Coupling the Timestamp of Licking Data with Fiber Photometry Calcium Imaging in Mice

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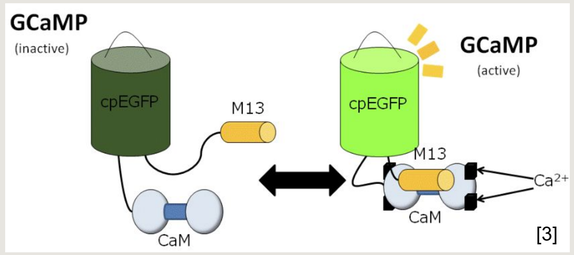
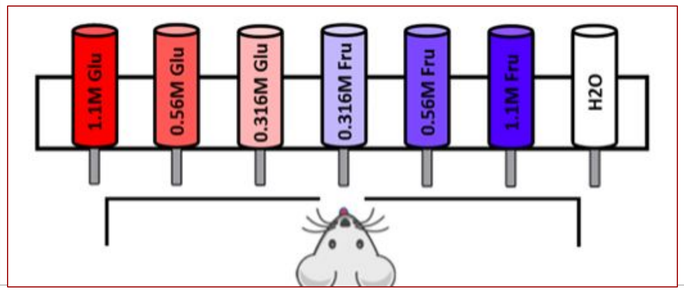
Abstract

Calcium plays a critical role in **neuronal signaling**, and changes in calcium levels can be used as a proxy of neural activity [1]. Calcium imaging with **fiber photometry** was developed to visualize and measure neural activity in animal models. This tool takes advantage of a genetically-encoded calcium indicator, GCaMP, which fluoresces when it binds to calcium under 470 nm excitation/510 nm emission wavelengths [2]. Bulk fluctuations in calcium activity in a particular neuronal population can be imaged with high temporal resolution in **awake, behaving animals.** The goal of this project was to register these changes in calcium activity while sampling specific tastants in order to reveal how a particular brain area is recruited for sensory-reward processing. We developed a code in MATLAB to register the timestamp of animals' licking responses to various sugar solutions that can be coupled to real-time fluctuations in fluorescence in a region of the hindbrain (laterodorsal tegmental nucleus, **LDTg**).



Test Structure

In a brief access taste test, the animal is presented with an array of tastant solutions (one at a time; water, 0.316M, 0.56M, 1.1M of glucose and fructose) in a randomized order. The solutions are presented for 10 second per trial, with a 7.5 second interval between each trial. This 10s trial begins once the animal initiates licking. Each lick is recorded by electronic lickometer and saved for later analyses. The test session lasts 20 minutes.



Objective

The aim of this project was to translate data output from the Davis Rig, which measures lick responses, into a timestamp format to be compared to the photometry output. We created a manuscript in MATLAB to convert the Davis Rig data to a line of timestamps corresponding to the original trial and lick.

Methods & Results

1. The code was constructed using the format of the existing table. In the script, the data is formed into a table with the **latency to initiate the first lick** and the inter-lick intervals (**ILIs**) in each trial, and inter-trial intervals (**ITIs**). This table is then reorganized into a line with each data point and the values are added to produce a **timestamp for each lick**.

Input

```
file = 'DavisRig648.xlsx';
range = 'B262:CD305';
data = xlsread(file,1,range)./1000;
range1='H11:H54';%latency input
data1=xlsread(file,1,range1)./1000;
data(isnan(data))=0;
data2=[7.5;10-sum(data,2)+7.5];
data2(end,:)=[];
latency=data2+data1;
total=[latency,data];
line=reshape(total',1,[]);
line1=line(line~=0);
time=cumsum(line1);
timeStamp=seconds(time)
```

Output

```
Columns 1637 through 1640

   1077.8 sec    1077.9 sec

Columns 1641 through 1644

   1078.6 sec    1078.7 sec

Column 1645

   1086.6 sec
```

2. Running the manuscript prototype revealed that the combined time of the trials often **ran below the twenty minute allotment** for trials. An additional section to the manuscript **adjusts the time loss** due to the motor movements by calculating the remaining time after the last lick and dividing it by total lick numbers.

Adjusted Output

```
perTrial =

  duration

   00:00:00.068
```

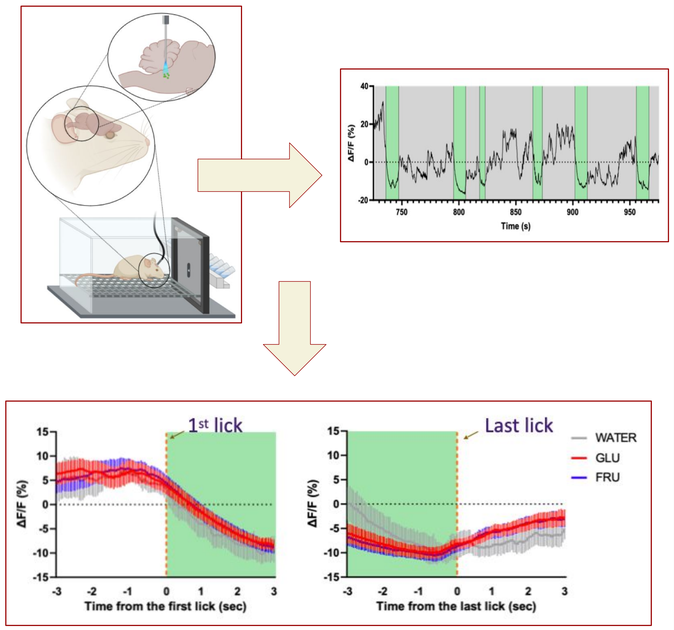
This variable shows difference remaining time divided by the toal number of licks. It is added to the original line of timestamps to adjust the output.

```
line2=line1+perTrial;
```

```
Columns 1642 through 1644

   00:19:51.9471    00:19:52.1250

Column 1645

   00:20:00.0000
```

Future Directions

Having a code that translates the Davis Rig data into a timestamp makes the data **compatible** with the fiber photometry output. The next step is to compare the timestamp output to the graph output of the photometry. This will create a graph marked with the time and solution indicating **patterns of neuronal activity** in relation to the different solutions. The ultimate goal is to **connect the licking behaviors** to the mice to their brain activity.



References & Acknowledgements

[1]Barnett, L. M., Hughes, T. E., & Drobizhev, M. (2017). Deciphering the molecular mechanism responsible for GCaMP6m's Ca2+-dependent change in fluorescence. PloS one, 12(2), e0170934.

[2]Rosenberg, S. S., & Spitzer, N. C. (2011). Calcium signaling in neuronal development. Cold Spring Harbor perspectives in biology, 3(10), a004259.

[3]http://molecular-ethology.bs.s.u-tokyo.ac.jp/labHP/E/EResearch/11.html

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