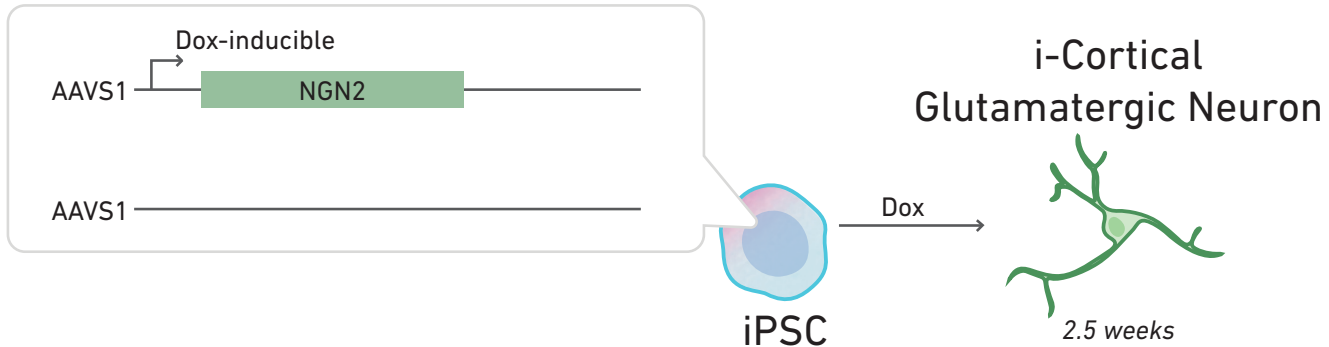


NGN2-induced Differentiation Protocol for KOLF2.1J AAVS1-TO-NGN2 KI/WT

Product Code: JIPSC002070

Cell line Name: AAVS-TO-NGN2 KI/WT

Parental Line: KOLF2.1J

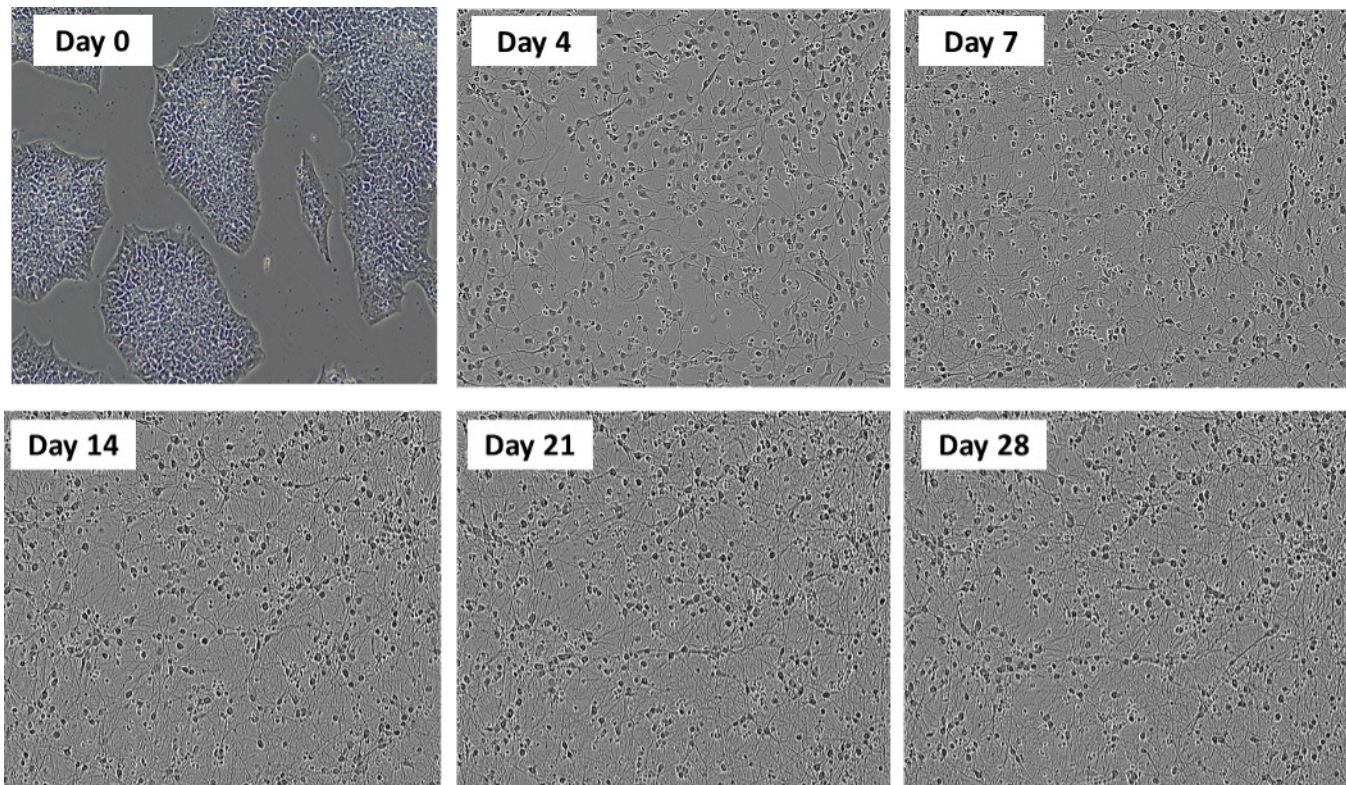


Protocols:

The standard protocol for thawing, propagating, and freezing KOLF2.1J lines is available on the JAX iPSC webpage (jax.org/ipsc).

