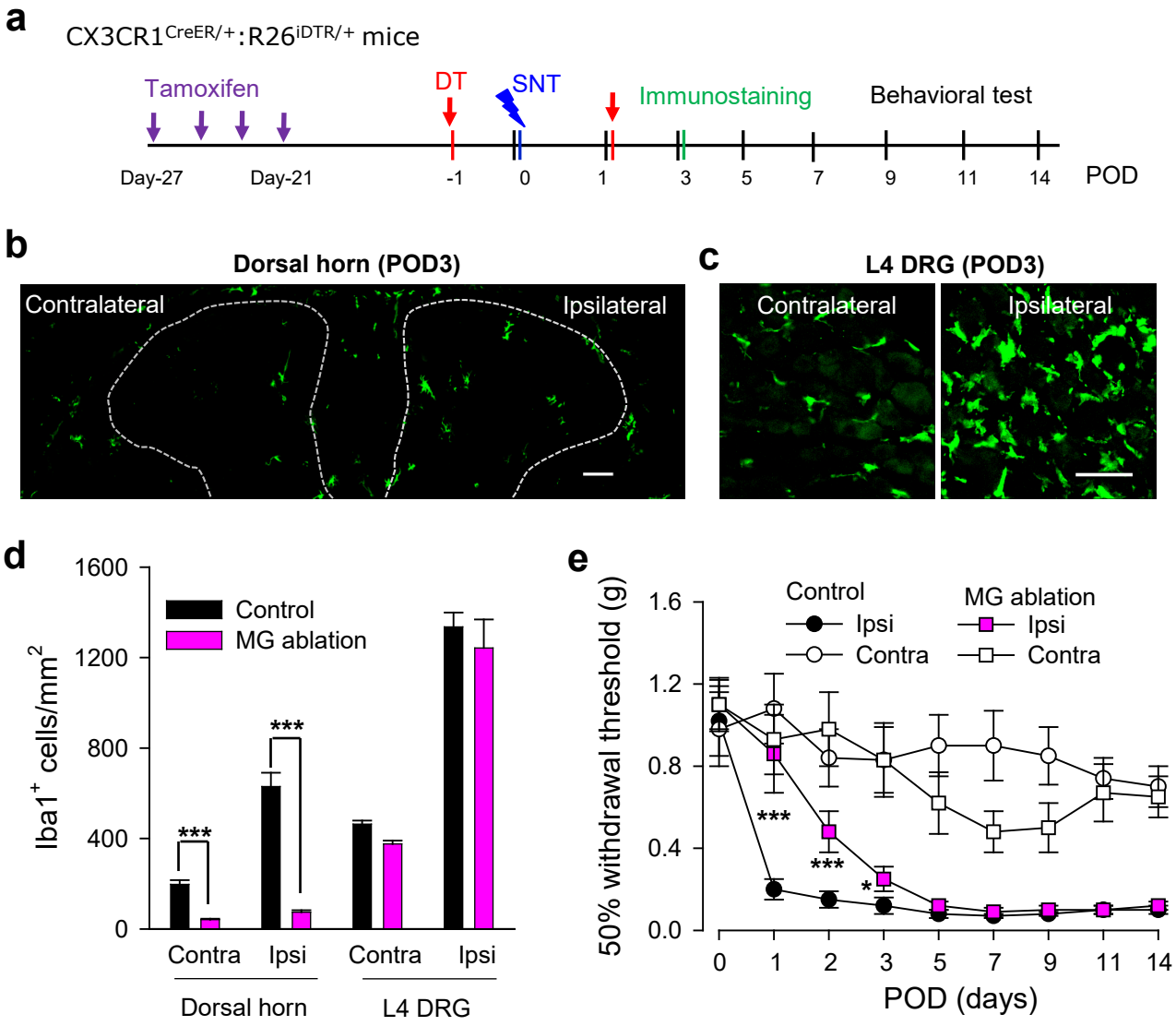
Microglia/monocyte ablation attenuated neuropathic pain





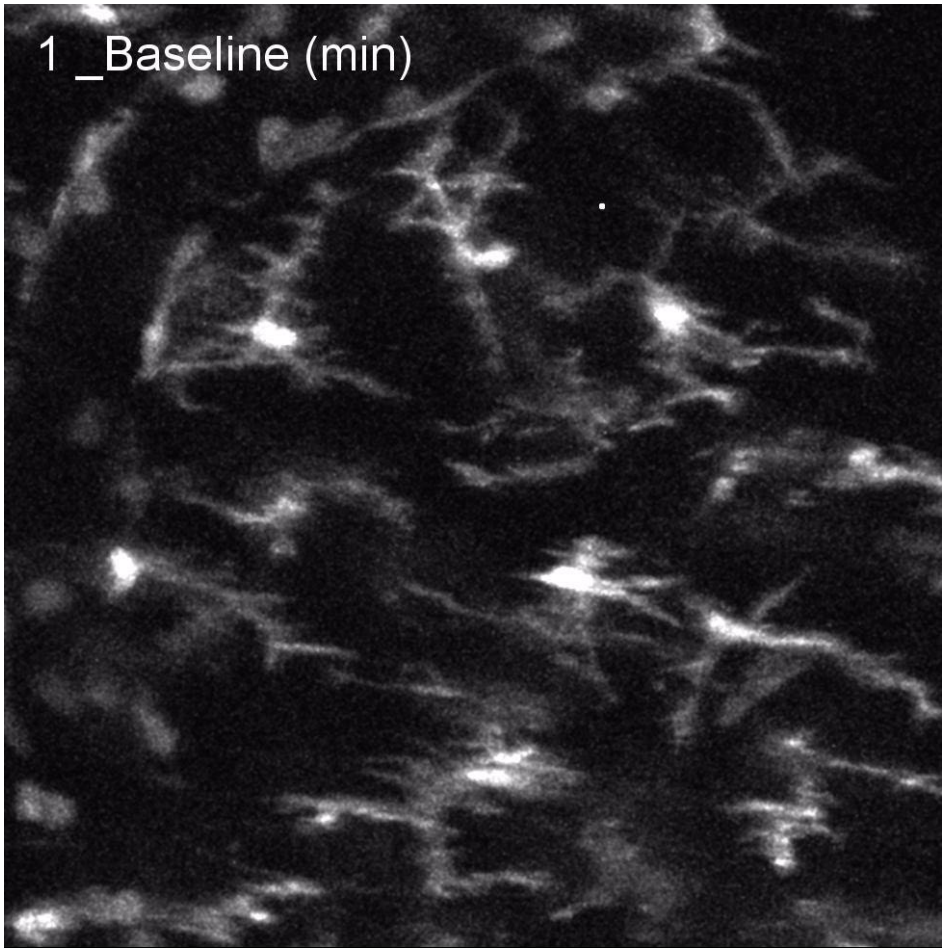


How about only microglia ablation?

