Immunohistochemical Characterization of the Human Olfactory System

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Abstract

Our project explores the connection between the olfactory system and neurodegeneration by examining the similarities and differences between olfactory receptor neurons in healthy and neurodegenerative olfactory tissue. We are using immunohistochemistry to mark specific olfactory receptor neurons, and subsequently scanning and imaging the tissue. We currently are topographically comparing the neurodegenerative tissue with healthy tissue. Through this project, we strive to better understand neurodegeneration.

Introduction

- The earliest symptoms of neurodegeneration appear in the olfactory system.[1]
- OSNs carry odorant information from the olfactory epithelium (OE) to specific glomeruli in the olfactory bulb (OB).
- We compare the topography of chemoreceptors in neurodegenerative tissue samples with those observed in healthy tissues.

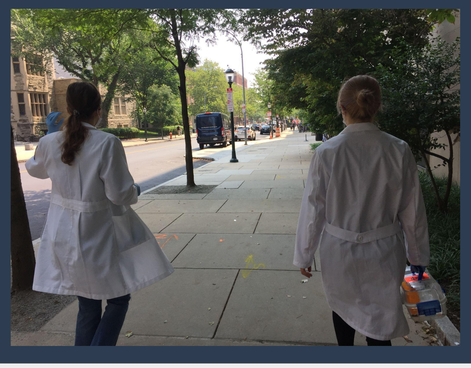
1Braak et al. 1996, J Neural Trans 103:455-490./

Discussion/Conclusion

- Study Objective: We want to compare olfactory receptor topography across neurotypical and neurodegenerative tissue.
- To accomplish this, we are using immunohistochemistry and annotating the images for analysis.
- Longterm Objective: The goal is to clarify the role of the olfactory system in early detection of neurodegenerative pathology and to explore its potential as a marker for early diagnosis

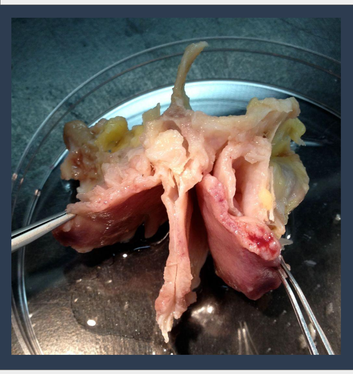
Methods

1 Tissue Pick Up



We pick up the tissue from the Department of Pathology with consent from the patient and/or their family.

2 Fixation and Dissection

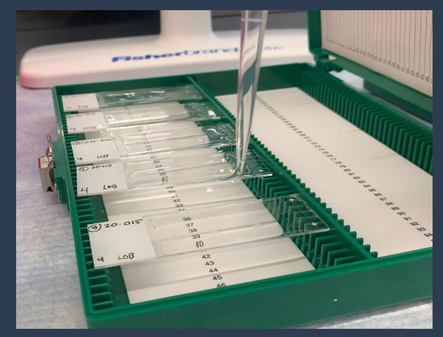


We fix the tissue in 1% paraformaldehyde, decalcify in EDTA, and place in sucrose and sodium azide.

3 Embedding and Sectioning

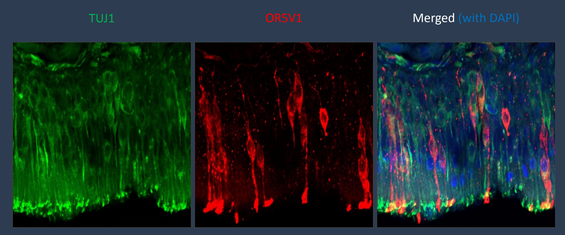
We dissect the tissues and then embed them in optimal cutting temperature compound to be flash frozen at -80 C. Later, we section the tissue at 16 µm on the cryostat.

4 Immunohistochemistry



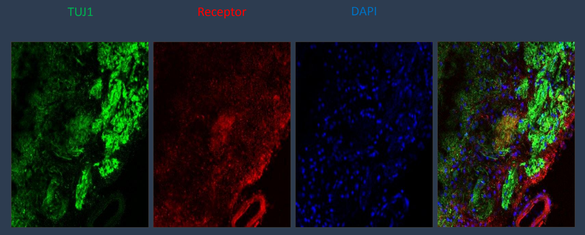
We use customized antibodies against chemoreceptors (OR51E2, OR5V1, OR10G7, and TAAR5) and DAPI. We stain the OE with Tuj1, a neuronal marker in green, and the OB with NCAM2 and VGlut2, a glomerular marker in green.

5 OE Annotation



We mark OSNs in OE and OB with Zeiss ZEN Imaging software. In the OE, OSN marking consists of: a strong DAPI signal (blue) in the soma with OR of interest (pink) and TUJ1 (green) overlapping around the circumference.

6 OB Annotation



In the OB, glomeruli marking consists of an overlap in OR of interest (pink) and NCAM2 and VGlut2 (green) without DAPI (blue).