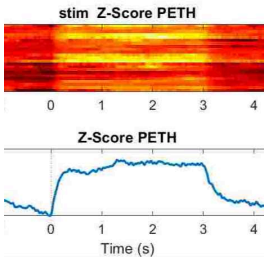
Aug 26, 2024

Fiber Photometry on Noradrenergic/Dopaminergic Neurons (Open Field)



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

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Disclaimer

The **protocols.io** team notes that research involving animals and humans must be conducted according to internationally-accepted standards and should always have prior approval from an Institutional Ethics Committee or Board.

Abstract

The goal is to monitor in vivo calcium activity in noradrenergic and dopaminergic neurons within the hindbrain and midbrain of mice using fiber photometry during an open-field setup.

Materials

- Doric fiber photometry system
- Custom patch cord for fiber connection
- Fiber optic cable (suitable for the wavelengths used)
- Mice with implanted optical fiber targeting dopaminergic neurons in the midbrain or LC neurons in the hindbrain.
- Open-field arena (with appropriate dimensions)
- Power meter
- Animal handling equipment (e.g., cages, transfer box)
- Behavioral tracking system (optional)

Troubleshooting

Pre-Experiment Preparation:

1 **Animal Preparation:**

1.1 Ensure that the mice have undergone appropriate surgical procedures for fiber implantation and calcium indicator expression in the relevant brain region targeting catecholaminergic neurons. Allow for recovery and acclimatization.

2 **Equipment Setup:**

2.1 Start up the Doric fiber photometry system and ensure that the light source is connected to the appropriate fiber optic cable and that the photodetector is ready.

Protocol Steps:

3 **Equipment Initialization:**

3.1 Turn on the System: Power on the Doric fiber photometry system.

3.2 **Loading Configuration:**

3.3 Open the data acquisition Doric software and load the appropriate experimental settings/configuration.

4 **Measuring Power:**

4.1 Measure the optical power at the tip of the fiber using the power meter.

4.2 Adjust the power output if necessary to ensure optimal excitation for the calcium indicator.

5 **Bleaching the Fiber:**

5.1 Immediately after verifying the power, bleach the fiber for a duration of 10 minutes at the desired excitation wavelength.

5.2 Ensure that the bleaching is uniform and that thermal management is maintained.

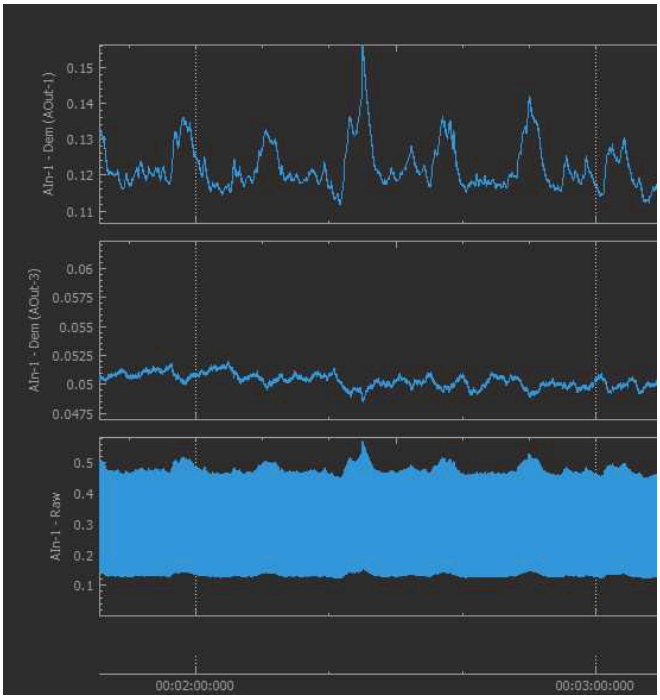
6 **Connecting the Mouse:**

6.1 Gently restrain the mouse and connect the custom patch cord to the implanted fiber and secure it appropriately. Note: Proper handling and acclimatization of the mouse prior to the experiment are essential to minimize stress and ensure reliable data collection. To mitigate stress, it is important to allow the mouse to become familiar with the experimental setting and equipment well ahead of the actual experiment. This can be achieved by gently handling the mouse for short periods over several days, providing a calm and quiet environment, and allowing the mouse to explore the open field arena before the actual recording. Also, proper handling techniques must be employed to minimize stress while connecting the patch cord to the mouse, as excessive restraint can lead to anxiety and impact the animal's physiological responses.

7 **Recording:**

7.1 Start the acquisition in the data acquisition software.

7.2 Monitor baseline calcium activity for an initial period (e.g., 1-2 minutes) before introducing any stimuli or behavioral tasks.



Upper panel: GCaMP signal.
Middle Panel: Isosbestic signal
Lower panel: Raw signal

7.3 Conduct open-field experiments, allowing the mouse to explore freely for 5-30 minutes while recording calcium activity continuously.

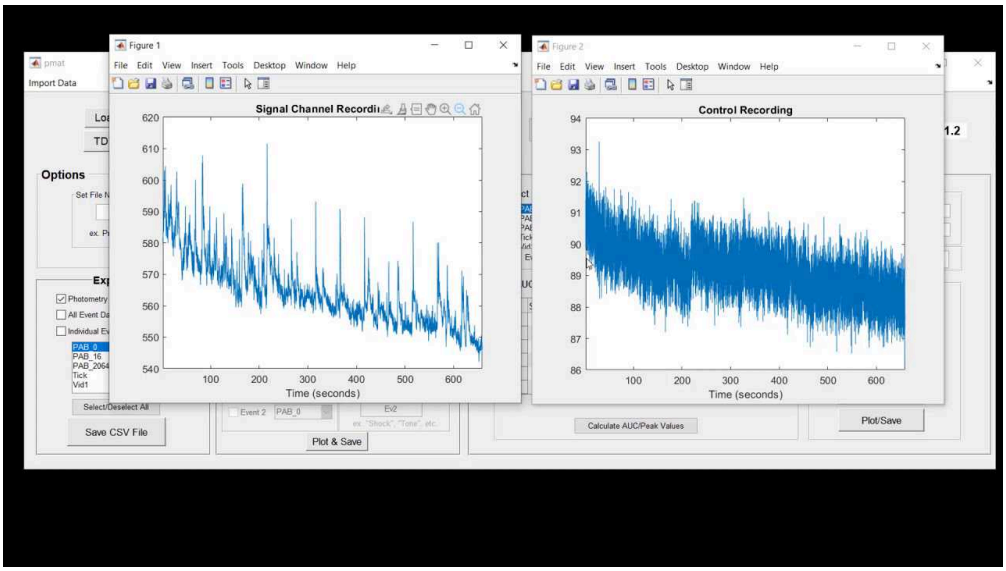
8 **Post-Experiment Procedures:**

8.1 After completion of the recording session, disconnect the mouse from the patch cord carefully.

8.2 Return the mouse to its home cage for recovery or monitoring.

8.3 Save all data collected during the experiment for subsequent analysis.

9 Data Analysis: Analyse the collected data using pMAT or any other compatible software. pMAT is an open source Matlab-based code.



https://github.com/djamesbarker/pMAT

Notes:

10 It may be helpful to include video recordings to correlate behavioral data with calcium imaging results.

11 This protocol outlines a general approach; adjustments may be necessary based on specific experimental conditions or equipment configurations.