

**The NMDA receptor GluN2C subunit controls cortical excitatory-inhibitory balance,
neuronal oscillations and cognitive function**

Subhash C. Gupta¹, Aparna Ravikrishnan¹, Jinxu Liu¹, Zhihao Mao², Ratnamala Pavuluri¹,
Brandon G. Hillman¹, Pauravi Gandhi¹, Dustin J. Stairs³, Ming Li⁴, Rajesh R. Ugale⁵, Daniel T.
Monaghan² and Shashank M. Dravid^{1*}

Author affiliations:

¹Department of Pharmacology, ³Psychology, Creighton University, Omaha, NE 68178 USA,

²Department of Pharmacology and Experimental Therapeutics, University of Nebraska-Medical
Center, Omaha, NE, ⁴Department of Psychology, University of Nebraska-Lincoln, Lincoln, NE
68588 USA, ⁵Department of Pharmaceutical Sciences, R.T.M. Nagpur University, Nagpur,
Maharashtra 440033 India.

Running title: GluN2C subunit regulates cortical E/I balance

Manuscript: Figures: 6; Tables: 1

Word count: Abstract: 242, Introduction: 497, Discussion: 1193.

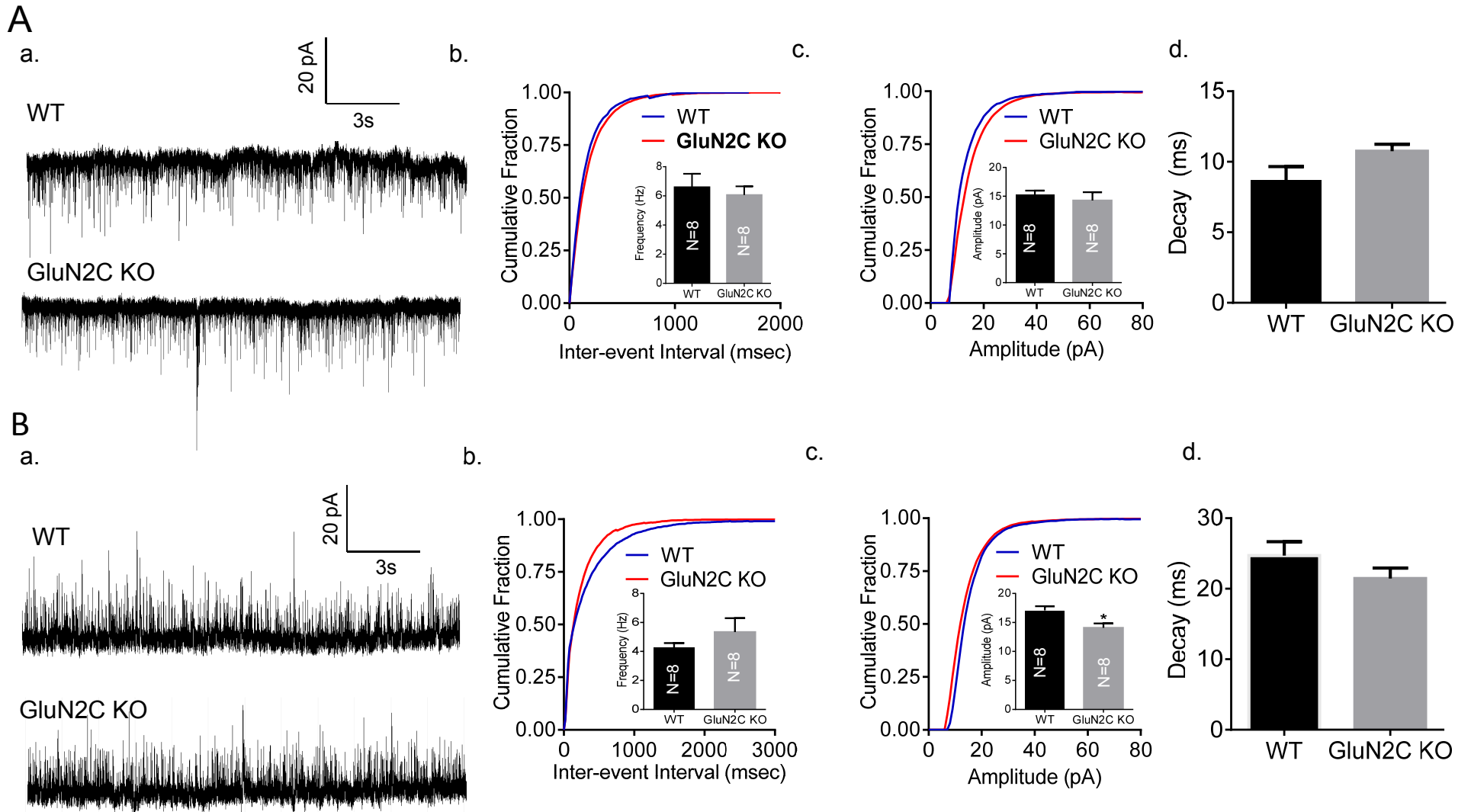
Corresponding Author: Dr. Shashank M. Dravid, Department of Pharmacology, Creighton
University, School of Medicine, 2500 California Plaza, Omaha, NE 68178, Phone: 402-280-
1885, Fax: 402-280-2142, Email: ShashankDravid@creighton.edu

Supplementary figure 1. (A) No significant differences in the frequency, amplitude or decay of sEPSCs of layer V pyramidal neurons in the mPFC were observed between WT and GluN2C KO mice (n=8 cells from 3-4 mice/genotype). Representative traces (a), cumulative probability plots for inter-event interval and amplitude and histograms for frequency and amplitude in inset (b, c) and histograms for decay (d). (B) No significant differences in the frequency or decay and a modest reduction in amplitude of sIPSCs of layer V pyramidal neurons in the mPFC in GluN2C KO mice (n=8 cells from 3-4 mice/genotype). Representative traces (a), Cumulative fraction plots for inter-event intervals and amplitude and histograms for frequency and amplitude in the inset (b, c) and decay kinetics (c, f). Only amplitude was modestly but significantly lower in GluN2C KO sIPSC (unpaired t-test with Welch's correction, *P = 0.0110).

