Multi-resolution open-top light-sheet (OTLS) microscopy for rapid 3D pathology

MECHANICAL ENGINEERING
UNIVERSITY of WASHINGTON

Lindsey A. Barner[1], Dr. Adam K. Glaser[1], Dr. Jonathan T.C. Liu[1,2](PI)
[1]University of Washington, Department of Mechanical Engineering, [2]University of Washington, Department of Pathology

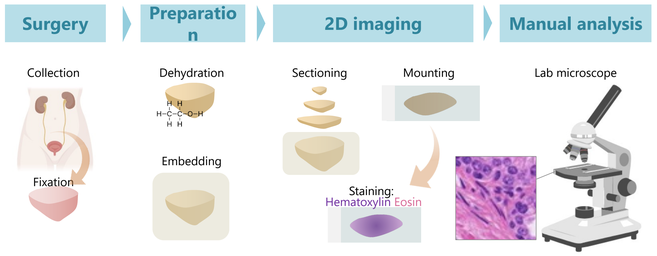
IMECE 2019-13009

IMECE Track 16-1
NSF Research Poster Competition

Background and motivation

The gold standard for prognosis and treatment stratification of cancer patients relies on conventional histopathology, a dated and labor-intensive technique that suffers from limited 2D sampling and inter-observer variability.

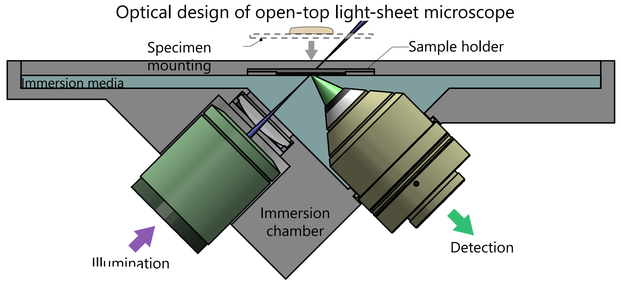
This compromises the accuracy of cancer diagnoses, which leads to over-treatment of patients with indolent cancer (non-life-threatening disease), or under-treatment of high-risk patients which leads to increased mortality.



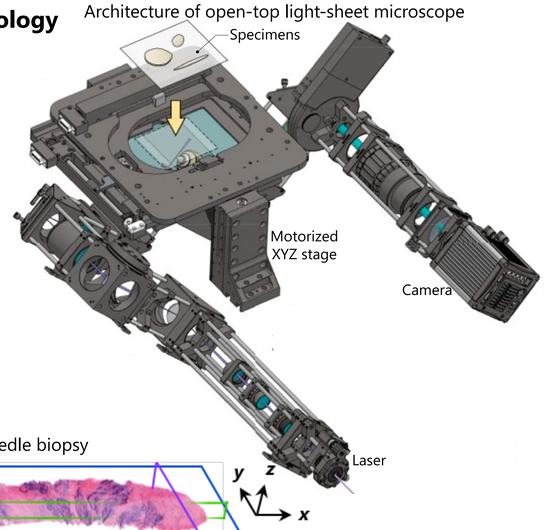
Prior work: Open-top light-sheet microscopy for rapid 3D pathology

We have developed open-top light-sheet microscopy for nondestructive, slide-free and volumetric pathology of clinical specimens. Intact tissues can be rapidly imaged over large volumetric fields of view. The utility of this technology has been demonstrated for several applications in clinical pathology, including volumetric assessment of core-needle biopsies *in toto*.

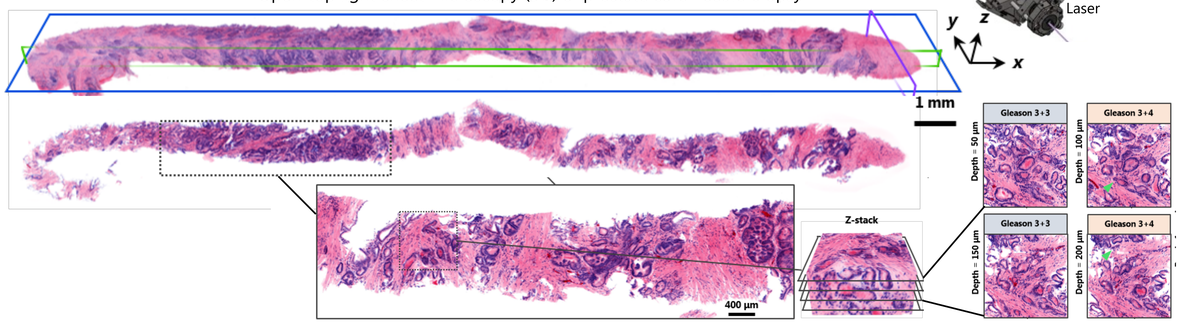
Optical design of open-top light-sheet microscope



