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We use this protocol and it's working

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Isolation of NeuN+ cells from brain tissue (for CUT and RUN)

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ABSTRACT

This protocol describes the steps to isolate NeuN+ cells from brain tissue in preparation for CUT and RUN

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Isolation of NeuN+ cells

45m

1 Nuclei were isolated from frozen tissue as described here.

2 Before FACSing, nuclei were incubated with Recombinant Alexa Fluor® 488 Anti-NeuN antibody [EPR1276 - Neuronal Marker (Abcam, Cat# ab190195, RRID:AB_2716282) at a concentration of 1:500 for 00:30:00 On ice as according to Spalding, K.L. et al. (2013). 30m

3 The nuclei were run through the FACS at 4 °C with a low flow rate using a 100 mm nozzle and 300.000 nuclei Alexa Fluor – 488 positive nuclei were sorted.

4 The sorted nuclei were pelleted at 1,300 x g for 00:15:00 and resuspended in 1 mL of ice-cold nuclei 15m wash buffer (20 mM HEPES, 150 mM NaCl, 0.5 mM spermidine, 1x cOmplete protease inhibitors, 0.1% BSA) and 10 µL per antibody treatment of ConA-coated magnetic beads (Epicypher) added with gentle vortexing (Pipette tips for transferring nuclei were pre-coated with 1% BSA), to perform CUT&RUN.