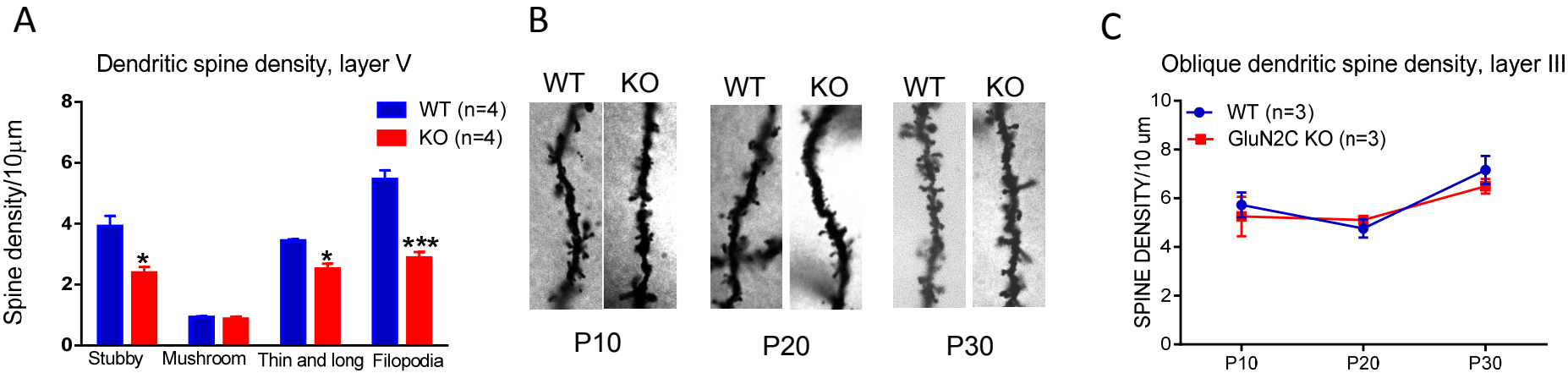
Supplementary figure 2: Changes in dendritic spine density in different layers, and for different spine types in the mPFC of GluN2C KO mice. (A) Dendritic spine density was significantly reduced in layer V of the mPFC in GluN2C KO mice; this reduction was observed for all spine types (*P = 0.01295, stubby and P = 0.0101 thin and long spines and ***P = 0.0007 for filopodia-like spines, unpaired t-test with Welch's correction, n = 4/genotype), except for mushroom spines. (B, C) No significant difference in the oblique apical dendritic spine density was observed between WT and GluN2C KO mice at P10, P20 or P30. (B) Representative images of Golgi-stained sections and (C) quantitative results.

Supplementary figure 3: No change in somatostatin labeling in GluN2C KO. The number of somatostatin-positive cells in the mPFC of GluN2C KO was not different from wildtype.

Supplementary figure 1



Supplementary figure 2



Supplementary figure 3

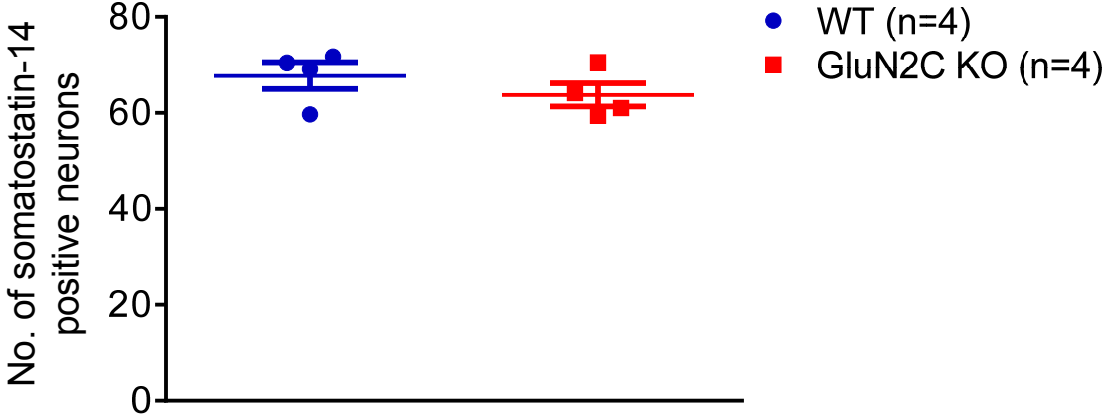
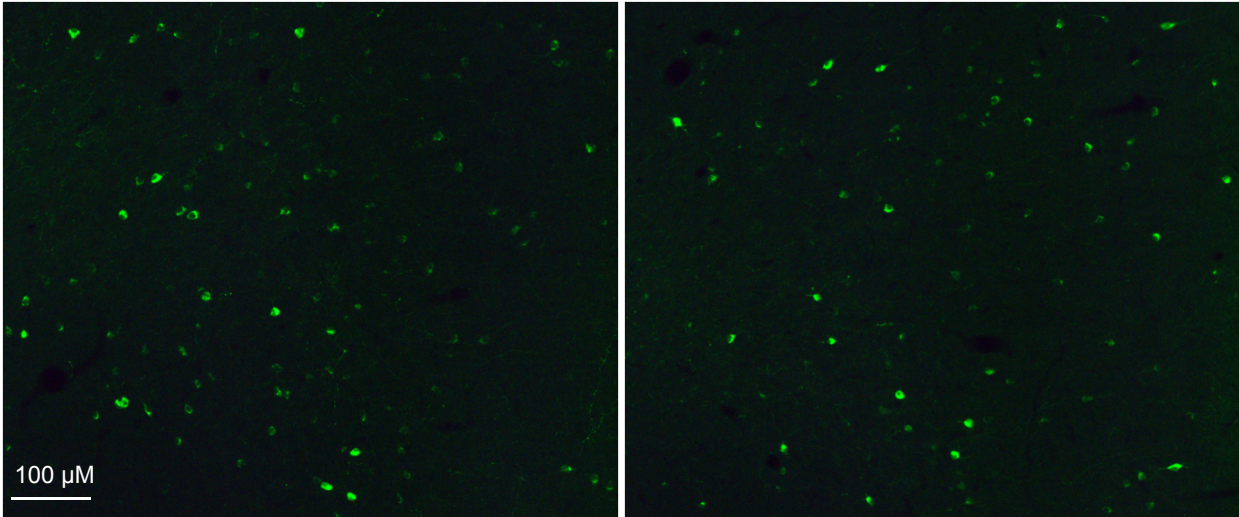