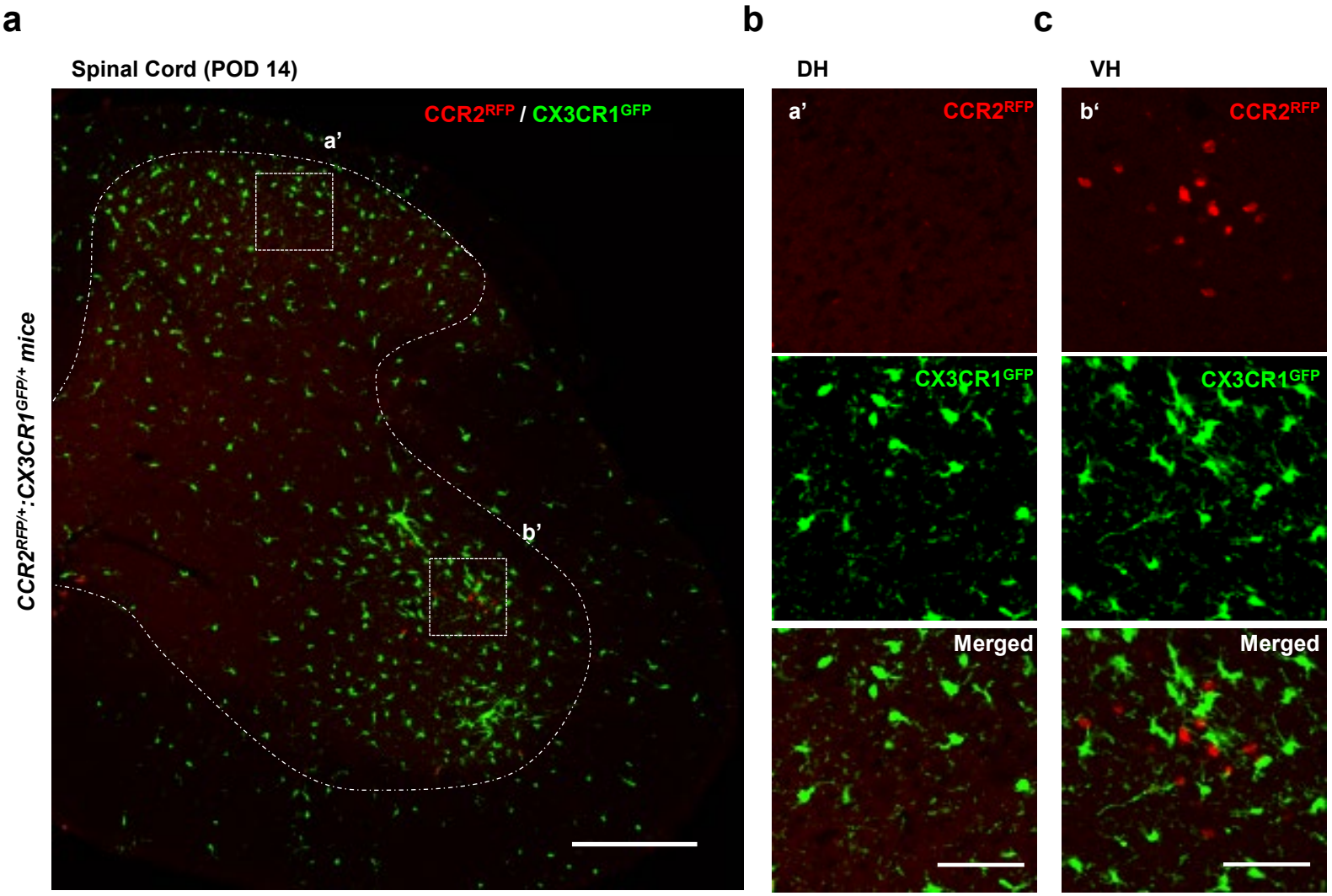


Resident microglia are required for chronic pain initiation

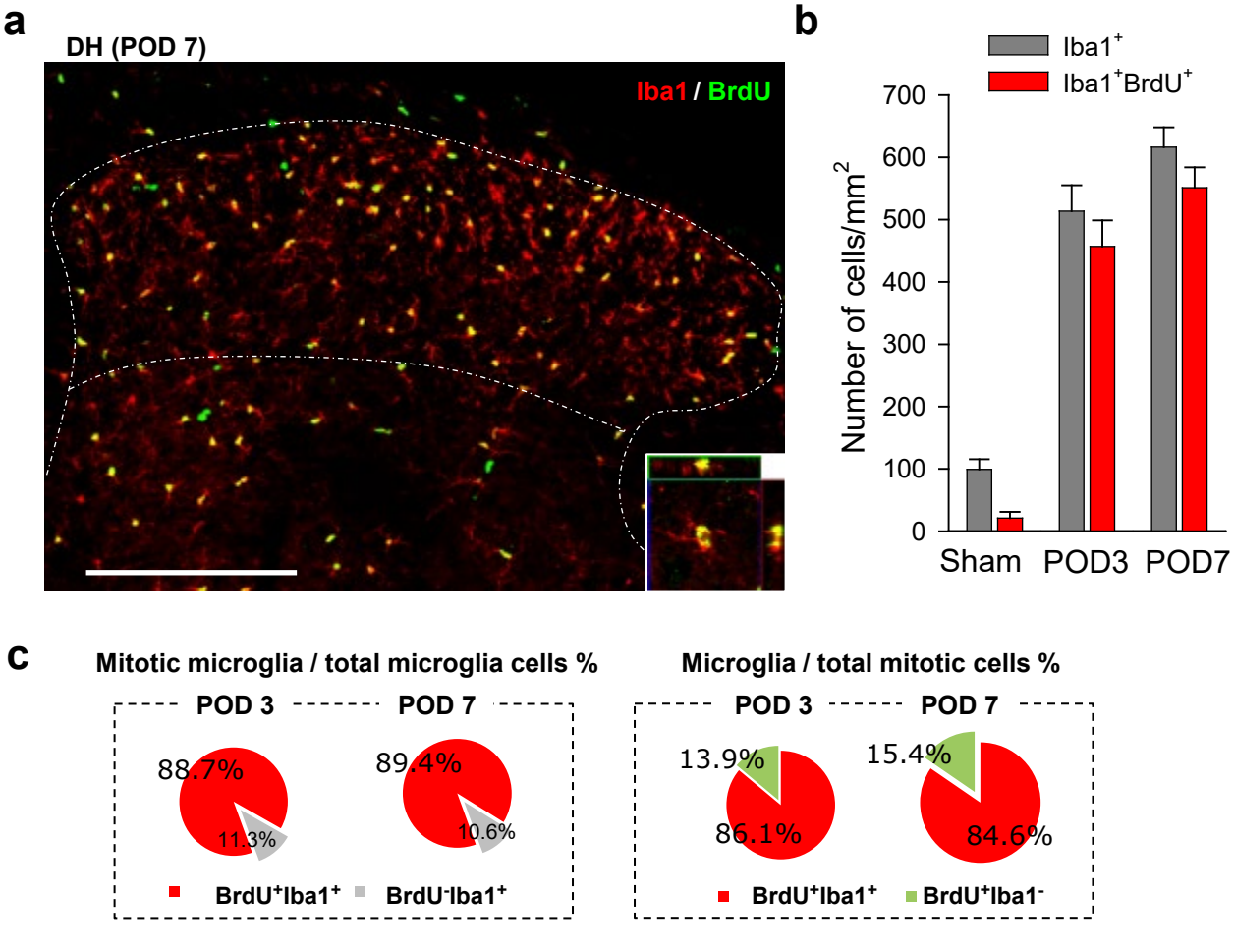
In vivo imaging of monocytes in the spinal cord