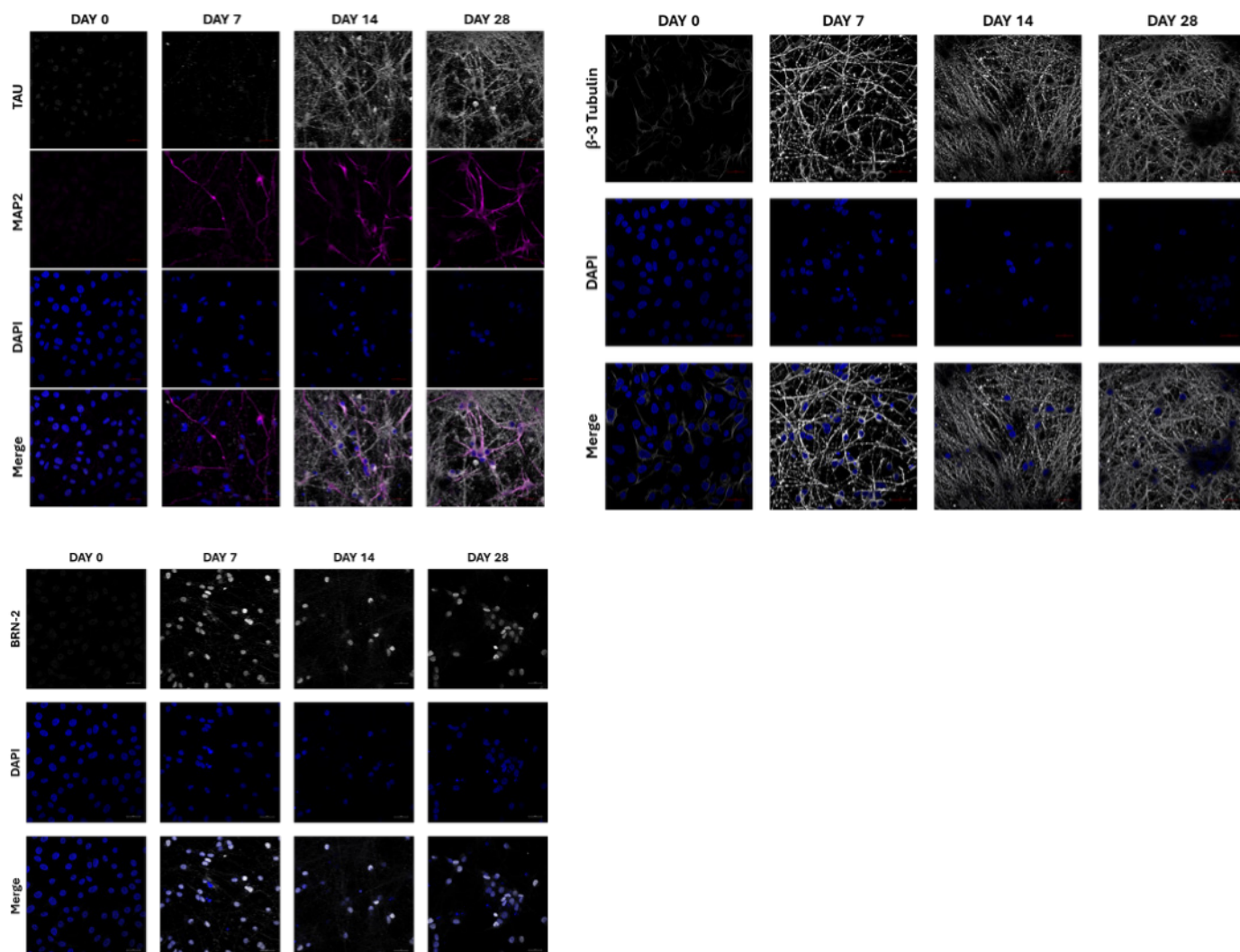
For a detailed NGN2-induced differentiation protocol, please visit:

<https://www.protocols.io/view/indi-transcription-factor-ngn2-differentiation-of-cvpxw5pn>