Figure 2A

Dendritic spine data in mPFC of wild type mice

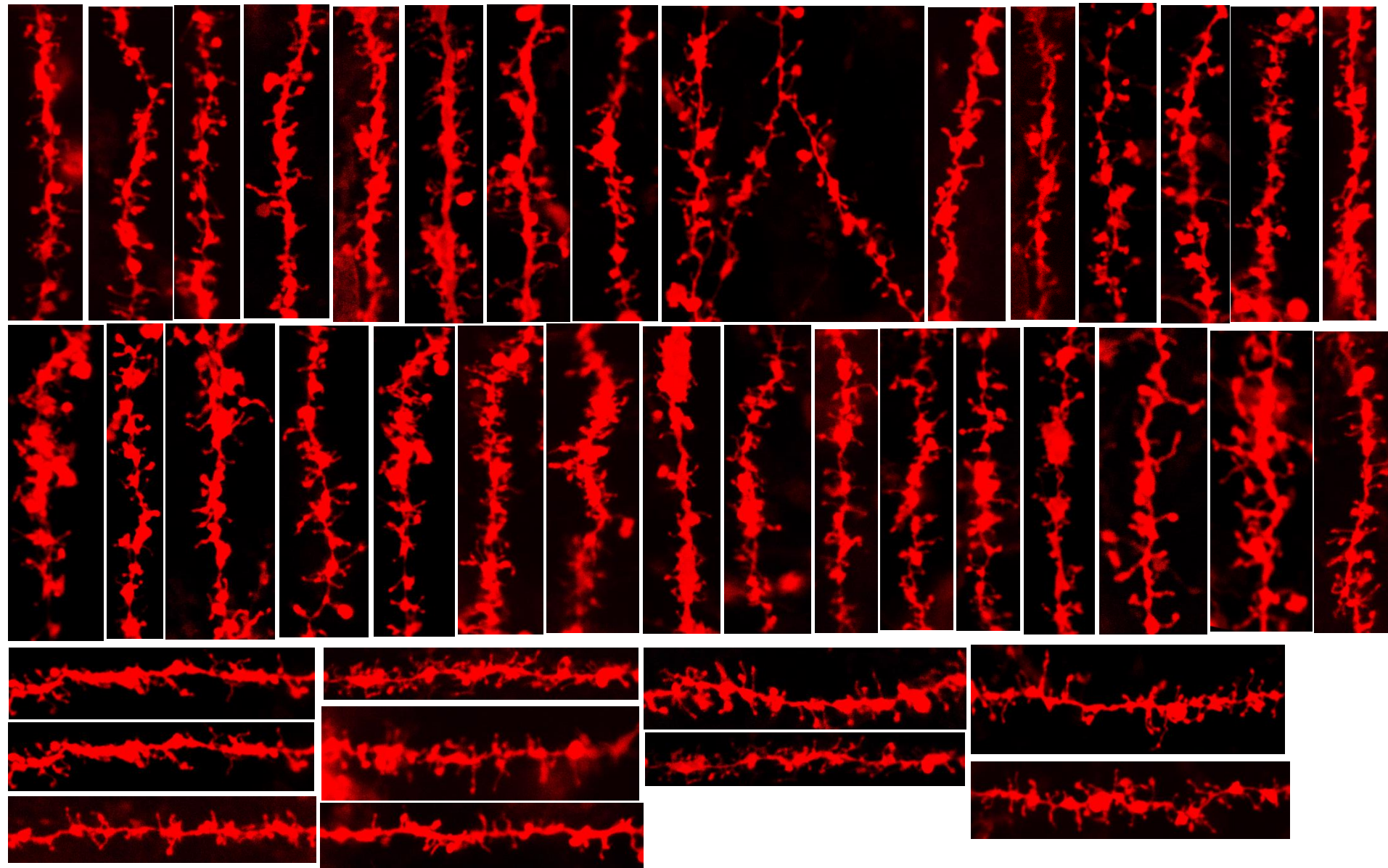


Figure 2A

Dendritic spine in GluN2C KO mice

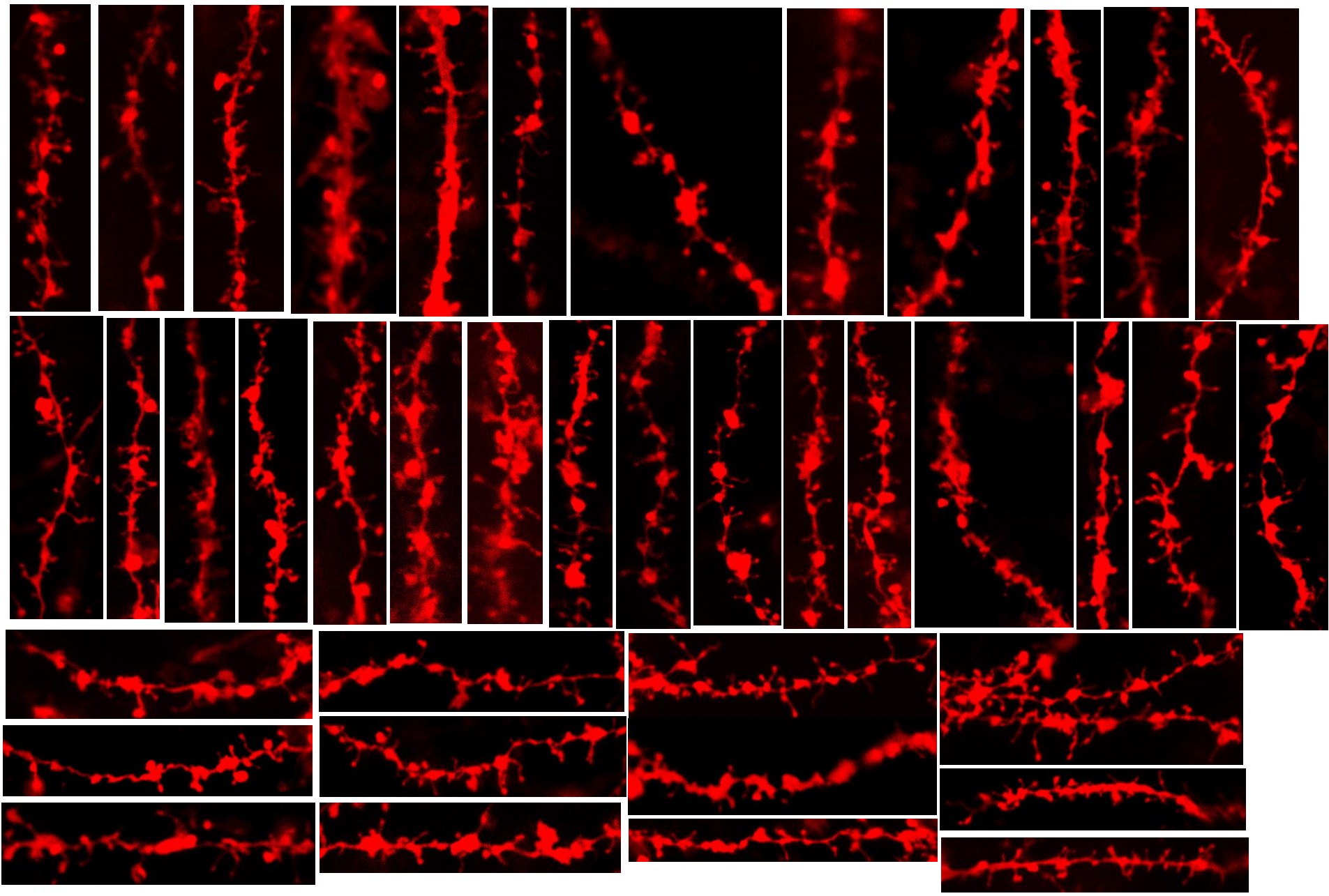


Figure 3A

WT (vGluT1-VGAT)

