

Automatic Segmentation and 3D Reconstruction of the Inner Ear Vasculature from Histology Slides

Project proposal

Aseem Jain

Table of Contents

<i>Clinical Motivation</i>	2
<i>Prior work</i>	2
<i>Goals and Significance</i>	3
<i>General Experimental Setup</i>	3
<i>Technical Approach</i>	4
<i>Milestones and Deliverables</i>	5
<i>Dependencies</i>	6
<i>Team Members/Role</i>	8
<i>Management Plan</i>	8
<i>Reference</i>	9

Clinical Motivation

Researchers have often relied on analysis of histopathology slides of the inner ear from deceased humans/mammals to understand the impact that different structures have on the various pathologies in otolaryngology. Understanding the pathophysiology of the inner ear remains crucial to guiding treatment strategies in otolaryngology. For instance, the stria vascularis, a structure in the inner ear analyzed on histology slides, has been thought to be critical to understanding sensorineural hearing loss in mammals [1]. Additionally, through studying histology slides of patients with inner ear malformation, researchers were able to create a better model for cochlear implants in this population [2].

One area of interest within the realm of inner ear histopathology is the study of vascular in the inner ear diseases. Currently, vascular disorders of the inner ear remain poorly understood. Some researchers have hypothesized that abnormal vasculature may play a role in pathologies such as vestibular neuritis [3]. These researchers note that being able to analyze the vasculature with 3D models of histopathology could prove to be valuable as it would enable them to better understand the positional relationship between vasculature and adjacent structures.

However, despite the importance of analyzing histology slides, most researchers still rely on manual methods for segmentation of structures within the inner ear. Manual segmentation techniques can also lead to poor interrater reliability [4]. Furthermore, few researchers have the tools to be able to generate 3D models of structures within the inner ear. While automatic techniques to extract relevant features from whole slide histology images (WSI) is rapidly expanding, little work has been done to automate segmentation histology slides of the inner ear and construction 3D models from these segmentations.

Prior work

Prior groups have tackled the issue of segmenting and analyzing WSI using neural networks. Segmenting WSI poses many challenges including 1) large image files often on the order of 1-2 GB per image 2) stain variability between slices 3) extracting clinically relevant features from large images. Khened et al proposed a generalized framework for addressing these issues; the researchers used an ensemble of neural networks that were based upon a Unet with various backbones to improve the overall accuracy of the model. This group primarily used a Jaccard Index, measure of overlap between generated labels and ground truth, to quantify their accuracy and achieved a score of .67-78 for certain datasets [4]. Guo et al focused on creating quick, yet still accurate, deep learning methods to analyze breast images. They used a patch-based method to extract cancerous regions and then evaluated these regions at different resolutions to gather coarse and fine features [5].

Previous groups have generated 3D reconstructions of histology slides for a variety of applications. The researchers mentioned in [3] manually aligned segmented histology slides together to generate a 3D model. Other researchers have used tools such as simple elastix to perform alignment and registration of histology images to generate 3D models of bladder cancers [6].

Goals and Significance

Goals

The key deliverable at the end of this project is to create software that can

- 1) Use deep learning methods to segment histopathology slides of the ear
- 2) Use these segmentations to create a 3D reconstruction of the ear

These goals can further be organized into

- Minimum: Create software to perform semantic segmentation of the inner ear of macaque(monkey) ears
- Expected: Software that can segment and reconstruct a 3D volume of the inner ear of monkeys
- Maximum
 - Write and submit a manuscript that details proposed new software
 - Use software to analyze human ears: extract clinically relevant features of such as volume, density of cells, curvature, etc

Significance

Completion of this project will enable researchers to analyze WSI of the inner ear rapidly and accurately compared to existing manual techniques. Additionally, by adding the ability to view these segmentations in 3D, researchers will have new insights into the role that structures such as the vasculature can play in various pathologies. Creation of this software could also facilitate the ability to study other structures within inner ear WSI. Overall, the proposed project has the potential to dramatically improve the way researchers analyze WSI within the inner ear.