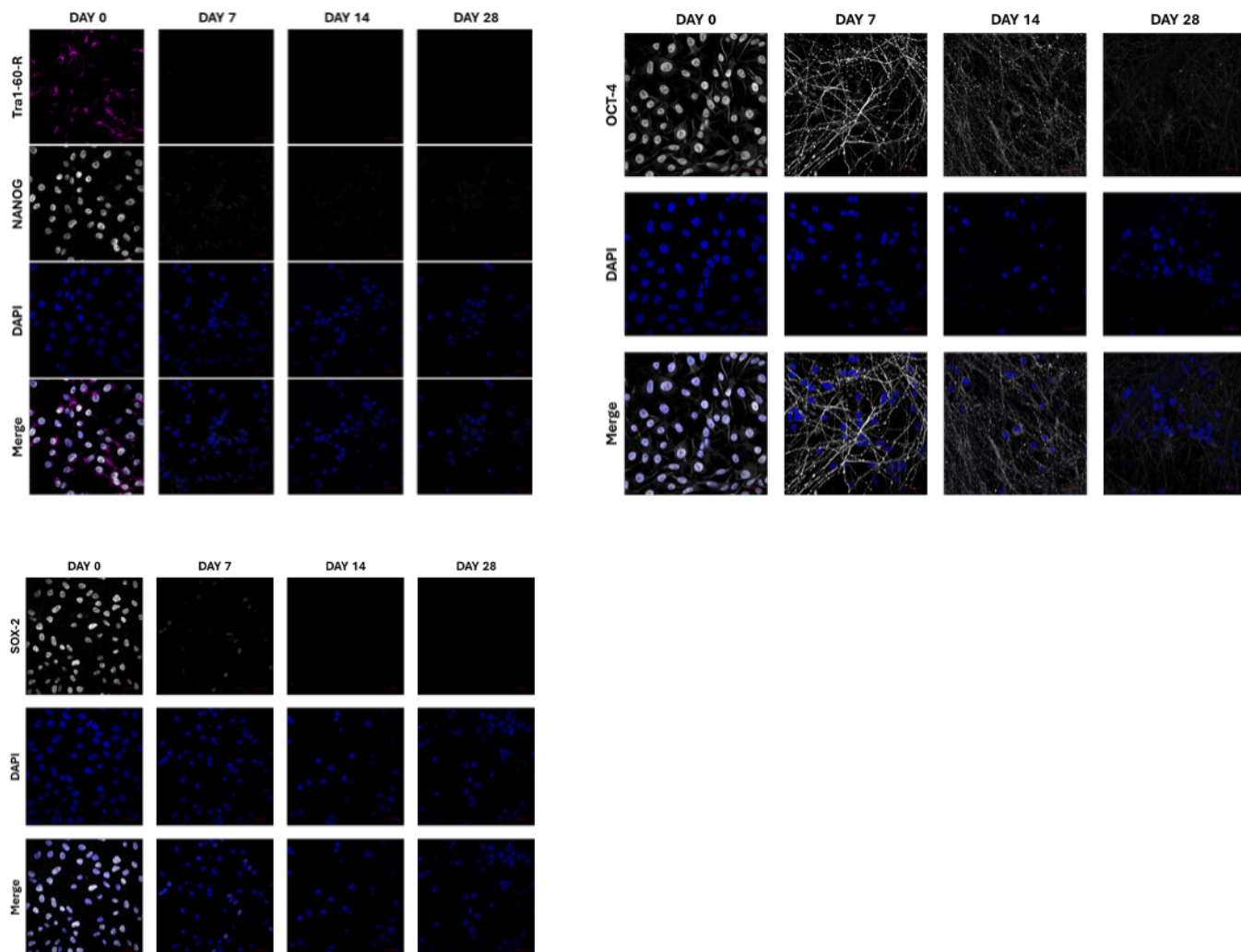


Longitudinal images of KOLF2.1J AAVS-TO-NGN2 neuron differentiation following doxycycline treatment.

On day 0 (prior to doxycycline addition), iPS cells remain in a pluripotent state. Upon doxycycline induction, iPS cells begin differentiating into neurons.



Induction of NGN2 in KOLF2.1J AAVS-T0-NGN2 cells upon doxycycline treatment leads to neuronal differentiation.
Immunofluorescence (IF) images confirm the expression of neuronal markers, including MAP2, Tau, β -3 tubulin, and BRN-2, highlighting the progressive acquisition of neuronal identity.



Doxycycline-induced NGN2 expression in AAVS-T0-NGN2 KOLF2.1J cells results in the gradual loss of iPSC markers.
 IF images demonstrate the progressive downregulation of Tra1-60-R, NANOG, OCT-4, and SOX-2 during differentiation.

Images were kindly provided by Nélío Oliveira (The Jackson Laboratory for Genomic Medicine, Farmington, CT, USA), Marianita Santiana and Erika Lara Flores (Center for Alzheimer's and Related Dementias (CARD), National Institute on Aging and National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, MD, USA).