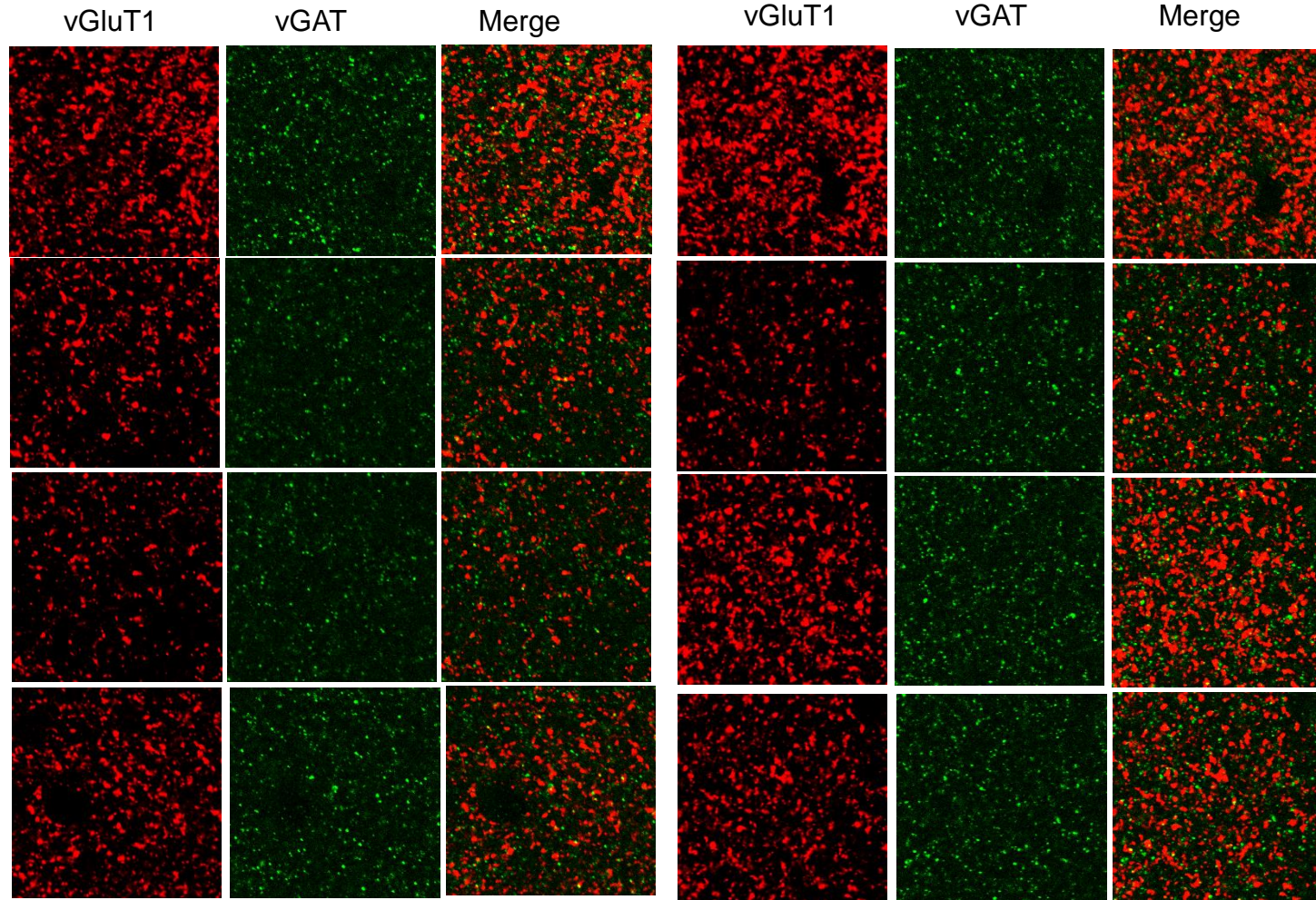


Figure 3A

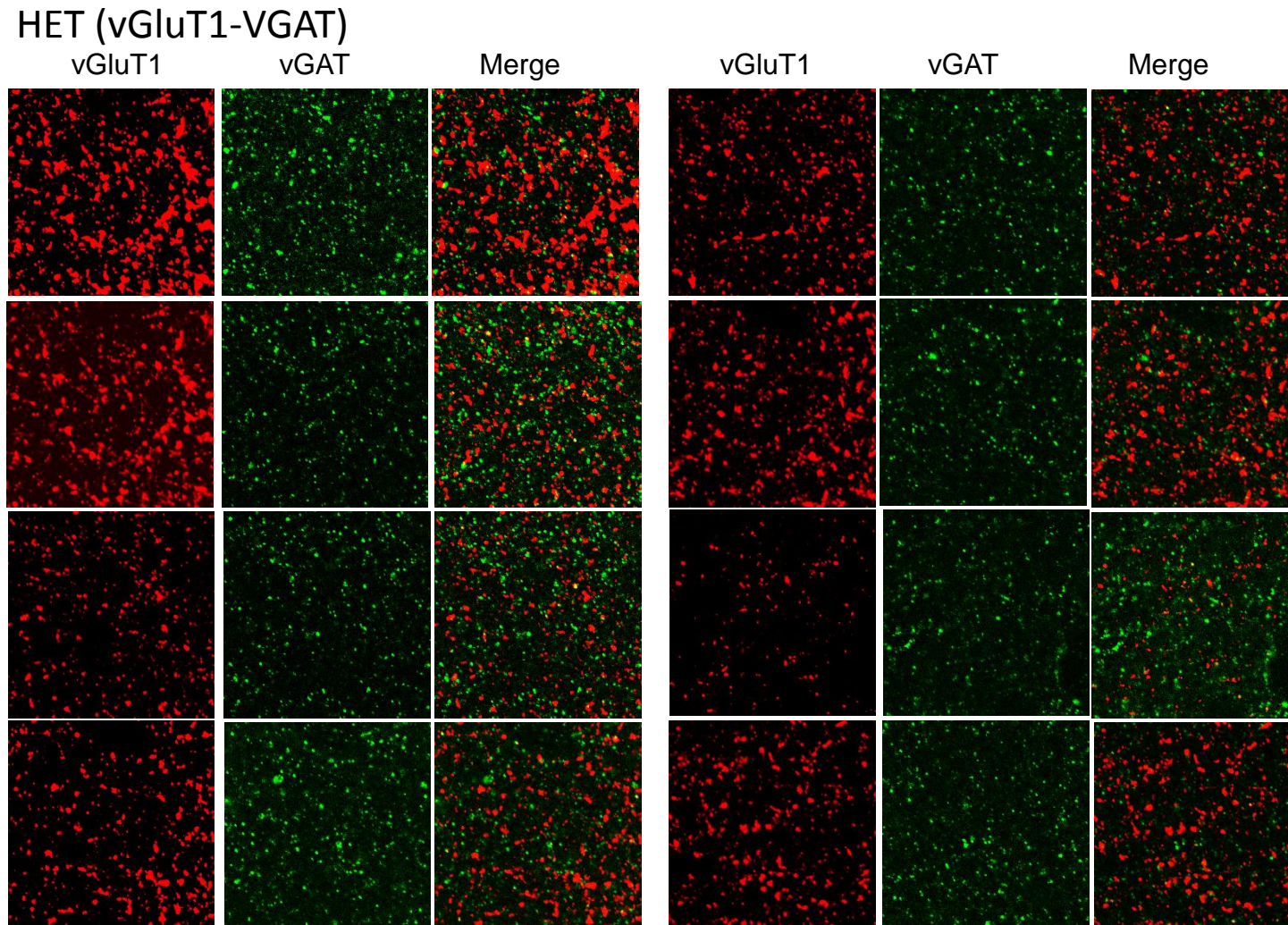


Figure 3A

GluN2C KO (vGluT1-VGAT)

