Towards an open high-performance platform for fully-automated analysis of whole organ light-sheet fluorescence microscopy data

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1. Analysis of whole organs

Light-sheet fluorescence microscopy (LSFM) allows quantitative three-dimensional analysis of whole organs. This includes the evaluation of structural changes such as a reduced number of glomeruli in kidneys [1] or the formation of bronchus-associated lymphoid tissue (BALT) caused by lung inflammation [2].

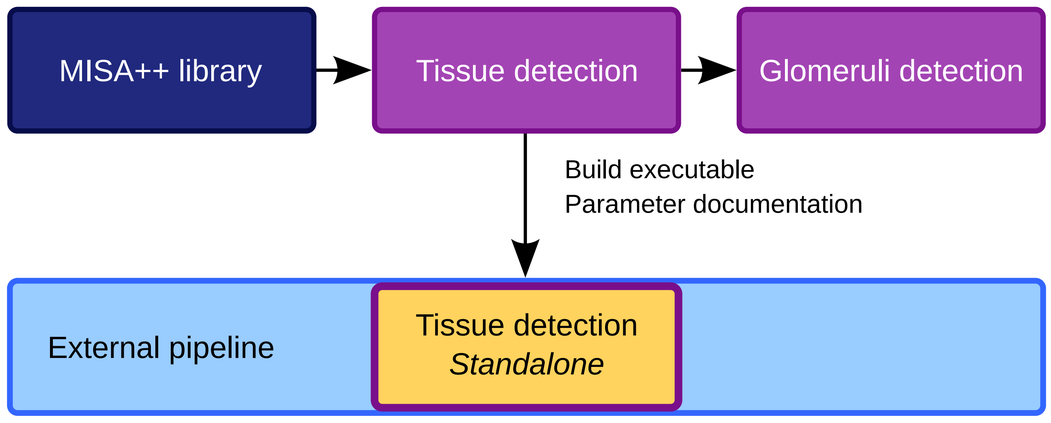
Memory intensive
Long processing } Large datasets
Repeated code Many common tasks for different data

2. Framework for automated analysis

MISA++

Modular Image Stack Analysis for C++

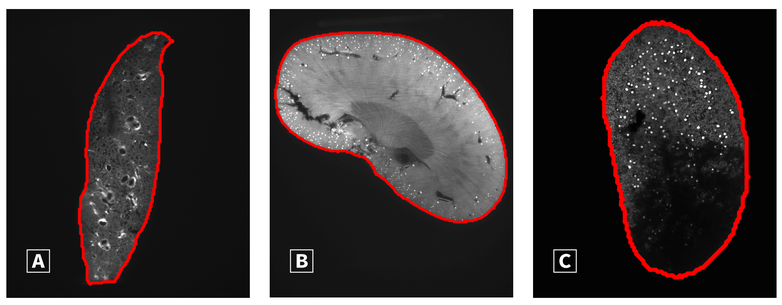
Memory-efficient Implemented in modern C++
Fast Automated parallelization of workloads
Flexible Reusable modules for easy extension and integration



MISA++ modules can be used in C++ code to create other modules or exported to a standalone executable that can be integrated into other pipelines or applications such as Fiji/ImageJ.

3. Tissue detection module

2D segmentation
Algorithms based on percentiles, superpixels or auto thresholding



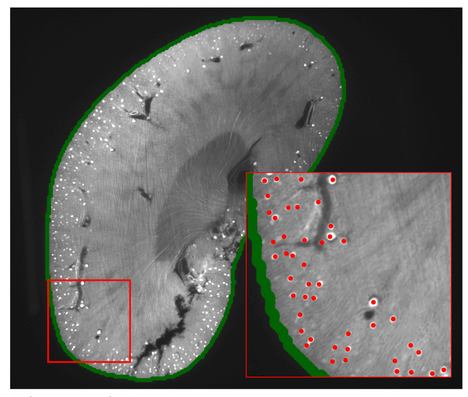
2D Segmented tissue of Lung A and Kidney B, C

Future: Graph assisted 3D object detection
Removal of false positive 2D tissue detection results

Quantification
Number of pixels, volume

4. Glomeruli detection module

Glomeruli are functional structures within the renal cortex that are damaged by diseases and toxins [1].

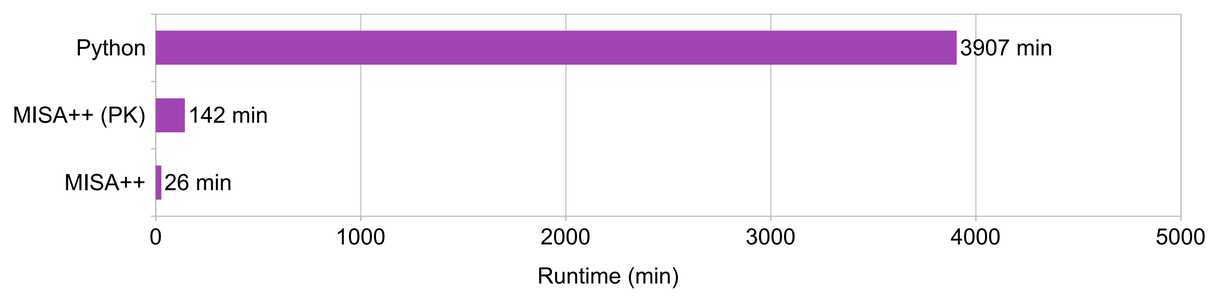


Glomeruli (red) inside the tissue (green)