No monocyte infiltration using CCR2^{RFP} / CX3CR1^{GFP} mice

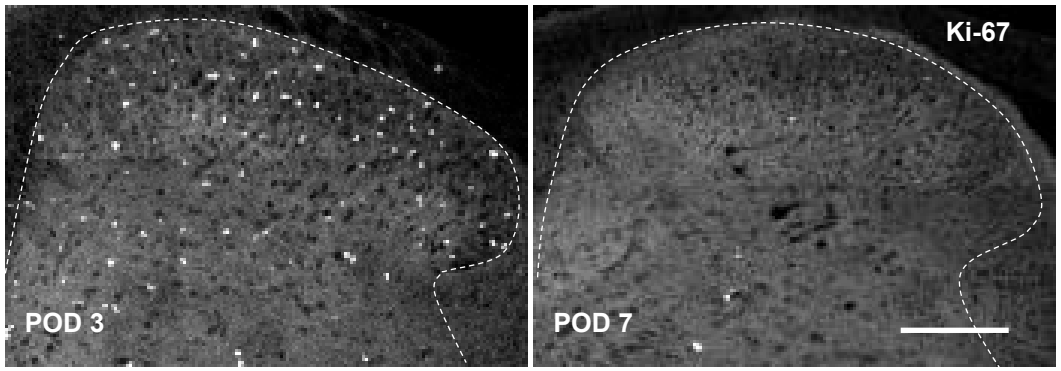


Microglial proliferation dominates microgliosis



Microglial proliferation peaks at day 3 after SNT

a



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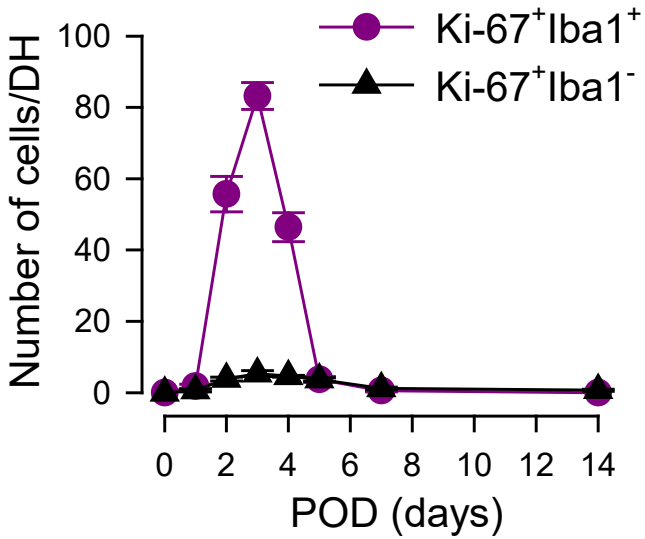
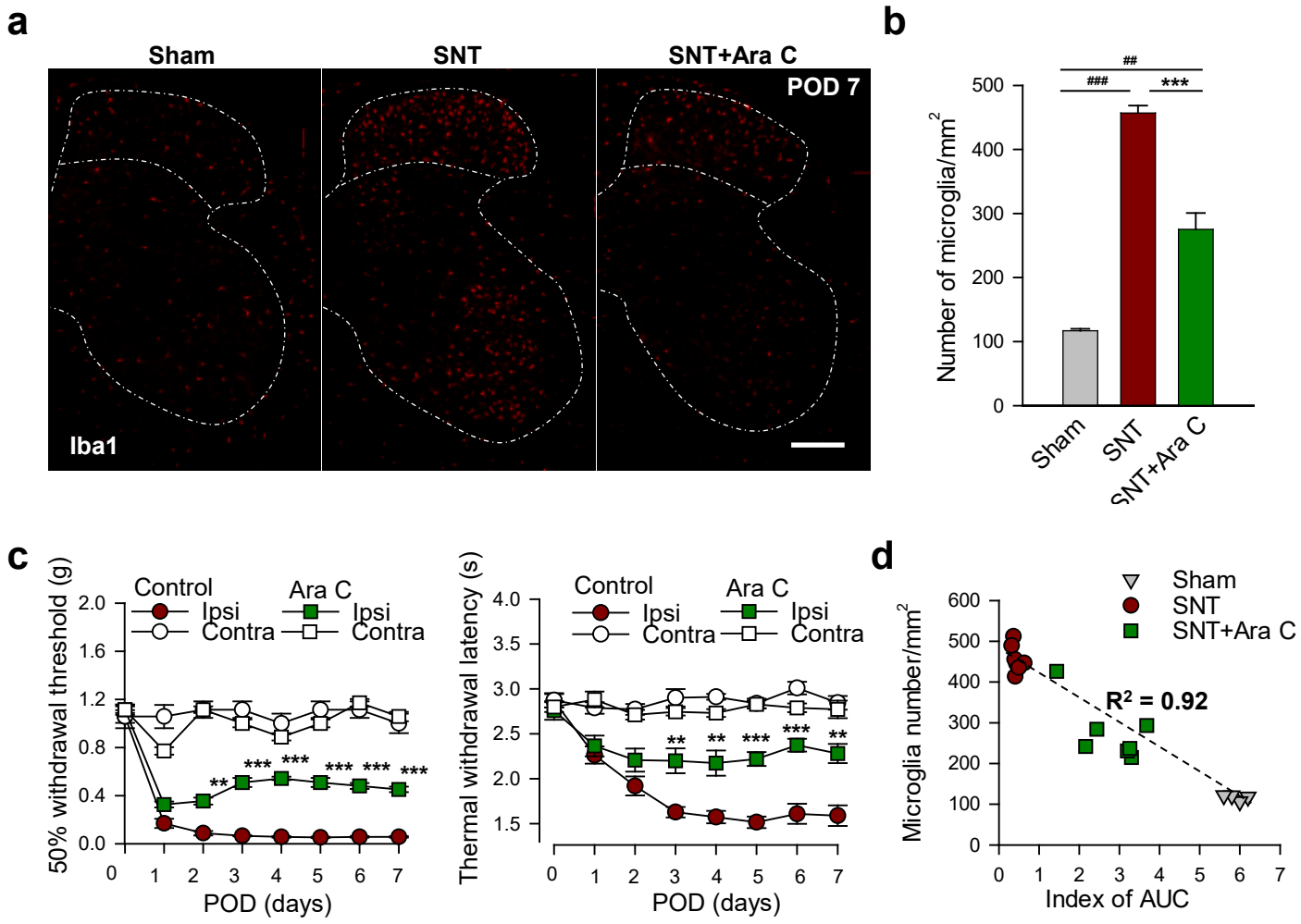
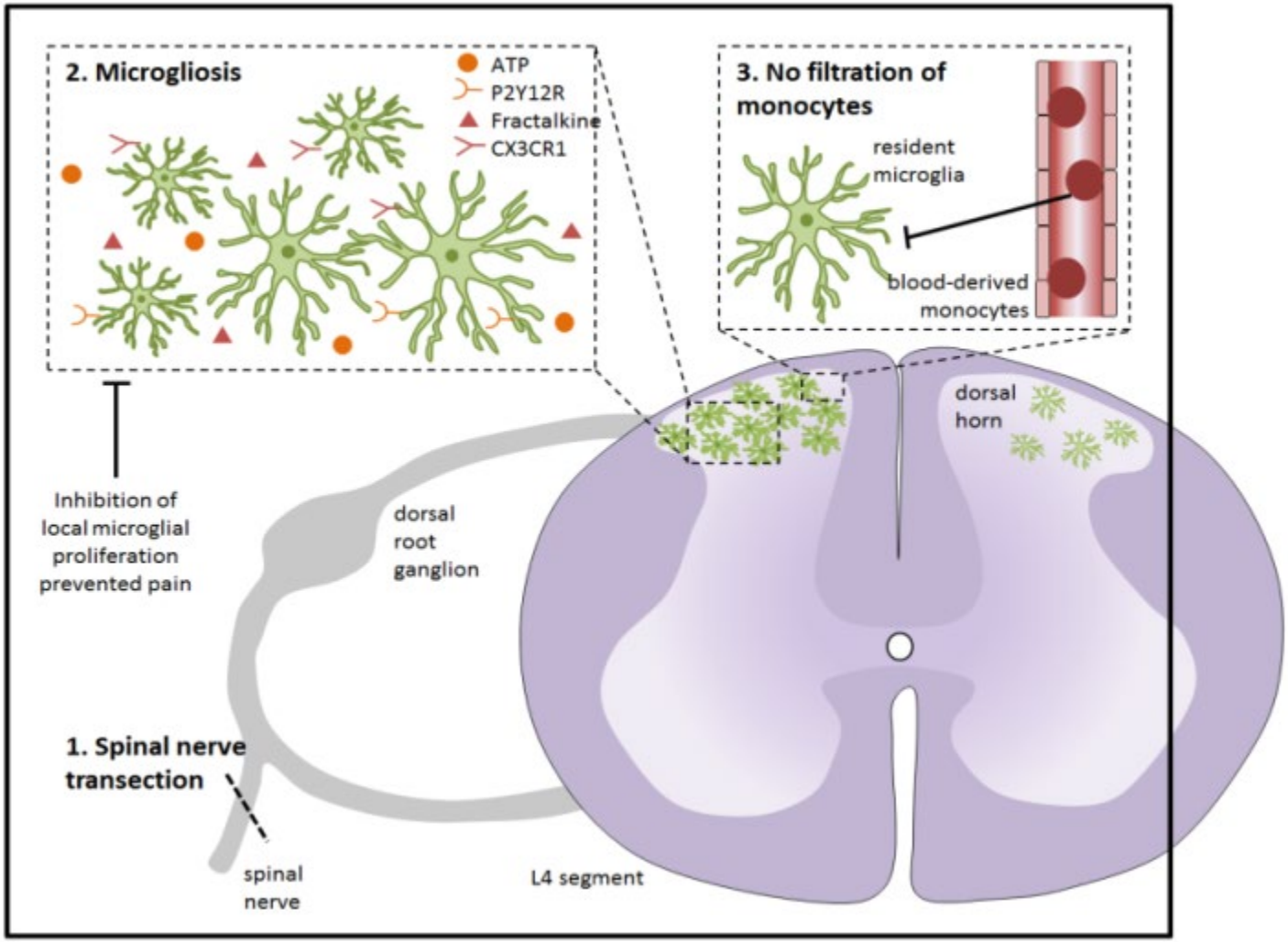


Microglial proliferation is correlated with neuropathic pain



Microglia and monocytes synergistically promote pain



Dr. Jiyun Peng

Now at Nanchang U

Peng et al., *Nat Commun*, 2016; Gu et al., *Cell Rep*, 2016

Question: chronic pain model without monocyte activation?

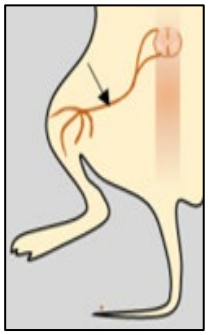
If so,

(1) What is microglial function?