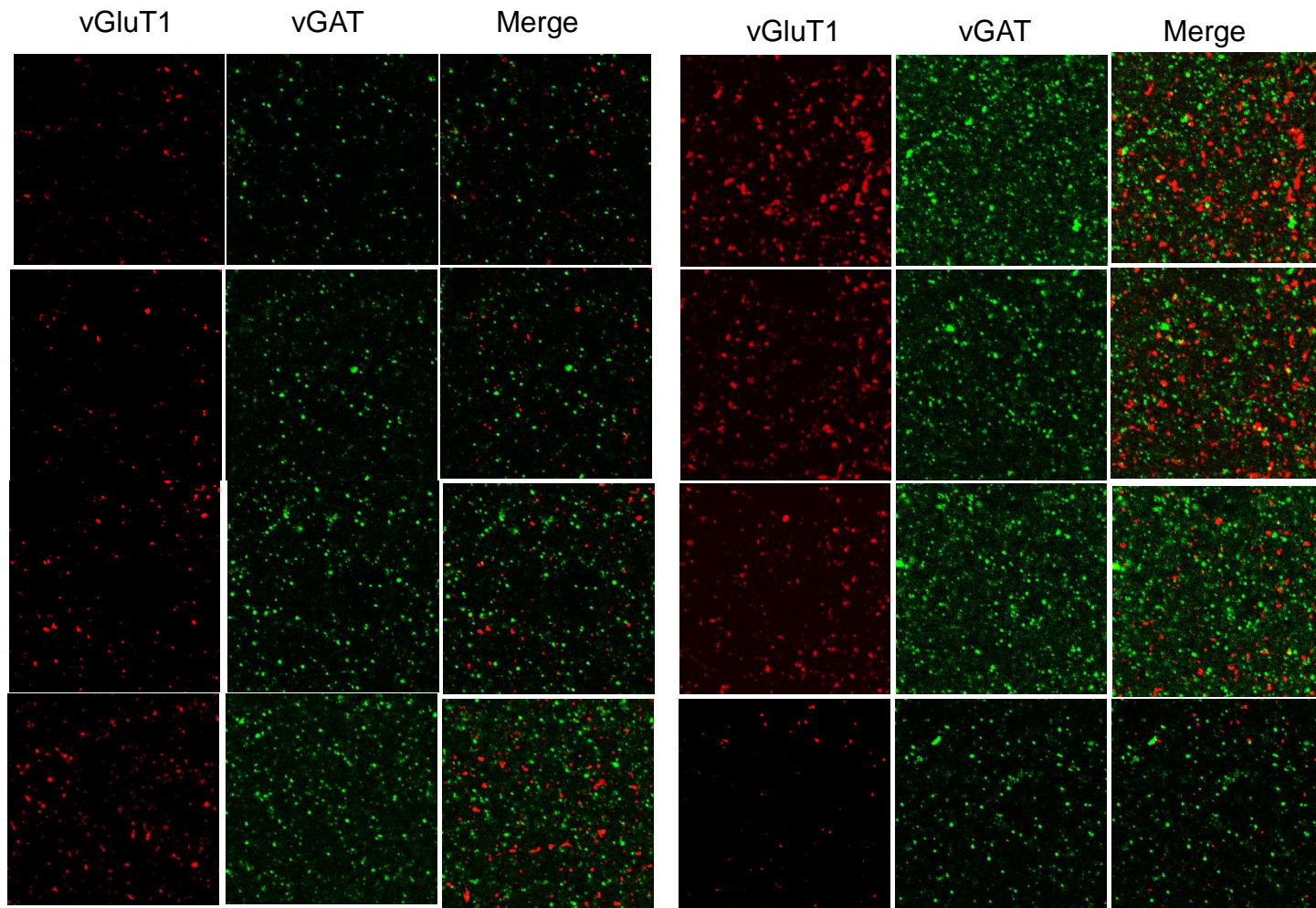


Figure 3C

PV-WT

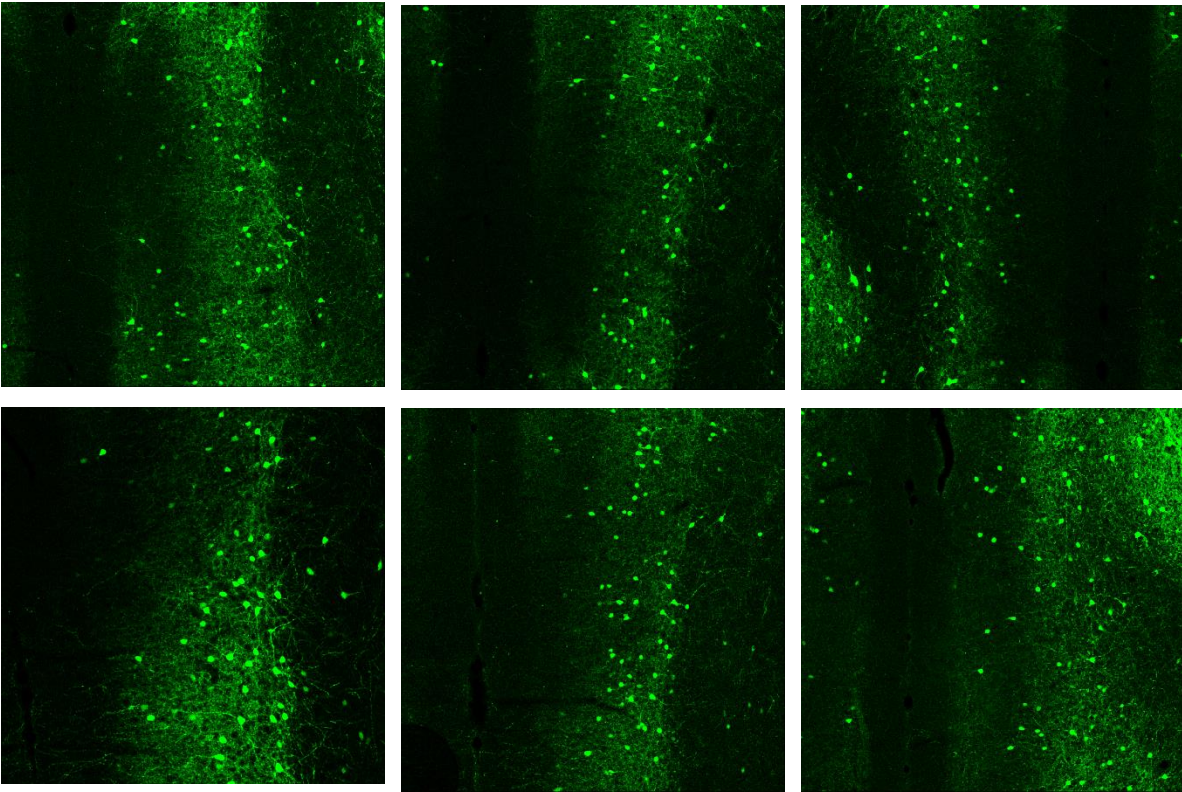


Figure 3C

