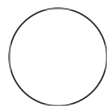




APR 07, 2023

Immunofluorescent staining for neuronal marker MAP2

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ABSTRACT

This is the protocol for immunofluorescent staining for neuronal marker MAP2.

OPEN ACCESS

DOI:

dx.doi.org/10.17504/protocols.io.j8nlkwj46l5r/v1

Protocol Citation: Qing Wang 2023.

Immunofluorescent staining for neuronal marker MAP2.

protocols.io

<https://dx.doi.org/10.17504/protocols.io.j8nlkwj46l5r/v1>

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Protocol status: Working

We use this protocol and it's working

Created: Apr 07, 2023

Last Modified: Apr 07, 2023

PROTOCOL integer ID:

80149

Keywords: ASAPCRN

1. Treat differentiated SH-SY5Y cells with 40 ug/mL eumelanin or pheomelanin or PBS for 24 hours.

- 2** 2. Process cells using Cytofix/Cytoperm™ fixation/permeabilization solution (BD554714, Thermo Fisher Scientific) and block with 5% normal goat serum.
- 3** 3. Add primary antibody against microtubule-associated protein 2 (MAP2)(30 µg/mL, OSM00036G, Thermo Fisher Scientific) and incubate at 4°C overnight.
- 4** 4. Add secondary antibody (1:1000, Alexa 594-conjugated, A11012, Thermo Fisher Scientific) and incubate for 2 hours at room temperature.
- 5** 5. Stain nuclei with DAPI.
- 6** 6. For MAP2-positive cell counting, three images are captured randomly from each well using FluoView FV300 confocal microscope under a 60x objective lens. MAP2 and DAPI channels are merged, and MAP2-positive cells in each random visual field of 0.045000 mm² are counted using ImageJ.