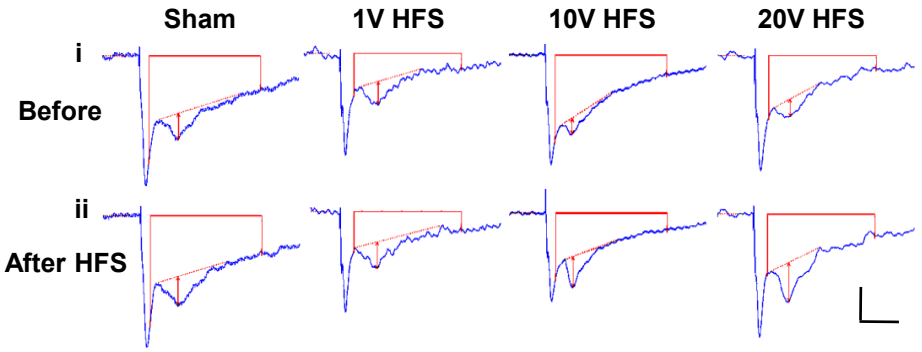
(2) What is microglial signaling involved?

HFS induces spinal LTP and chronic pain

a



Sciatic nerve sti, *in vivo* spinal recordings



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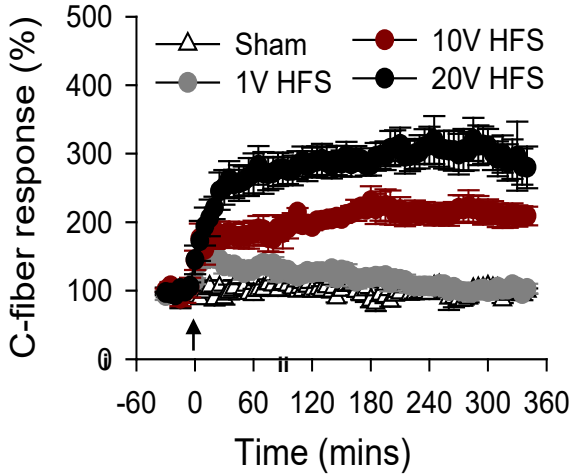
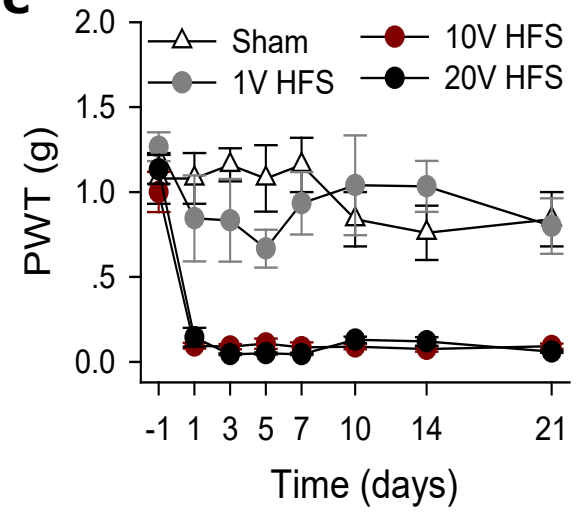


c



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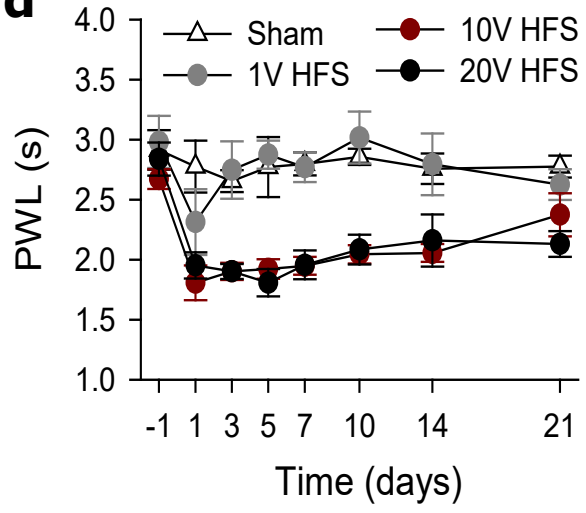
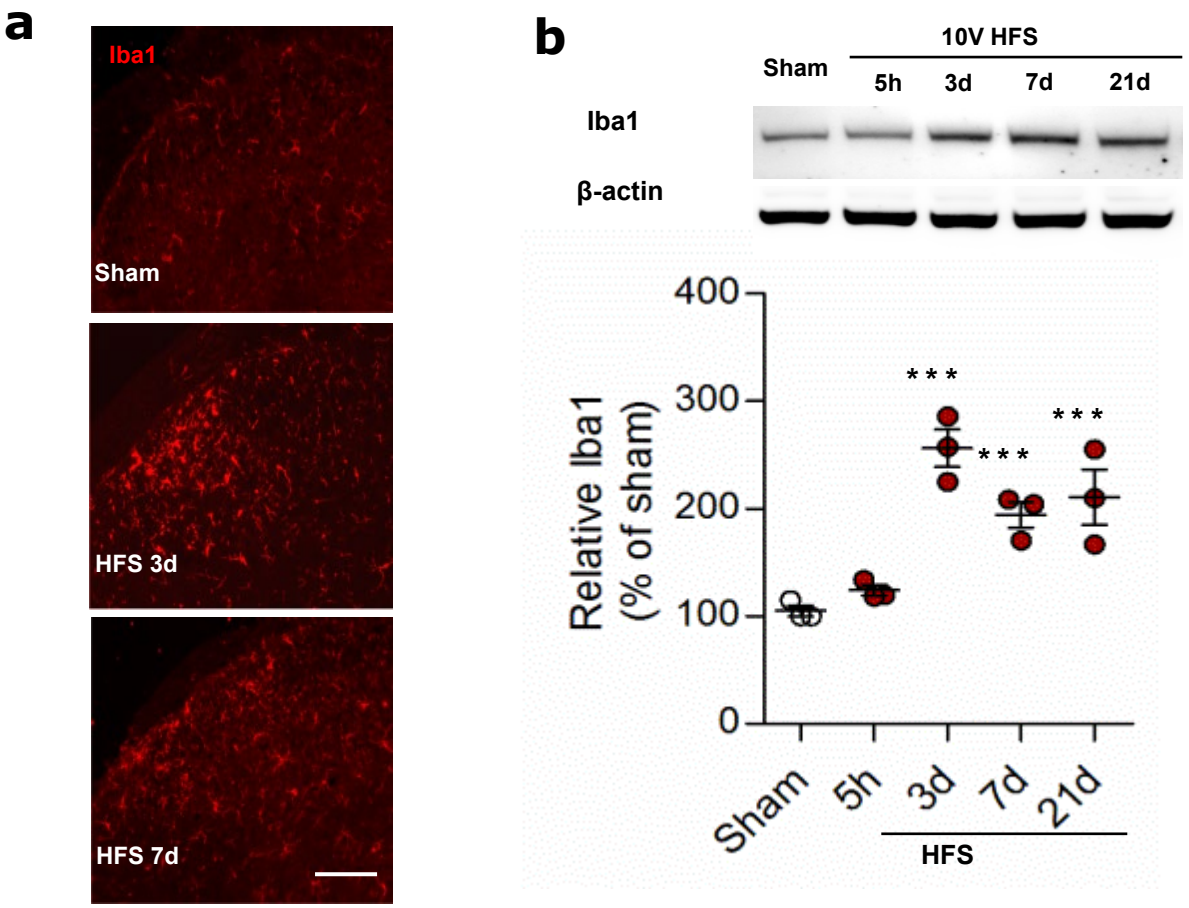


In collaboration with Dr. Xianguo Liu (Sen Yat-sen University)

