Analyzing Input and Output Neurons in the Olfactory Bulb of Mice

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Introduction

In Dr. Storace's lab, we aim to determine the olfactory bulb's role in sensory processing and olfactory perception.

After decades of research, we now know that the olfactory bulb is the first stage in the olfactory processing pathway. However, its defined role in the olfactory information processing remains unidentified *(Storace and Cohen, 2017).*

The organization of input and output neurons in the olfactory bulb:

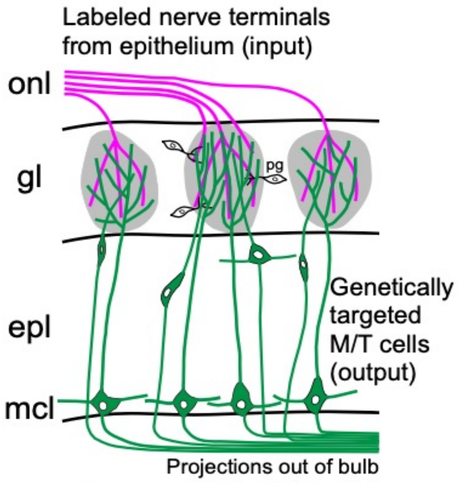


Figure 1. Storace and Cohen, 2017

Throughout these two semesters, the objective of my project has been to image a coronal section of the olfactory bulb, in order to determine its different components:

- Input neurons, output neurons and glomeruli.

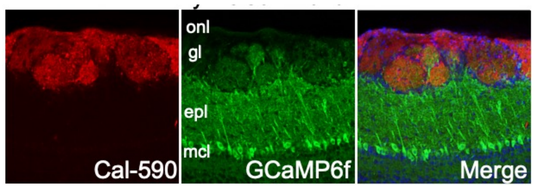


Figure 2. Storace and Cohen, 2017

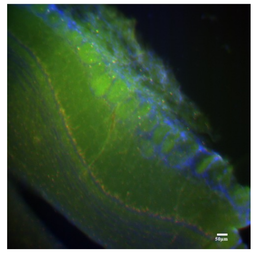
Methods

- **Subjects:** transgenic mice that have been genetically engineered to express distinctive genes that code for fluorescent proteins.
- **Procedure:**
  - Using a vibratome, I cut the brain in 50 µm thick coronal sections, mounted them on a glass slide, stained them with DAPI (a DNA staining gel), and covered the slide with a coverslip, leaving them for at least 24 hours in the lab's fridge to solidify the gel.
  - After 24 hours, I observed them under the fluorescent microscope. Using this technology and changing the colored lens in the microscope, we could distinguish between the different structures:
    - The input neurons labeled with green sensors, the output neurons labeled with red sensors, and the glomeruli.
  - To document my findings, I used the camera on the microscope and a computer software to capture the images. Finally, I used ImageJ to convert the files and save them as pictures in a computer.

Results

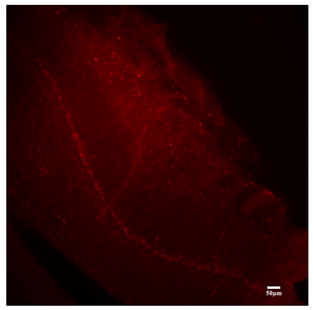
Rcamp, GCaMP

- 10x augmentation
- Red, green, and blue sensors

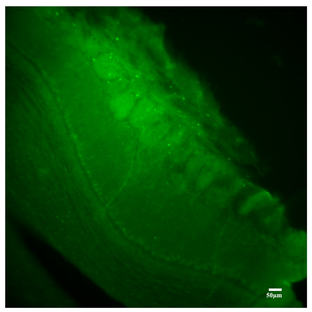


Composite

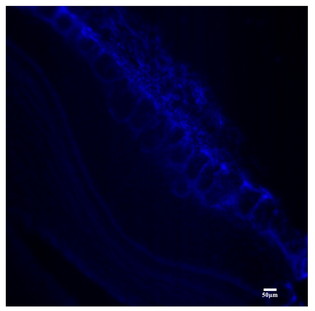
RCaMP, Output neurons



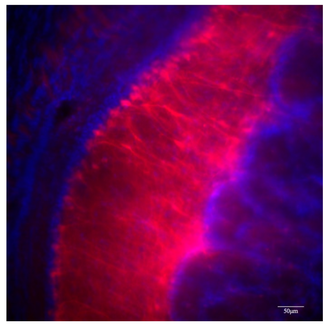
GCaMP Input neurons and glomeruli



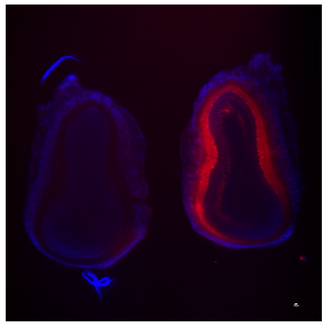
DAPI

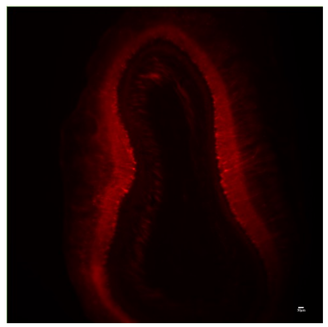


Other Examples:



RCaMP, 20x, Composite





RCaMP, 2.5x & 5x, Composites

Results cont.

- After imaging the sections, we concluded that the experiments went amiss. We are still troubleshooting to determine what caused the unsuccessful imaging. However, we know what the images *should* have looked like.
- The image below was taken at the lab when carrying out previous imaging. This is what the input neurons labeled with green sensors should have looked like.

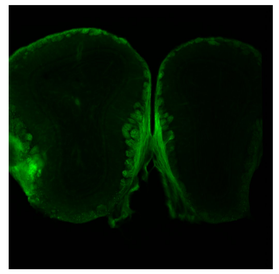


Figure 3. GCaMP6s located in the olfactory nerve terminals .

Analysis

After performing an analysis of the neurons in the olfactory bulb, I now better understand how the most basic neurological pathway in the olfactory bulb works.

- The input neurons (green), located in the nose, gather the sensory information from the organ and sends it to the bulb. The input neurons transmit the information to the output neurons (red) in the bulb, which carry it to the rest of the brain. The input and output neurons exchange the sensory information in the glomeruli, a region specific to different protein receptors where they overlap each other.

References

- Storace DA, Cohen LB. (2017). Measuring the olfactory bulb input-output transformation reveals a contribution to the perception of odorant concentration invariance. Nat Commun. Jul 19;8(1):81.