Tissue segmentation
MISA++ module

2D glomeruli segmentation
Klingberg et al. or Localized Otsu + Shape

3D object detection and quantification
Number of glomeruli, volume, diameter

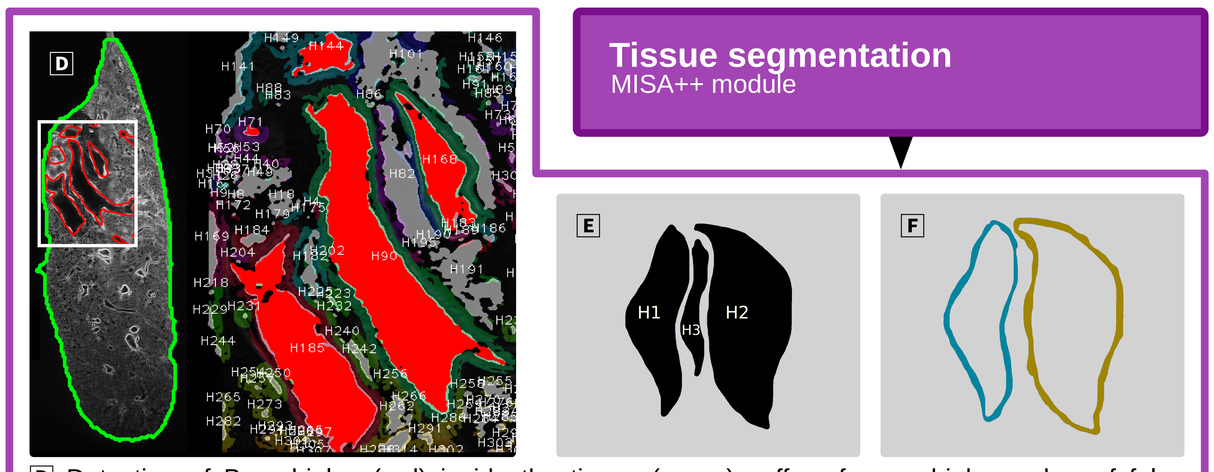


In comparison to the implementation published by Klingberg et al. [1] (Python), MISA++ calculates the same work up to 148 times faster if all images are available at the same time. If the analysis is done per kidney (PK), the calculation is still approximately 27 times faster. Parallelization using 30 threads.

5. Bronchioles detection module (In progress)

Automated assessment of Bronchus-associated lymphoid tissue (BALT) to investigate lung infection requires to segment the bronchioles of the lung, visible as holes with strong borders in LSFM images. We are currently developing an approach to segment those highly irregular structures.

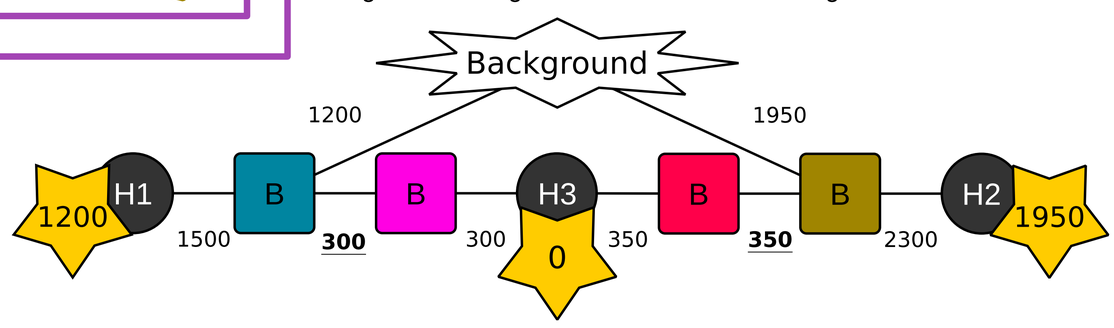
Tissue segmentation
MISA++ module



D Detection of Bronchioles (red) inside the tissue (green) suffers from a high number of false positive holes and borders, making it difficult to segment true positive objects. To solve this issue, holes E are segmented independently from borders F.

Border splitting
Borders (colored) shared by multiple holes (black) are split between the holes.

A graph is built, with holes and borders as edges. The edge weight is the number of neighboring pixels between holes/borders and to the background. The score of a hole (star) is the summarized flow to the background. The goal is to remove all B-B edges.



References:
[1] Anika Klingberg et al., “Fully Automated Evaluation of Total Glomerular Number and Capillary Tuft Size in Nephritic Kidneys Using Lightsheet Microscopy,” Journal of the American Society of Nephrology: JASN 28, no. 2 (February 2017): 452–59, https://doi.org/10.1681/ASN.2016020232.
[2] David Twapokera Mzinza et al., “Application of Light Sheet Microscopy for Qualitative and Quantitative Analysis of Bronchus-Associated Lymphoid Tissue in Mice,” Cellular & Molecular Immunology, February 12, 2018, https://doi.org/10.1038/cmi.2017.150.

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