General Experimental Setup

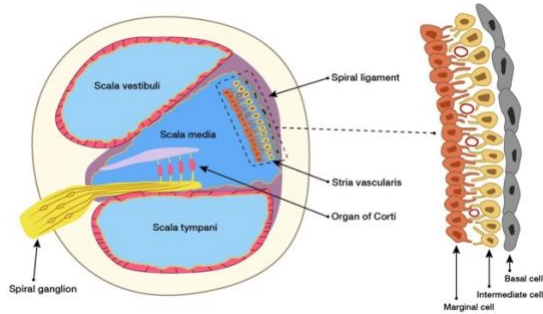
Temporal bone slices of 4 monkey ears have already been sectioned for this project. Each ear contains 81 slides spaced 10 micrometers apart. The ears themselves are assumed to have no pathologies associated with them. To convert these slices into digital images, an Olympus microscope has already been setup in the Lauer lab at JHMI. The members on this project have already been trained to use this microscope to generate these images.

To facilitate quicker creation of the software and the associated deep learning framework, access to a virtual GPU has already been provided. The configuration of the GPU is a GeForce RTX 3090 with 32 Gb of RAM. Additional software requirements will be run locally on the team lead's MacBook Pro.

Technical Approach

Broadly, the technical approach can be divided into two phases. 1) Creation of the deep learning framework for semantic segmentation of inner ear vasculature. 2) Alignment of segmentations and generation of 3D model

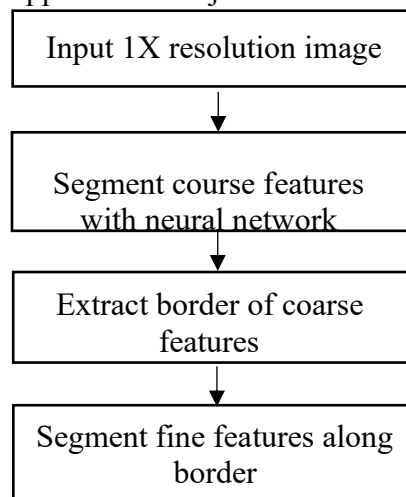
Phase 1 Deep Learning Framework



Data Acquisition: To create a deep learning framework, some WSI (40 images) of monkey ears must first be manually segmented. Segmented features will include inner ear vasculature, Scala media, Scala vestibuli and cranial nerve VIII.

Model Training: Like the approach outlined by Guo et al; various 300x300 patches will be acquired from the labeled data. Fine features will be extracted at the 20X resolution of the images while coarse features will be extracted from the 1X resolution. A U-net will be used initially to perform the semantic segmentation.

Semantic Segmentation: A coarse to fine approach may be used for semantic segmentation. This approach is subject to future change.



A 1X input image will be used initially to extract coarse/large features such as Scala vestibuli, CN VIII, etc. Once these features are extracted, a border will be created around them. Given that most vasculature is positionally nearby coarse features, the border will serve as a search boundary to extract and segment fine features from the 20X resolution. This method speeds up the processing time as only a few patches are analyzed along the search boundary. This method also addresses the challenge of extracting small features relative to coarse features. A similar approach was proposed in Guo et al for cancer segmentation.

Phase 2

Phase two of the project involves aligning and generating a 3D model of the inner ear vasculature. Segmented WSI from Phase 1 can be aligned with tools such as simple elastix. Open source software such as Slicer 3D can be used to render 2D segmentations into a 3D model.

Milestones and Deliverables

	Activities	Deliverable	Proposed Completion date
Minimum	Scan and Label Appropriate structure from 1 monkey ear	40 labeled WSI; 81 scanned slides total	3/6
	Implement U-Net with proposed algorithm	Functional software that segments vasculature from WSI	3/16
	Validation of U-Net on test data	Internal validation report that analyzes non-labeled slides	3/23
Expected	Align segmented images	Software that can align WSI slides; validate against manual alignment	4/4
	Reconstruction in 3D	3D mesh file that can be assessed by neurotologist	4/11
Maximum	Manuscript writing	Submitted manuscript	4/26
	Analysis of human temporal bone	3D mesh file that can be assessed by neurotologist. Report that includes clinically relevant features such as volume, curvature, etc.	5/1