Architecture of open-top light-sheet microscope



Open-top light-sheet microscopy (3D) of prostate core needle biopsy

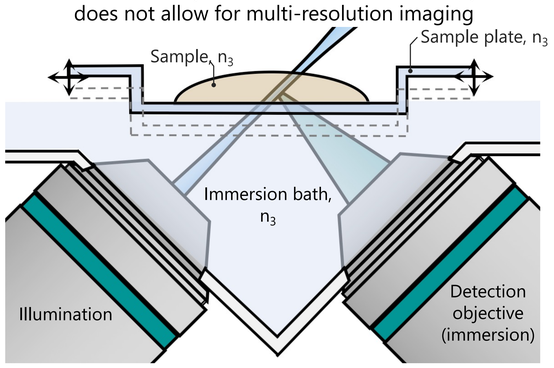


Goal: *Multi-resolution* open-top light-sheet microscopy for rapid 3D pathology

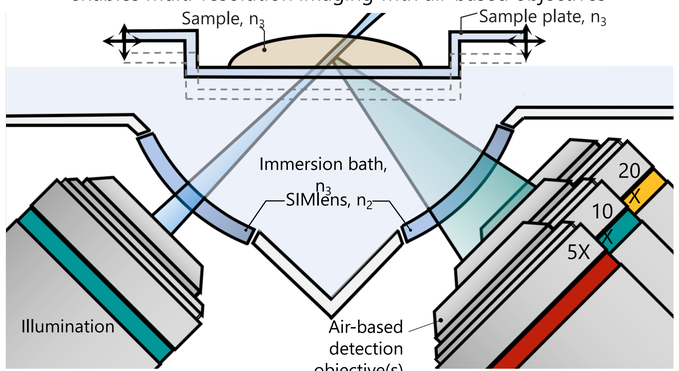
In current histology practice, pathologists first view slide-mounted histology sections at low magnification to rapidly inspect large tissue regions before subsequently imaging ambiguous tissue regions at higher magnification. Similarly, the ability to perform multi-resolution (i.e. multi-magnification) imaging in OTLS microscopy would improve its utility in 3D pathology, further accelerating the translation of OTLS into clinical settings.

Therefore, we aim to enable multi-resolution OTLS microscopy by developing a **solid immersion meniscus lens (SIMlens)**, which enables the use of air-based detection objectives that can be easily switched for multi-magnification imaging (unlike immersion-based objectives).

Immersion-objective-based OTLS microscopy:
does not allow for multi-resolution imaging



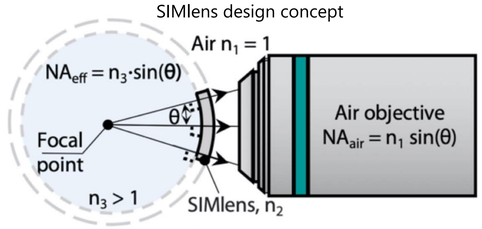
SIMlens-based OTLS microscopy:
enables multi-resolution imaging with air-based objectives



Experimental setup and results:

Design and validation of a SIMlens to enable multi-resolution imaging in light-sheet microscopy

