

JAN 24, 2024

OPEN BACCESS



DOI:

dx.doi.org/10.17504/protocols.io. 5jyl8pxz6g2w/v1

Protocol Citation: Mai-Anh Vu, mwhowe 2024. Targeted optogenetic stimulations. **protocols.io**

https://dx.doi.org/10.17504/protoc ols.io.5jyl8pxz6g2w/v1

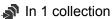
License: This is an open access protocol distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working

Created: Jan 09, 2024

Last Modified: Jan 24, 2024

Targeted optogenetic stimulations



Mai-Anh Vu¹, mwhowe¹

¹Department of Psychological and Brain Sciences, Boston University, Boston, MA, United States

Team Cragg



Mai-Anh Vu

ABSTRACT

We have developed a new micro-fiber array approach capable of chronically measuring and optogenetically manipulating local dynamics across over 100 targeted locations simultaneously in head-fixed and freely moving mice, enabling investigation of cell-type and neurotransmitter-specific signals over arbitrary 3-D volumes . This protocol includes the steps for targeted optogenetic manipulation. Please contact us (mwhowe@bu.edu) if you are interested in using this technique.

Oct 24 2024



PROTOCOL integer ID: 93204

Keywords: ASAPCRN

Funders Acknowledgement:

Aligning Science Across Parkinson's (ASAP) Grant ID: ASAP-020370

Targeted optogenetic stimulations

- To couple light into individual optical fibers in our array for targeted optogenetic manipulations, simultaneously with imaging, we integrated a programmable digital mirror device (DMD, Mightex Polygon1000 Pattern Illuminator DSI-K3-L20) into the light path of our imaging microscope (see Multi-color fiber array imaging protocol) as detailed via the substeps below:
 - **1.1** Excitation light was provided by a 3.2W, 465nm laser (Mightex, LSR-040-0465), which was coupled to the DMD with an optical fiber.
 - 1.2 Light from the DMD was coupled into the objective path by a dichroic (Chroma 570lpxr).
 - 1.3 570nm excitation and emission filters (see Multi-color fiber array imaging protocol) enabled simultaneous imaging of red fluorophores during stimulation.
- 2 Control of light patterning and stimulation parameters was achieved with PolyScan2 control software (Mightex) and custom MATLAB functions.
- A calibration step was performed prior to stimulations to align the camera view with the PolyScan2 software, allowing us to design patterns of circular light (~40µm diameter for each spot) to target individual fibers.

- 4 Transmission efficiency through 50µm diameter fibers in our arrays was calculated at approximately 39% based on comparisons between power at the objective and transmitted light through individual fibers.
 - 4.1 For stimulations of dopamine release, a light spot of ~750 µW (measured at the objective) was used, resulting in an estimated power density of 175 mW/mm2 (0.29 mW total power) at each implanted fiber tip.
 - 4.2 For stimulations of D1 expressing neurons, a light spot of ~600 µW at the objective was used, resulting in an estimated power density of 141 mW/mm2 (0.23 mW total power) at the fiber tip.
- 5 We estimated relative excitation light intensity and excitation area as a function of distance from the fiber tip by applying a light scattering model developed by Yona et. al 2016 with a scattering coefficient of 140 cm-1 approximated for the striatum based on Azimipour et al 2014 and Al-Juboori et al. 2013 and an activation threshold for ChR2 of 1 mW/mm² 70,71.
- 6 Light pulse trains for stimulation (30Hz, 4ms pulse width, 1s or 5/10s durations for DA and D1 neuron stimulation respectively) were programmed in PolyScan2 and triggered with 5V digital pulses controlled via MATLAB and sent from a NIDAQ board (National Instruments PCIe 6343) to the Polygon. Stimulations were triggered randomly (inter-stimulation interval 30-60 seconds) for 5-10 minutes per session for all stimulation experiments.