Dependencies

This project requires digital WSI that will be provided by the Lauer Lab. Outside of this dependency most requirement of this project will be based on open source platforms. Other dependencies are outlined below

Dependencies	Need	Status	Follow up	Contingency Plan	Planned	Hard
Access to sectioned Macaque Ears	Used to create training	Completed	n/a	Use existing temporal bone scans	2/16	2/23
Access to Olympus Microscope	Need to scan sectioned slices of macaque Ears	Completed	n/a	n/a	2/16	2/23
Label Validation	Need to have Dr. Lauer/ Dr. Creighton/ Dr. Ward validate accuracy of labeled training data	In progress	n/a	Validate with post-doc in Lauer lab	2/28	3/6
Access to Virtual GPU	Need access to virtual GPU to create deep learning model	Completed	n/a	Run deep learning framework locally	n/a	n/a

Team Members/Role

- Aseem Jain: *Masters Student, MS4 University of Cincinnati*
 - Project Lead: Design/Validate algorithm to accomplish Phase I/II
- Dianela Perdomo: *MS2 Johns Hopkins*
 - Will assist with scanning/labeling slides; will also assist with manuscript write up
- Mentors
 - Francis (Pete) Creighton, MD: Expertise in lateral skull base surgery.
 - Bryan Ward, MD: Expertise in neurotology.
 - Amanda Lauer, PhD: Expertise in various pathologies related to otology.
 - Russel Taylor, PhD: Expertise in medical imaging, computer-integrated surgery.

Management Plan

- Meetings: Biweekly meetings with medical student until scans are completely segmented; as-needed meetings with mentors.
- Monthly Committee meetings (March, April): These meetings will serve to have all the members of the team assess the progress/provide input on the direction of the project
- Additional communication with mentors will happen over email and/or text
- Documentation management
 - Code will be maintained on a private Git repository.
 - Google docs will be used for basic report writing, final report may be generated in word and/or overleaf

Reference

1. Yu W, Zong S, Du P, Zhou P, Li H, Wang E, Xiao H. Role of the Stria Vascularis in the Pathogenesis of Sensorineural Hearing Loss: A Narrative Review. *Front Neurosci*. 2021 Nov 19;15:774585. doi: 10.3389/fnins.2021.774585. PMID: 34867173; PMCID: PMC8640081.
2. Monsanto RDC, Sennaroglu L, Uchiyama M, Sancak IG, Paparella MM, Cureoglu S. Histopathology of Inner Ear Malformations: Potential Pitfalls for Cochlear Implantation. *Otol Neurotol*. 2019 Sep;40(8):e839-e846. doi: 10.1097/MAO.0000000000002356. PMID: 31361687; PMCID: PMC7377297.
3. Büki B, Mair A, Pogson JM, Andresen NS, Ward BK. Three-Dimensional High-Resolution Temporal Bone Histopathology Identifies Areas of Vascular Vulnerability in the Inner Ear. *Audiol Neurootol*. 2022;27(3):249-259. doi: 10.1159/000521397. Epub 2021 Dec 29. PMID: 34965531; PMCID: PMC9133178.
4. Khened, M., Kori, A., Rajkumar, H. *et al*. A generalized deep learning framework for whole-slide image segmentation and analysis. *Sci Rep* 11, 11579 (2021). <https://doi.org/10.1038/s41598-021-90444-8>
5. Guo, Z., Liu, H., Ni, H. *et al*. A Fast and Refined Cancer Regions Segmentation Framework in Whole-slide Breast Pathological Images. *Sci Rep* 9, 882 (2019). <https://doi.org/10.1038/s41598-018-37492-9>
6. Jansen, I., Lucas, M., Savci-Heijink, C.D. *et al*. Three-dimensional histopathological reconstruction of bladder tumours. *Diagn Pathol* 14, 25 (2019). <https://doi.org/10.1186/s13000-019-0803-7>