PV-HET

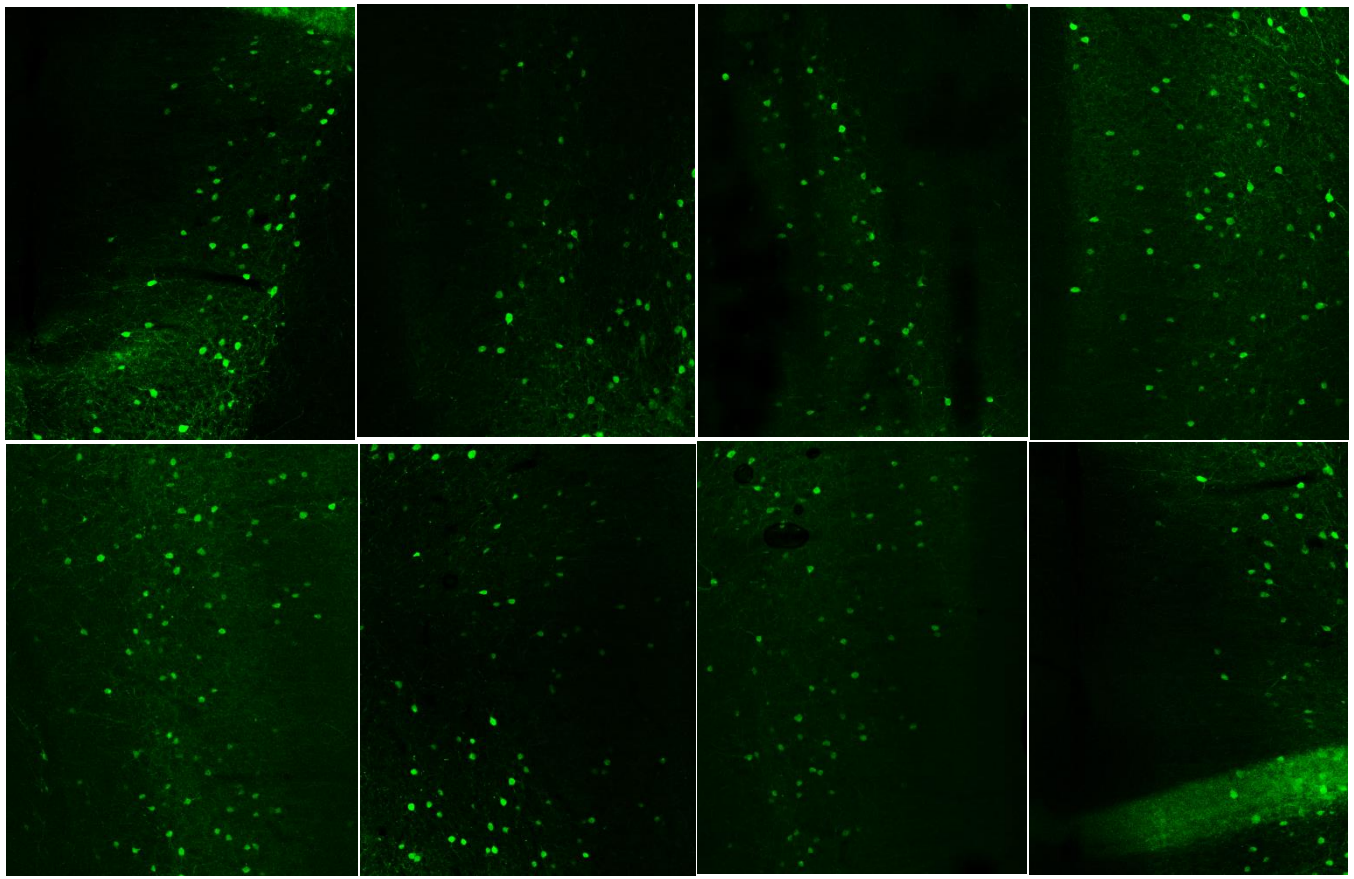


Figure 3C

PV-GluN2C KO

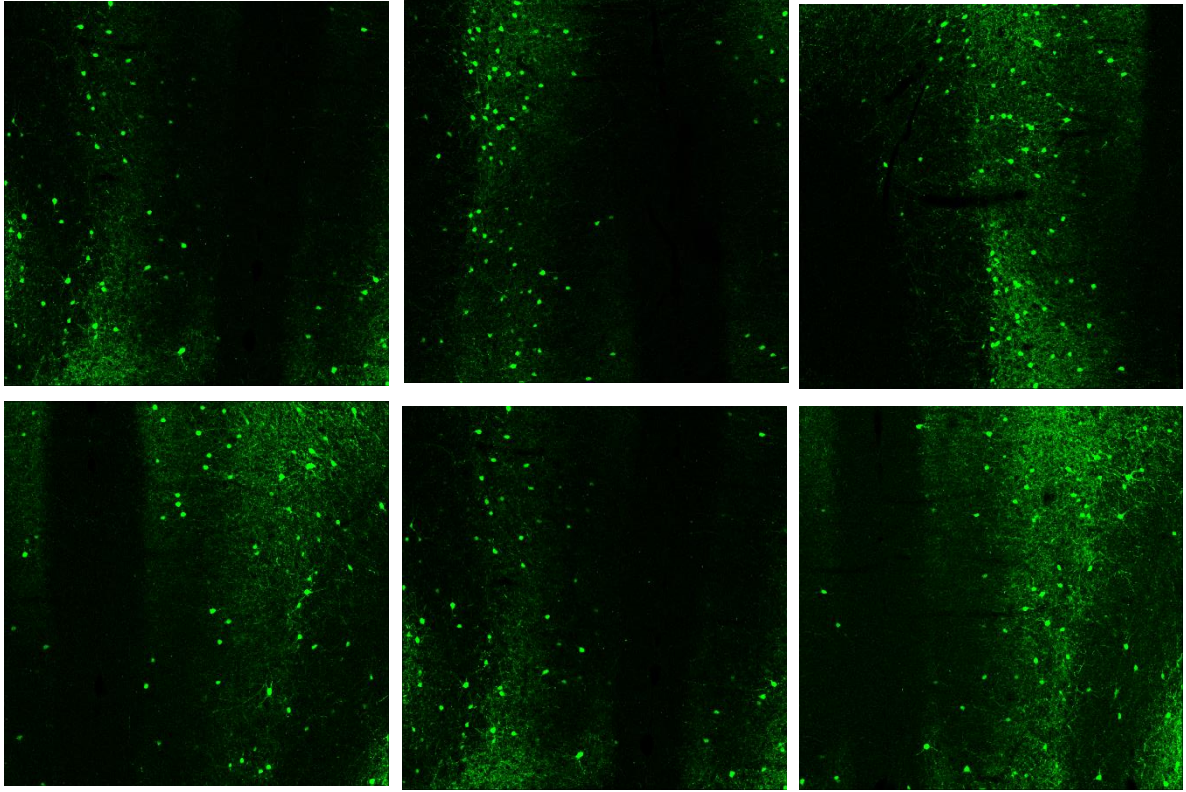