HFS increases long-term microglial activation



HFS-induced chronic pain is a unique model

(1) Short-term stimulation but long-lasting pain

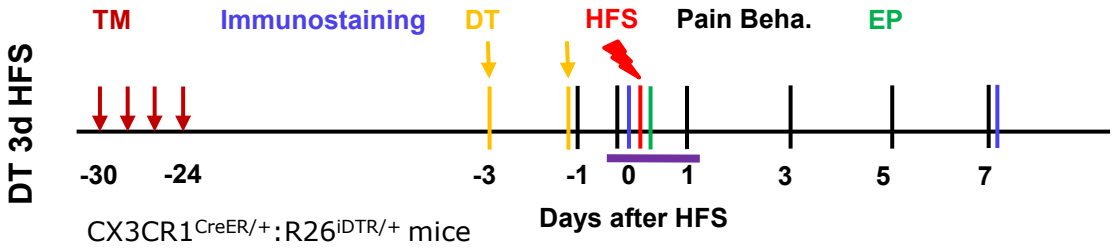
(2) No nerve injury

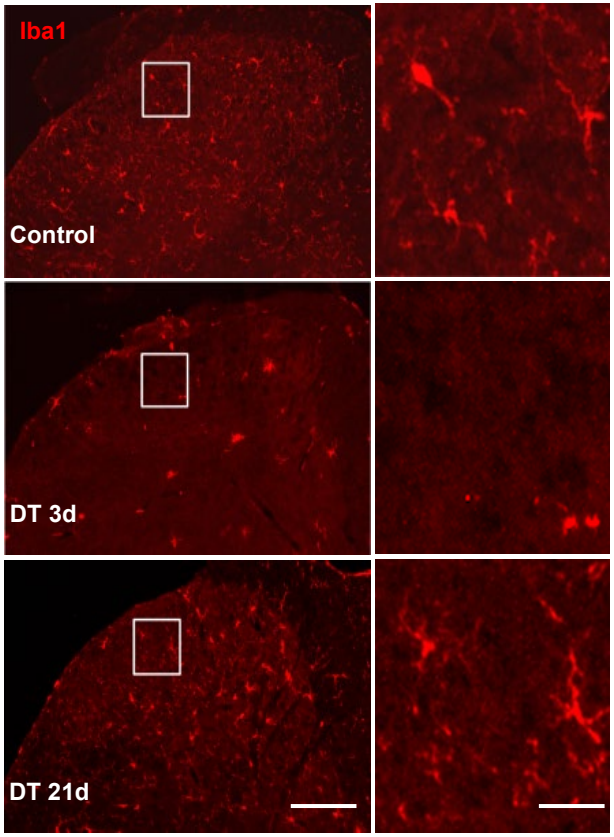
(3) Minimal peripheral inflammation

(4) Dramatic microglial activation

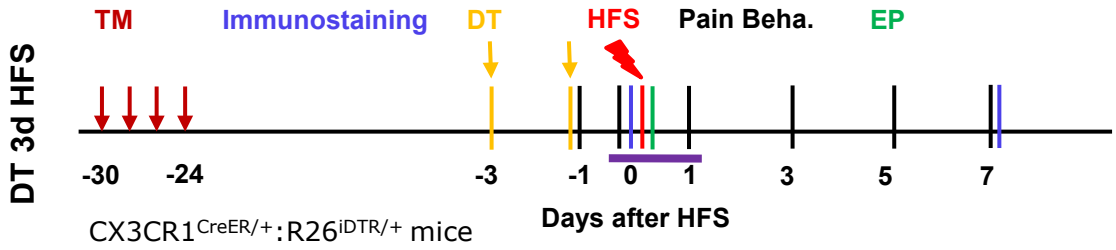
What is the role of microglia in HFS-induced chronic pain?

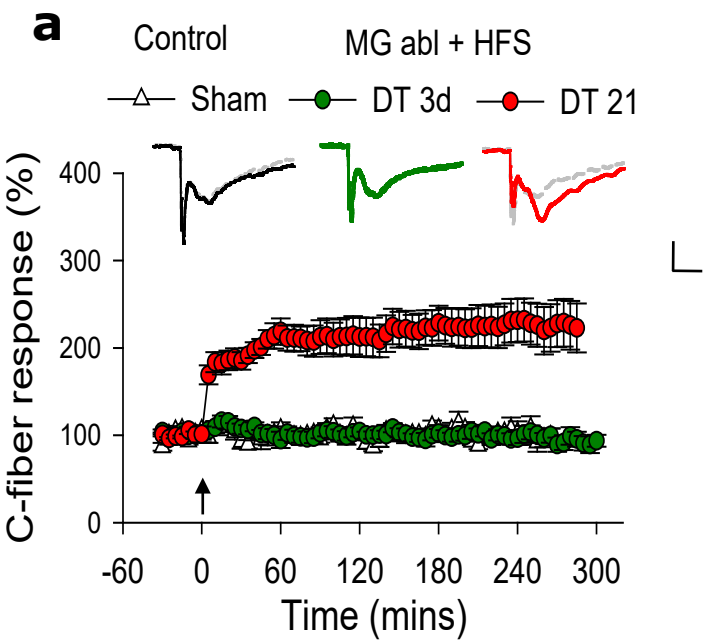
What is the role of microglia?

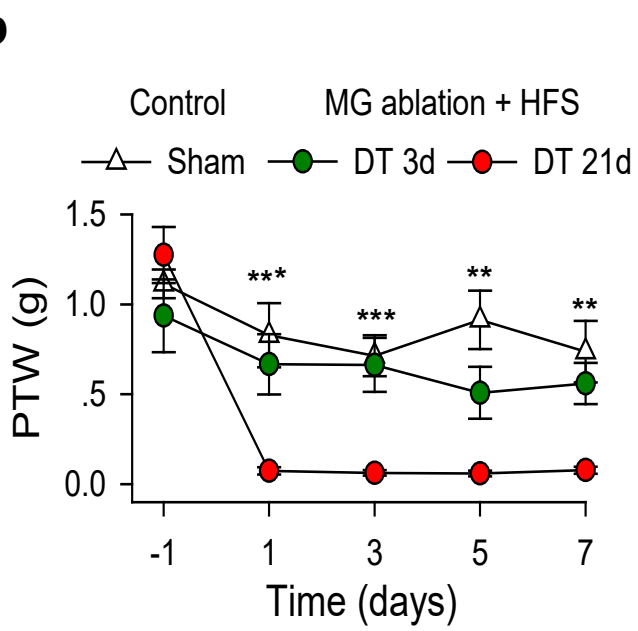




What is the role of microglia?

