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Optical Imaging-Guided Cell Transplantation for Improved Cell Loading Efficacy

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Background

Optical imaging-guided transplantation of mouse cerebral cortex cells combines the advantages of optical imaging and precise neurointerventions. In neuroscience research, optimizing delivery procedures in the mouse cerebral cortex is critical for the accurate and efficient manipulation of neural circuits.

Objective

We hypothesized that using the pulse-elevation mode of cell delivery can significantly improve cell loading efficiency in the needle track of the mouse brain.

Methods

Mouse neural stem cells (C17.2 cells) stably expressing the fluorescent protein Venus through transduction with lentiviral vector FCIV were injected at the concentration of 2.5×106 cells/ml to 0.6 % low-melting agarose [1] or the mouse brain at the coordinates: AP -3.35 mm, ML 1.75 mm using the stereotaxic instrument. The target depth for cell transplantation was set at 500 µm. Before injection, the needle was extended to 700 µm and then withdrawn to a target depth of 500 µm. Infusion program settings: step1: rate: 0.1 µL/min, 20 nL; delay 20 s and withdraw to 0.4 µm; step2: rate: 0.1 µL /min, 20 nL; delay 20 s and withdraw needle to 0.3 µm; step3: rate: 0.1 µL /min, 20 nL; delay 20 s and withdraw needle to 0.2 µm. Leica DMi8 was used to capture fluorescent images in vitro. For the in vivo experiment, 2-photon imaging was applied on days 0, 1, and 2 [2].

Results

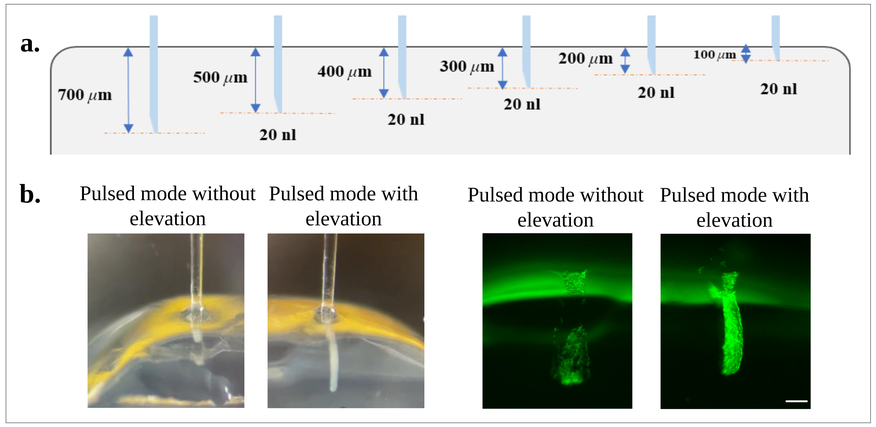

Figure 1 (a) Design of in vitro experiments; (b) images of needle tracks under a Leica microscope. Scale bar in (b), 100 µm.

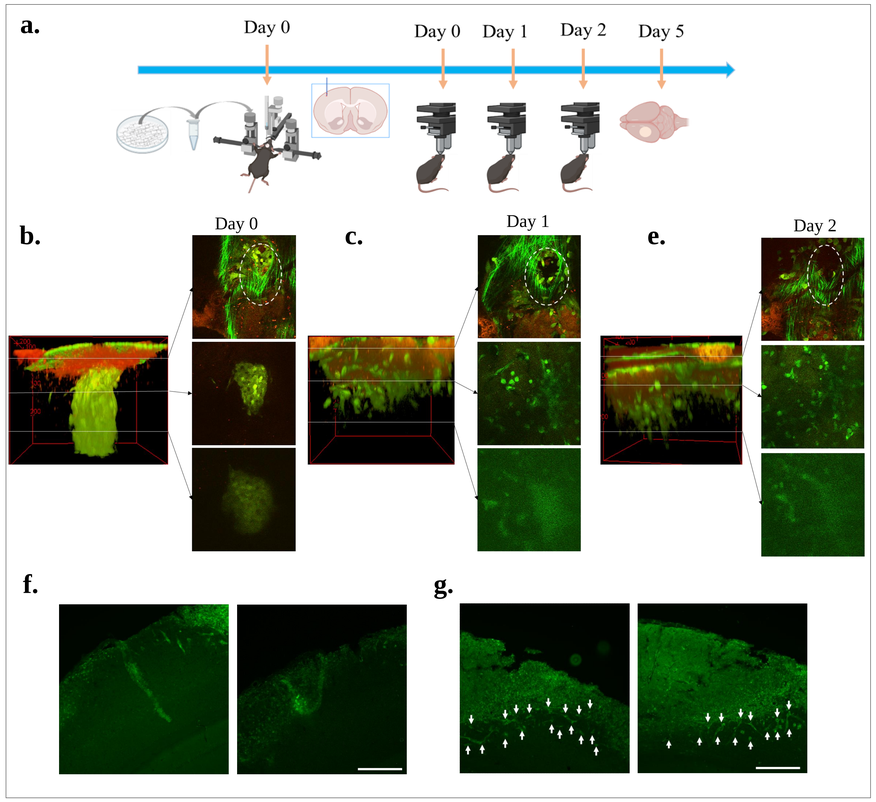

Figure 2 (a) Design of in vivo experiments; (b) 3D view of the imaging stack showing transplanted cells in needle track on day 0 (c), 1 (d), and 2 (e); (f,g) Needle tracks and transplanted cell in brain slices. Scale bar in (f, g), 332 µm.

In the in vitro experiment (Figure1a), we found a significant difference between the pulse-elevation and pulse groups. The cell signal in the pulse-elevation group needle track was significantly higher than in the pulsed group (Figure 1b). In vivo(Figure 2a, b), using the same infusion procedure, we observed a clear needle track post-surgery on day 0, and cells were confined to the needle track (Figure 2b). On day 1 and day 2, we imaged the same brain area but found scattered cells around the injected area, implying active migration of C17.2 cells out of the injection site (Figure 2c and 2e). The ability to observe clear needle tracks on the brain slices and transplanted cells provided additional evidence of the success and accuracy of the cell transplantation process(Figure 2f and 2g). These observations suggest that the pulsed-elevation injection can precisely deliver and hold cells within the needle track and improve loading efficiency to the mouse brain.

Conclusions

We established a platform for optical imaging-guided cell transplantation into the mouse cerebral cortex. Through in vitro and in vivo experiments, we found that the pulse-elevation injection method significantly increases cell transplantation's efficacy and improves cell loading efficiency. Further evaluation of this preliminary findings is warranted

References

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2. Liang Y, Walczak P. *Long term intravital single cell tracking under multiphoton microscopy*. J Neurosci Methods. 2021 Feb 1;349: 109042. doi: 10.1016/j.jneumeth.2020.109042. Epub 2020 Dec 16. PMID: 33340557.