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Protocol status: Working
We use this protocol and it's working

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Immunofluorescent Staining of Fixed Mouse Brain Tissue Sections

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ABSTRACT

This protocol describes steps for immunofluorescent staining of free floating fixed mouse brain tissue sections.

MATERIALS

- 1X PBS (UCSF Stem Cell Core)
- Triton X-100 (Sigma Aldrich 93443)
- Fetal bovine serum (UCSF Stem Cell Core)
- Primary antibody
- Secondary antibody
- Blocking buffer: 0.2% Triton, 4% FBS in 1X PBS
- Glass microscope slides (Fisher 22-038-103)
- Anti-fade mounting medium with or without DAPI (Vector Laboratories H140010)

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1 Free floating sections in 12-well plates, all steps performed at room temperature.

1.1 2 x 10min PBS washes.

1.2 2 x 10min 0.2% PBS-Triton (PBS-T) washes.

1.3 Block 1hr in blocking buffer.

1.4 Incubate in primary antibody diluted in blocking buffer overnight.

1.5 3 x 10min 0.2% PBS-T washes.

1.6 Incubate in secondary antibody diluted in blocking buffer for 2hrs.

1.7 2 x 10min PBS-T washes.

1.8 2 x 10min PBS washes.

1.9 Mount sections on glass slides, add mounting media and coverslip.