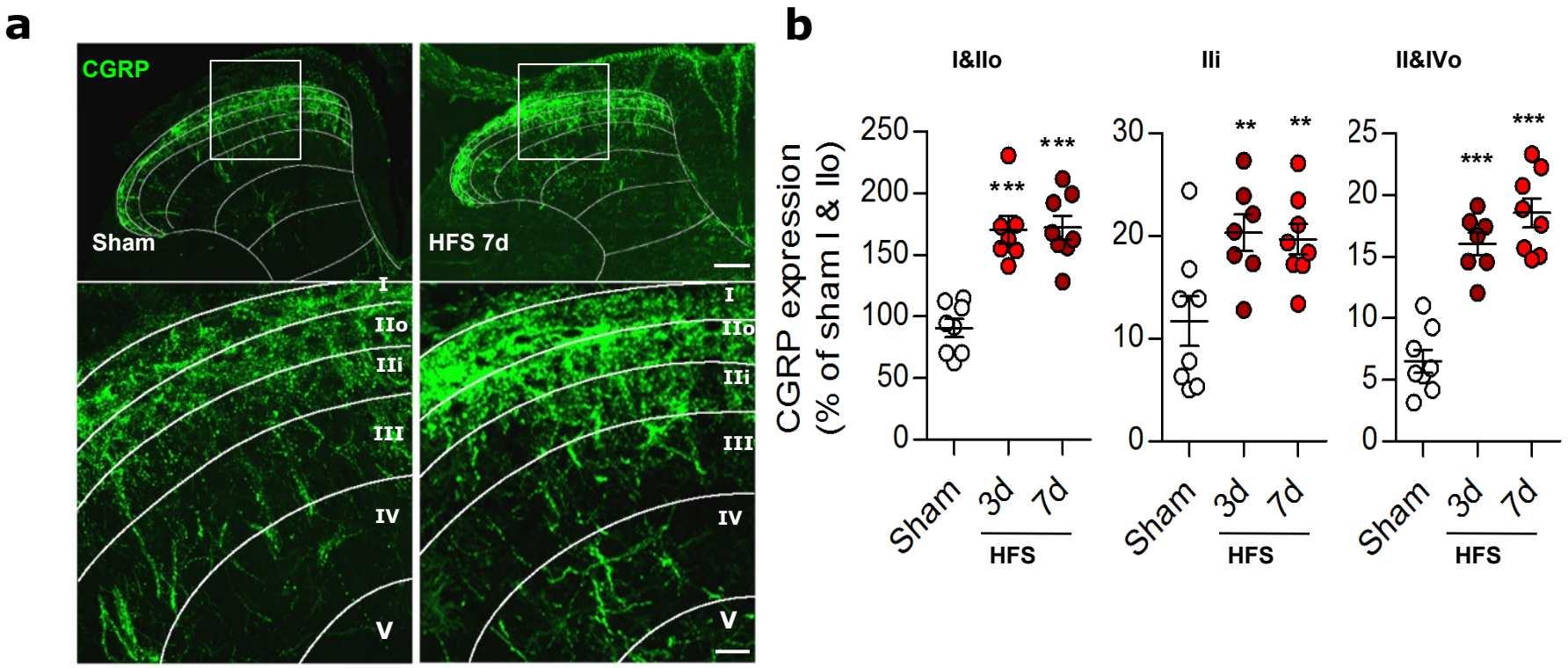






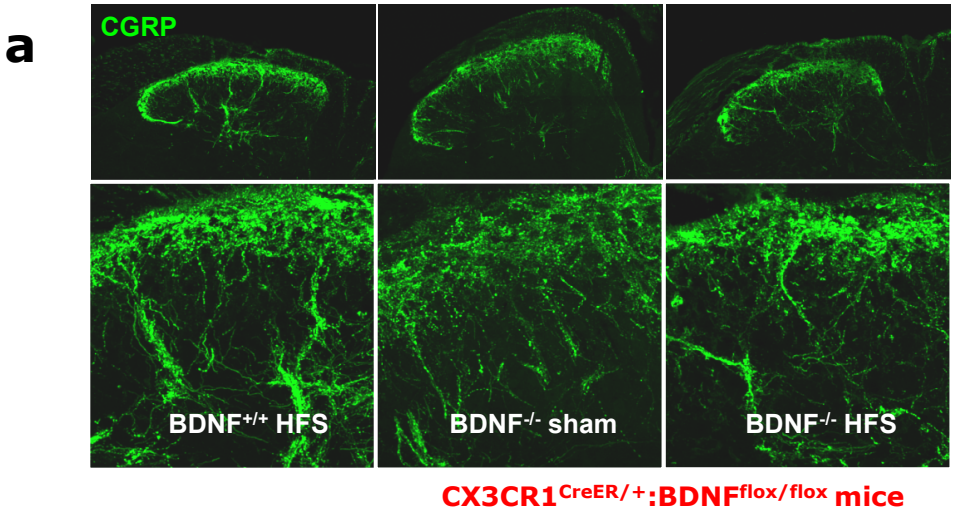
What is microglial mechanism underlying acute to chronic transition?

HFS increases the nerve sprouting

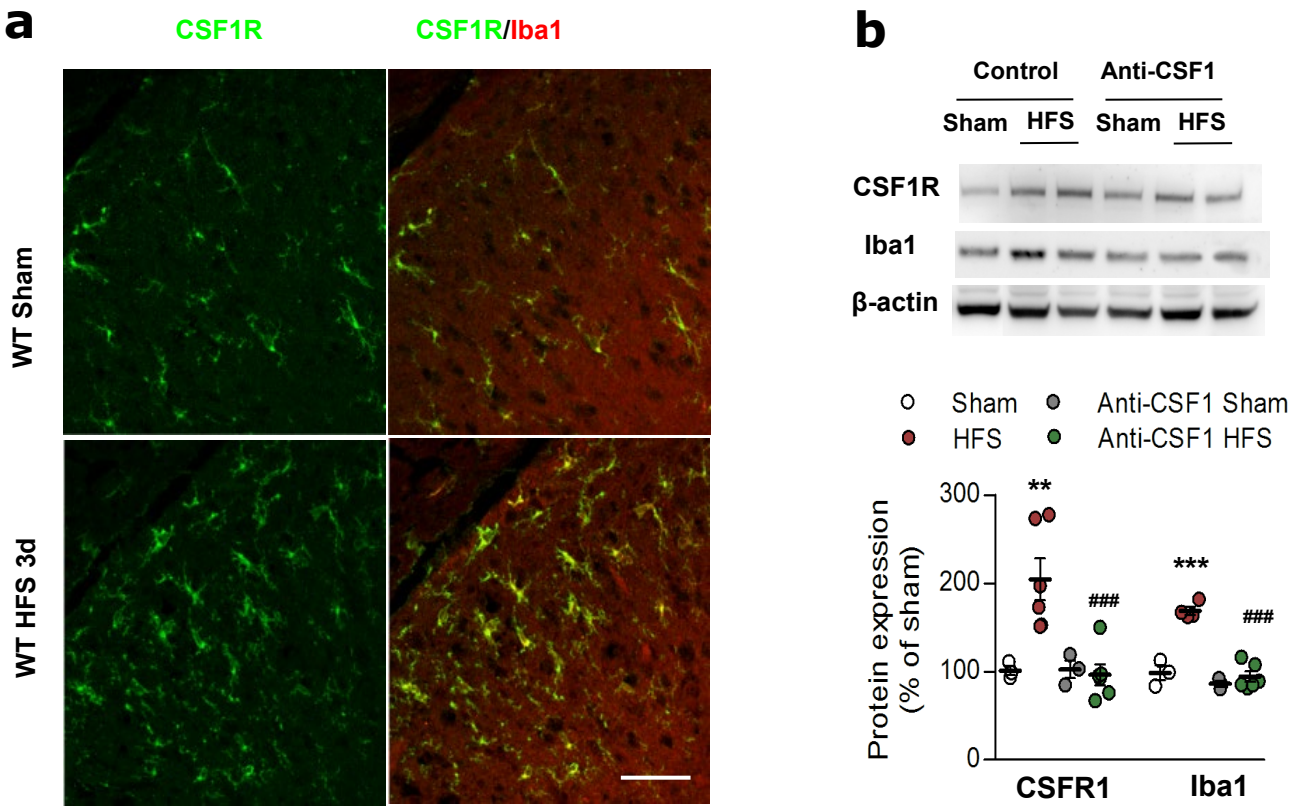


Microglial BDNF involved?

Microglial BDNF in HFS-induced nerve spouting and pain

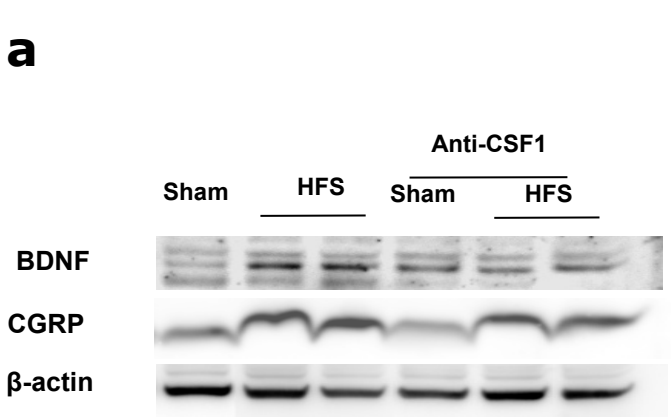


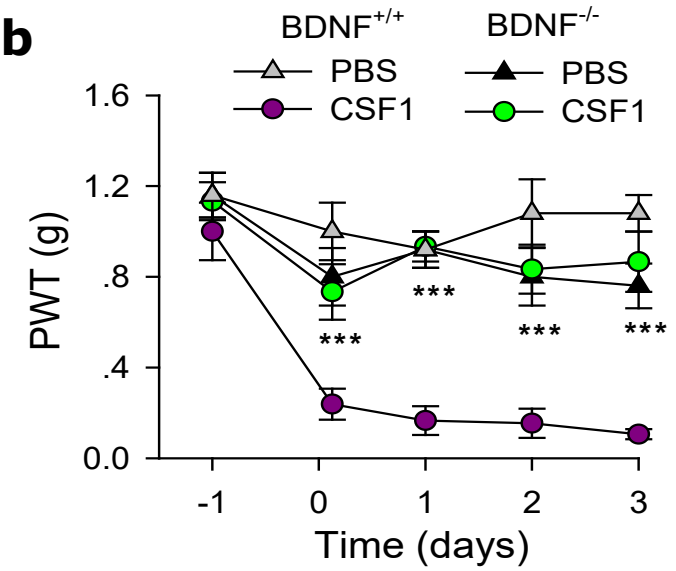
How microglia get activated after HFS?