Figure 3E

WT-NeuN and GAD67 staining

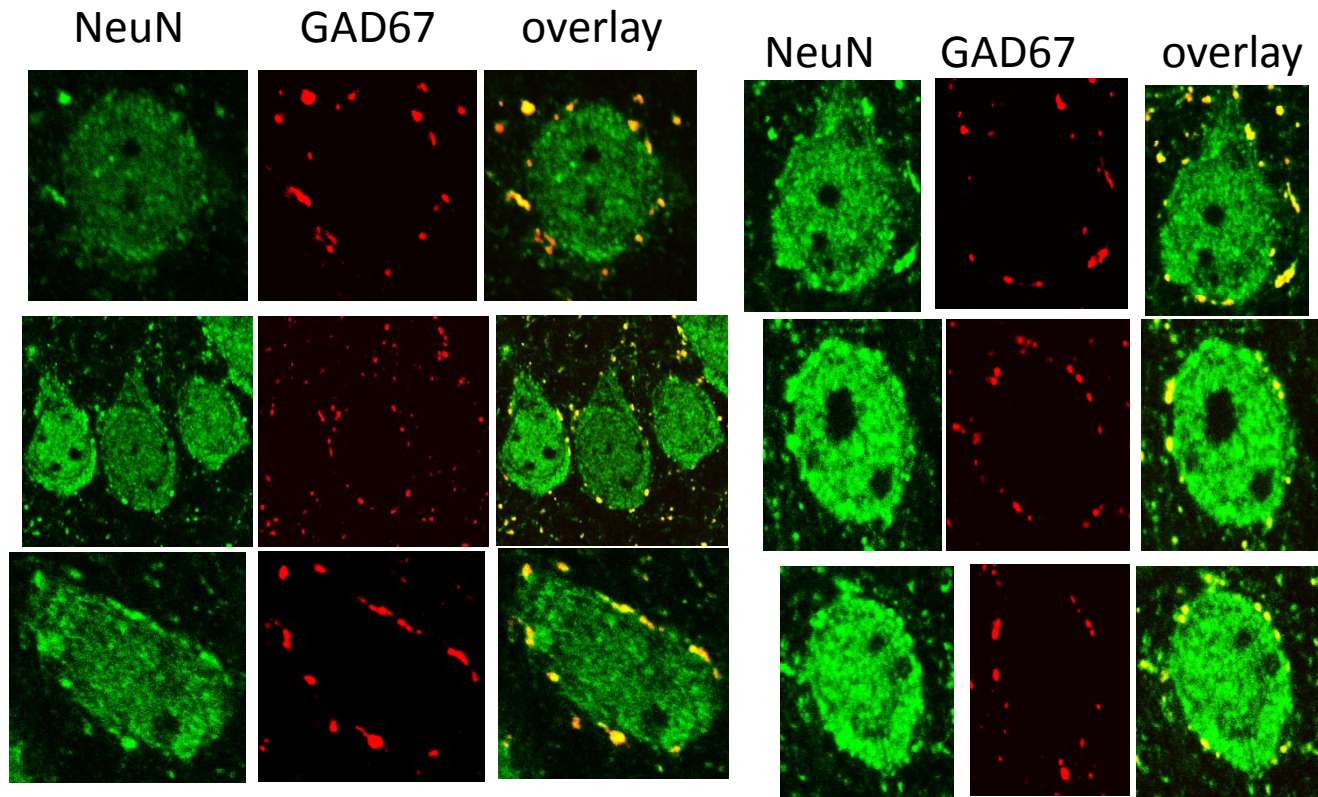


Figure 3E

