

Visualization of neuronal dendrites in human-derived cerebral organoids for use in studies of neurotoxicity

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Human-derived cerebral organoids represent an exciting new method for studies of neurotoxicity, as they hold the potential to both replace animal models and better recapitulate the human condition [1,2]. Herein, organoids were cultured as according to Pamies et al. [3]. After 3 weeks, they were fixed with 4% paraformaldehyde. Immunocytochemistry was performed to visualize mature neuronal bodies (NeuN, magenta) and dendrites (MAP2, green). Organoids were then imaged on a Zeiss LSM 800 confocal laser scanning microscope with 20x magnification. Image processing was accomplished using Fiji ImageJ.

Materials

Primary antibodies

- Mouse anti-NeuN (PhosphoSolutions, 583-FOX3) ([1:1000])
- Chicken anti-MAP2 (Aves, MAP2) ([1:1000])

Secondary antibodies

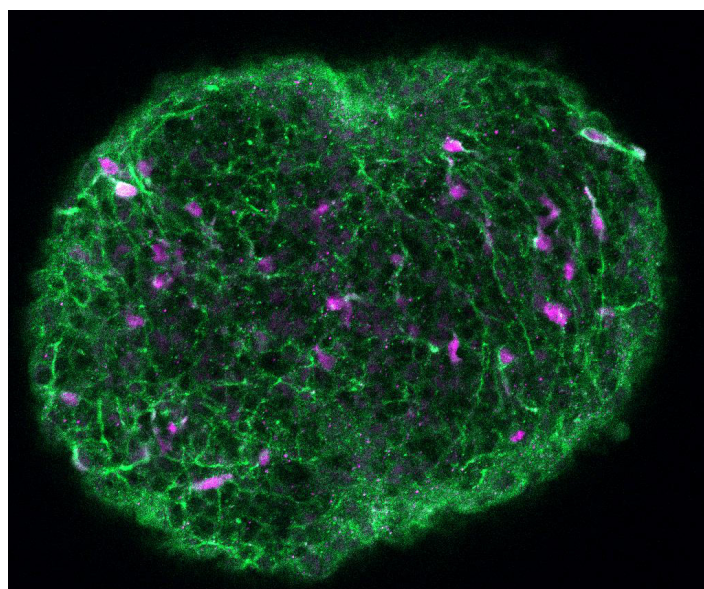
- Alexa Fluor 647 AffiniPure donkey anti-mouse IgG (Jackson ImmunoResearch, 715-605-151) ([1:400])
- Alexa Fluor 488 AffiniPure donkey anti-chicken IgY (IgG) (Jackson ImmunoResearch, 703-545-155) ([1:400])

Mounting media

- ProLong Diamond anti-fade mountant (ThermoFisher, P36961)

Imaging dish

- Micro-insert 4 well in μ -Dish 35 mm, high (ibidi, 80406)



References

1. Hartung T. Food for thought look back in anger – what clinical studies tell us about preclinical work. *ALTEX*. 2013; 30: 275-291.
2. Johnson SC, Pan A, Sun GX, Freed A, Stokes JC, et al. Relevance of experimental paradigms of anesthesia induced neurotoxicity in the mouse. *PLoS One*. 2019; 13: e0213543.
3. Pamies D, Block K, Lau P, Gribaldo L, Pardo-Villamizar CA, et al. Rotenone exerts developmental neurotoxicity in a human brain spheroid model. *Toxicol Appl Pharmacol*. 2018; 354: 101-114.

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