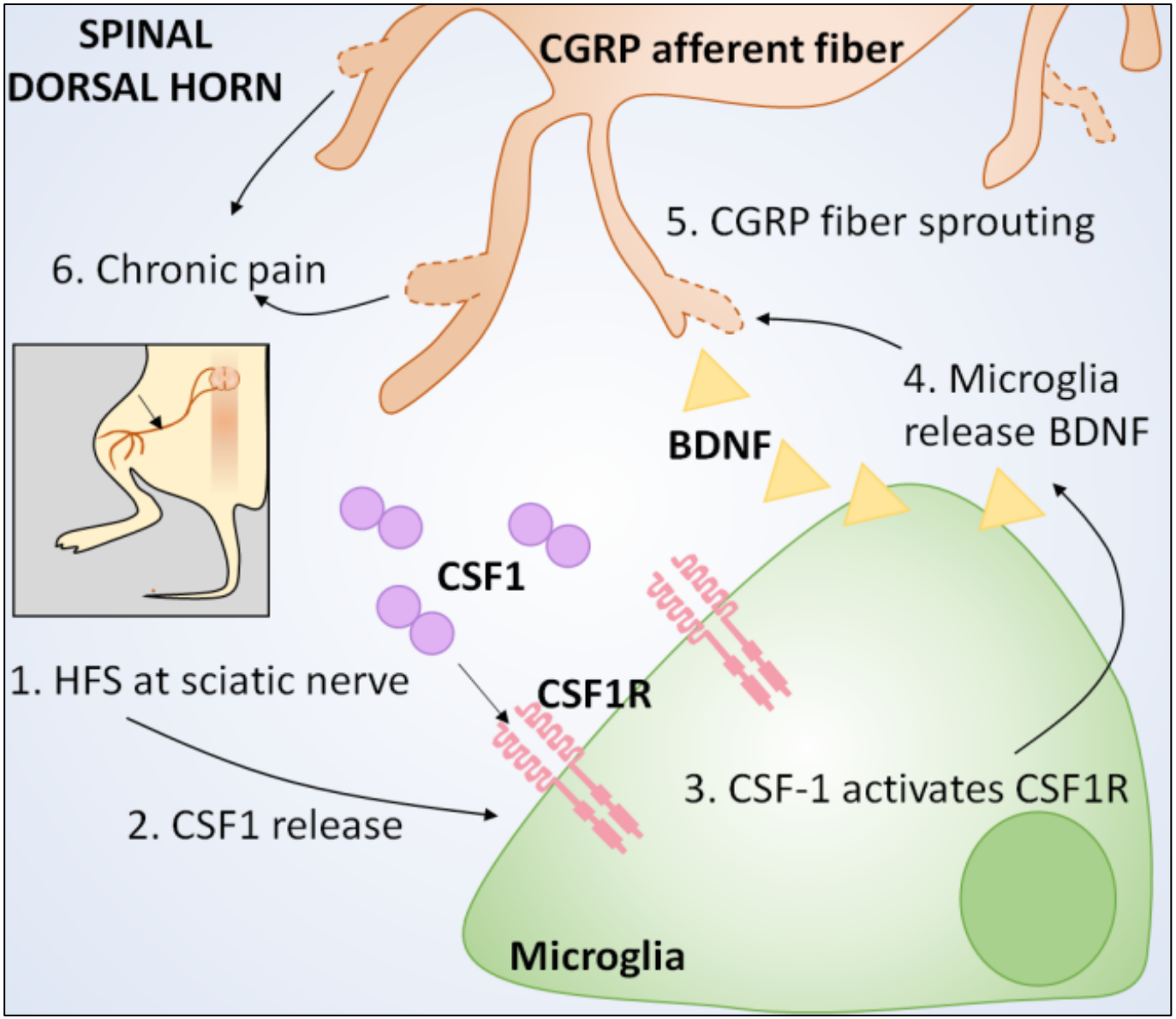
HFS activates microglia via CSF1 signaling

Microglial CSF1R and BDNF?





CSF1 signaling is coupled to BDNF in HFS-induced pain



Zhou et al., *Cell Rep*, 2019

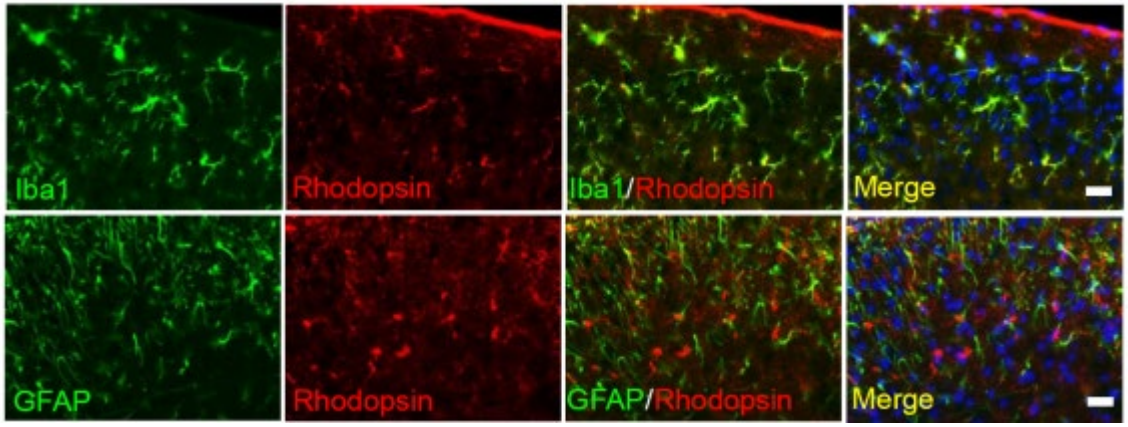
Dr. Lijun Zhou

Now at Sun Yat-sen U

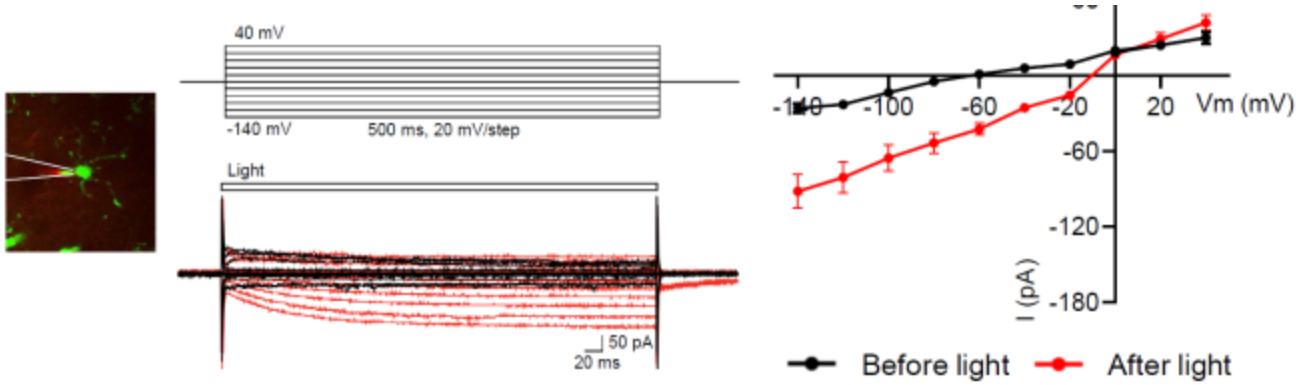
How can we directly manipulate microglia to study their function in chronic pain?