GluN2C HET-NeuN and GA67 staining

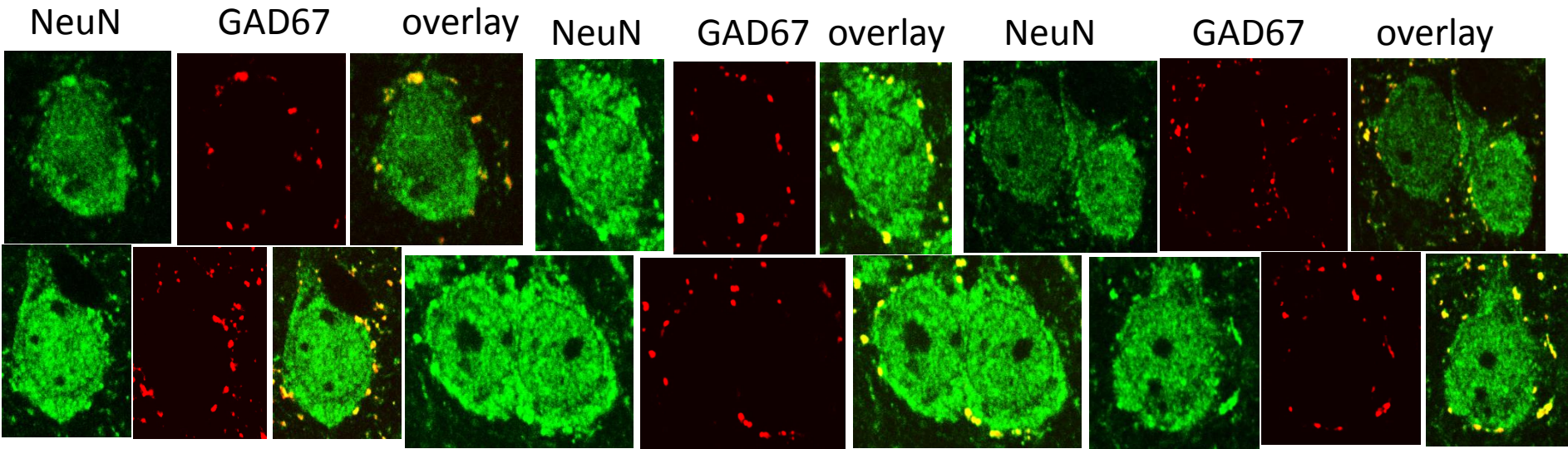


Figure 3E

GluN2C KO- NeuN and GA67 staining