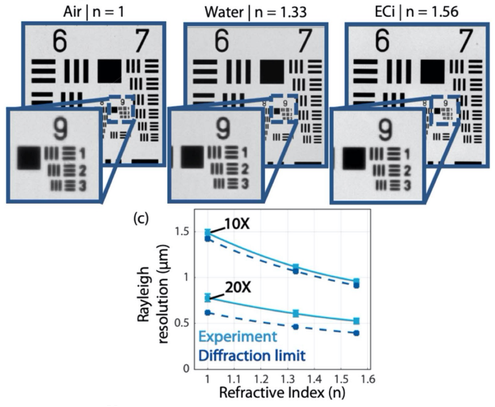
SIMlens design concept



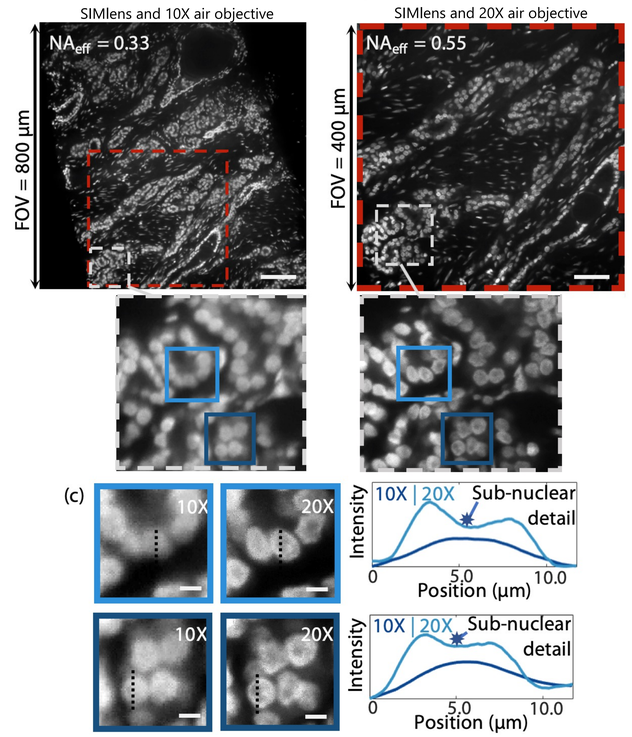
A SIMlens provides a wavefront-matching interface between an air-based objective and any immersion media. This provides a number of advantages in context of OTLS microscopy:

- *Boost in NA by a factor of n_3*
- *Accommodates samples with a wide variety of clearing protocols*
- *Enables vertical movement of the sample (in contrast to 1st-generation OTLS microscopy)*

Performance validation of a SIMlens:
Images of resolution target in various immersion media



Multi-resolution imaging demonstration of a prostate core-needle biopsy with SIMlens-based light-sheet microscopy

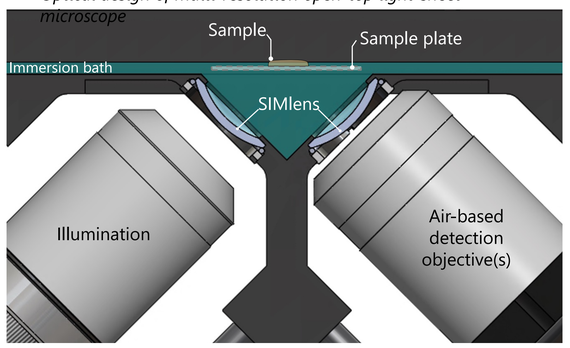


The same region of the tissue is imaged with 10X and 20X air-based objectives and a SIMlens, demonstrating the ability to rapidly switch between objectives to zoom into regions of interest in the specimen.

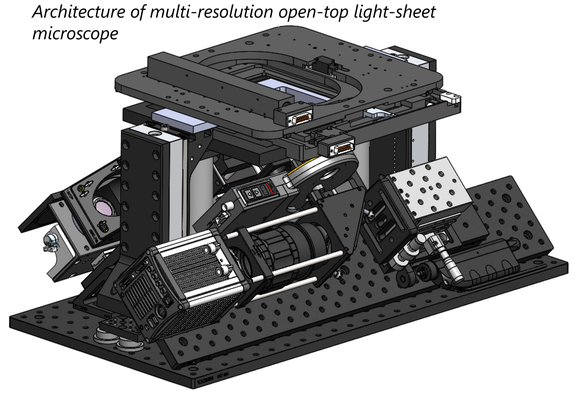
Future studies:

Now that the design and performance of a SIMlens has been validated, our goal is to design and assemble a SIMlens-based OTLS microscope for imaging post-operative prostatectomy specimens. The ability to rapidly screen at low magnification and subsequently zoom in at high resolution would enable rapid screening for lymphovascular invasion, a prognostic indicator that due to its sparse distribution in prostate tissue is difficult to identify with 2D conventional histology.

Optical design of multi-resolution open-top light-sheet microscope



Architecture of multi-resolution open-top light-sheet microscope



Conclusion:

We have developed a SIMlens, a wavefront-matching lens element designed for multi-resolution open-top light-sheet microscopy. In addition to enabling multi-resolution imaging of clinical specimens in 3D, a SIMlens provides a number of advantages over previous OTLS architectures.

References and publications:

Barner, L. *et al.* Solid immersion meniscus lens (SIMlens) for open-top light-sheet microscopy. *Optics Letters.* **44** (18) 4451-4454 (2019).

Glaser, A. *et al.* Light-sheet microscopy for slide-free non-destructive pathology of large clinical specimens. *Nature Biomedical Engineering.* **1** (0084) 1-10 (2017).

Glaser, A. *et al.* Multi-immersion open-top light-sheet microscopy for high-throughput imaging of cleared tissues. *Nature Communications.* **10** (1038) 1-10 (2019).

Acknowledgments: NSF GRFP (Barner), Achievement Rewards for College Scientists Foundation (Barner), Prostate Cancer Foundation, U.S. Department of Defense (PC170176), NIH