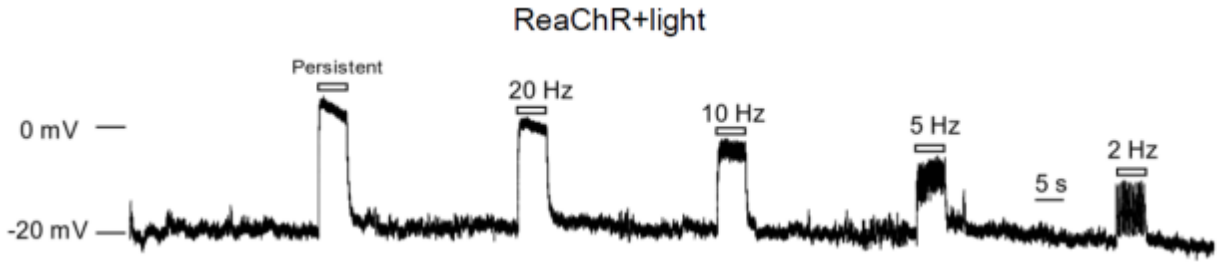
Optogenetics and chemogenetics

Red-activated channelrhodopsin (ReaChR) in microglia

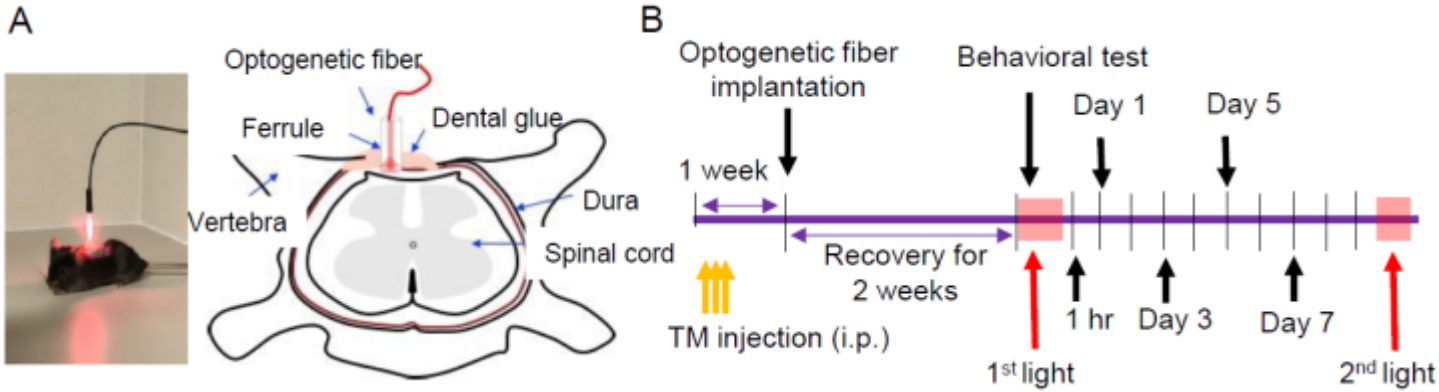


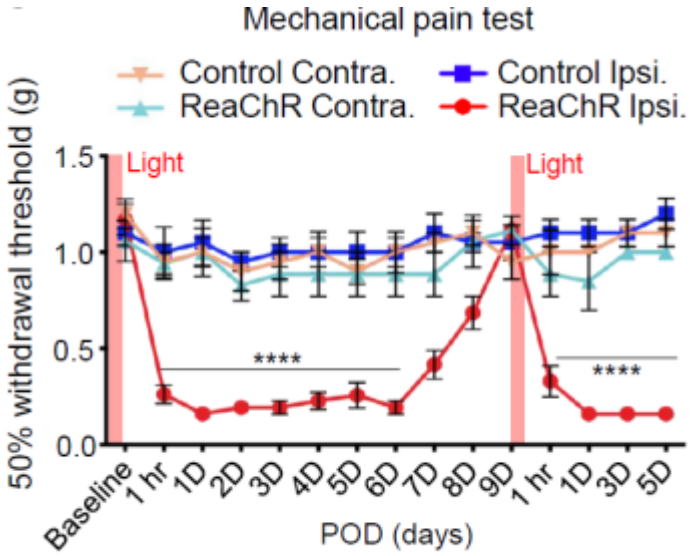
CX3CR1$^{creER/+}$: R26$^{LSL\text{-}ReaChR/+}$ mice

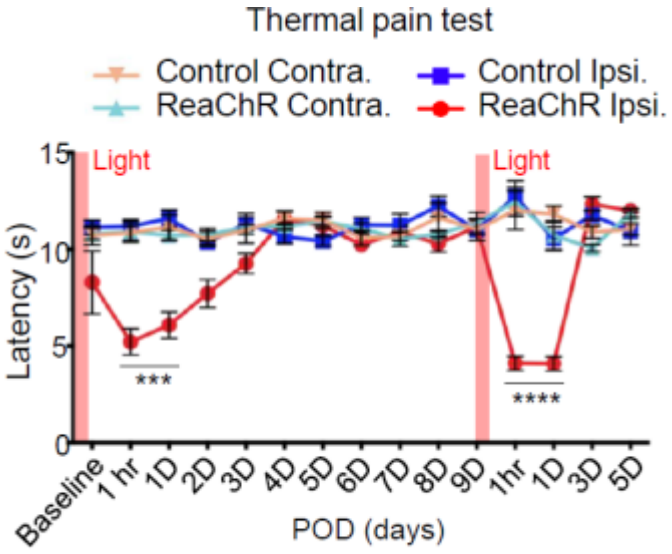




Activation of microglial ReaChR triggers chronic pain

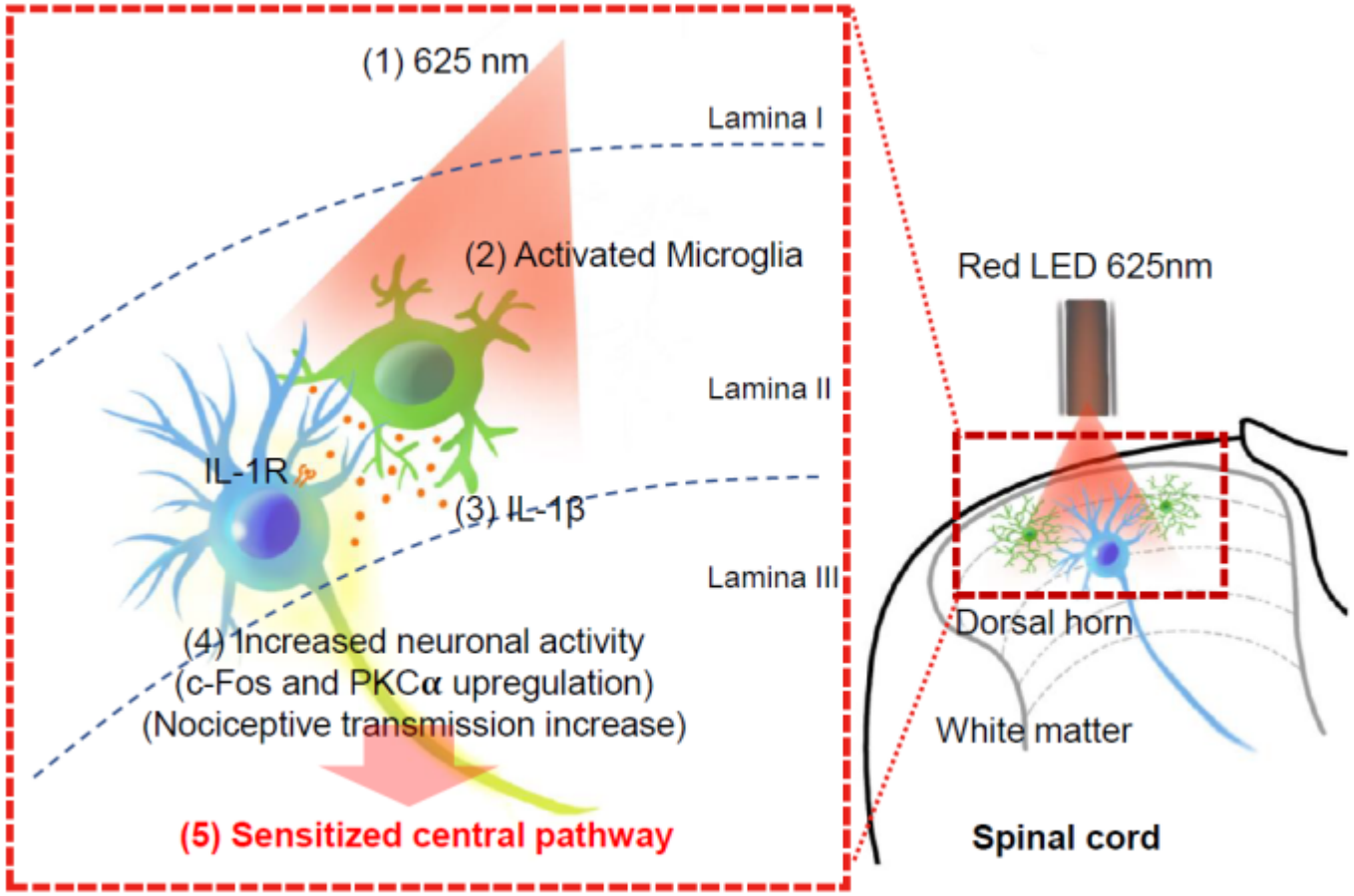






625 nm, red LED, 20 Hz, for 30 min

Microglial ReaChR induces chronic pain via IL1β signaling



Dr. Min-Hee Yi

Yi et al., BioRxiv, 2020; Yi et al., *Plos Biol*, under revision

Summary

- **How microglia sense neuronal activity?**

  Microglia sense neuronal hyperactivity via ATP-P2Y12 signaling

  Microglia sense neuronal hypoactivity via NE-β2 signaling

  U shape microglial responses to neuronal activity

- **What is the function of microglia in chronic pain?**

  Microglial proliferation induces microgliosis and neuropathic pain after nerve injury

  HFS-induced chronic pain requires microglial CSF1R signaling

  Optogenetic activation of spinal microglia induces chronic pain via IL1β signaling

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