

ENHANCING BIOMARKER QUANTIFICATION WITH AI THROUGH QUPATH

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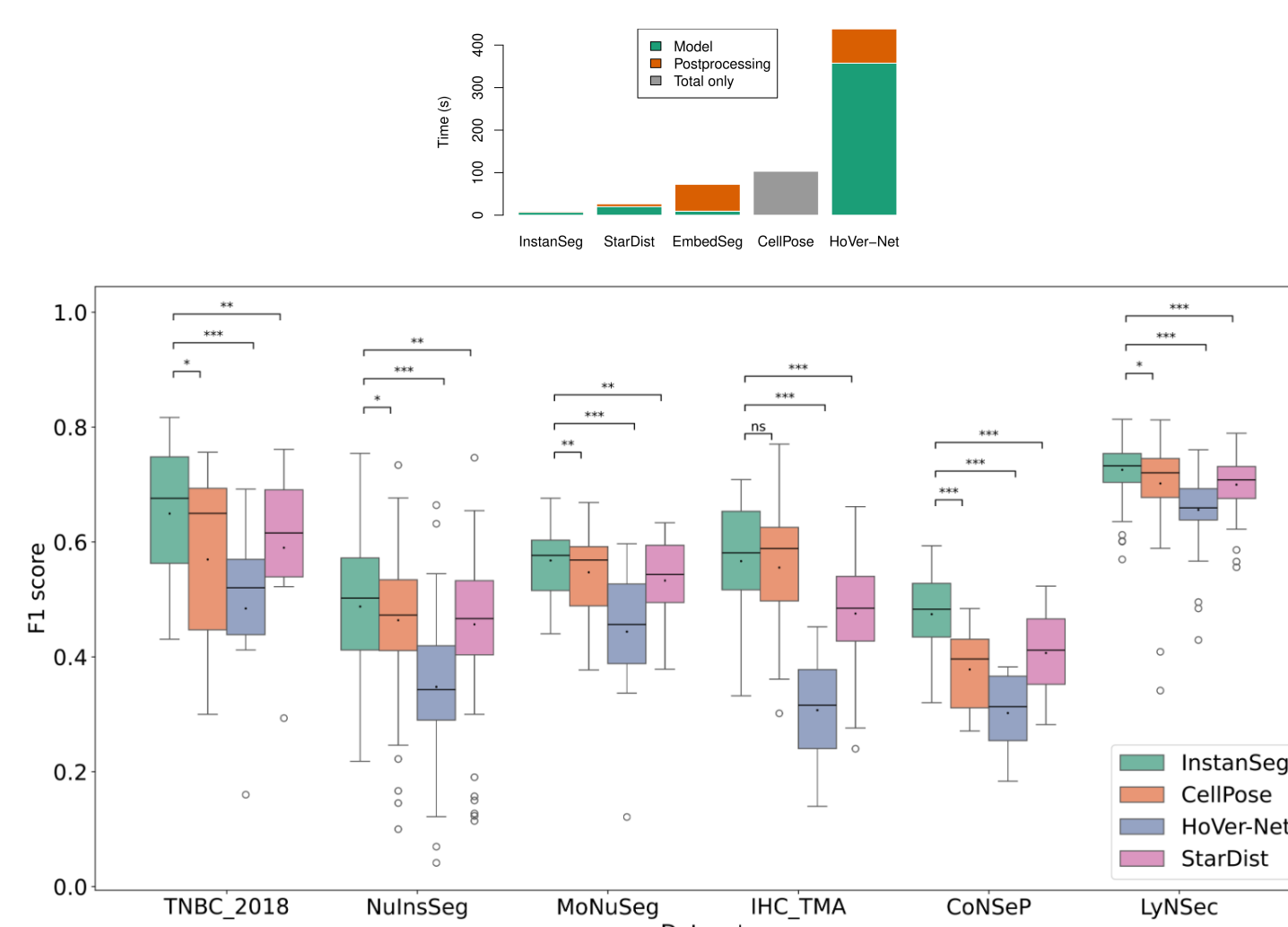
QuPath: open source bioimage software



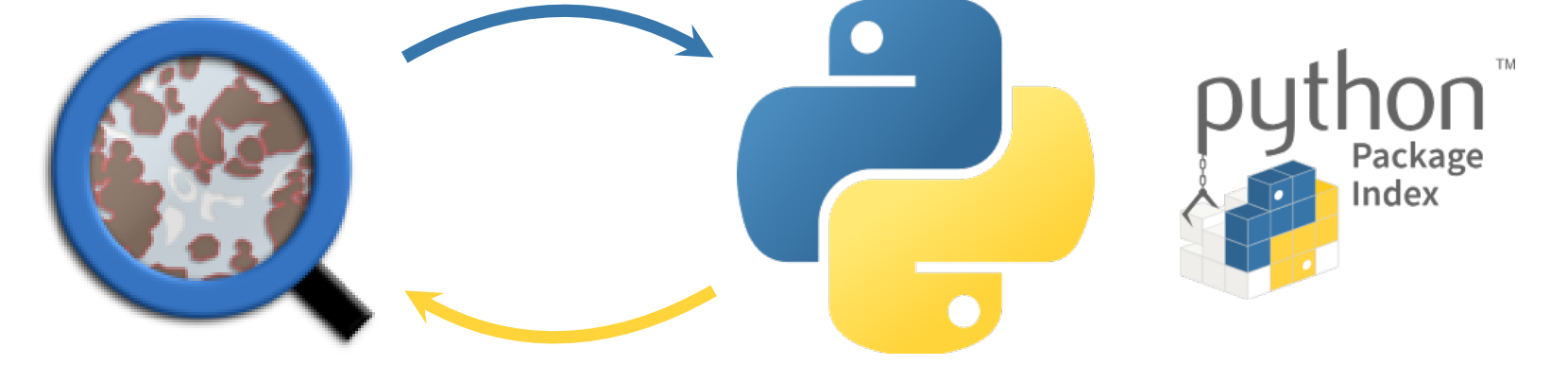
- > 800,000 software downloads
- > 5,800 paper citations
- Widely used in industry and academia
- Powerful annotation, visualization, ML tools
- Designed for large 2D brightfield H&E and IHC images
- Capable with large fluorescence images (< 10 channels)
- Increasingly well-adapted for high-dimensional images (> 10 channel, Z-stacks, time)

InstanSeg: deep learning cell detection

- State-of-the-art instance segmentation of:
 - Brightfield (nuclei)
 - Fluorescence/multiplex (nuclei and membrane)
 - Single-channel (nuclei)
 - Other objects (training required)
- Accurate and fast



QuBaLab: python toolkit for QuPath



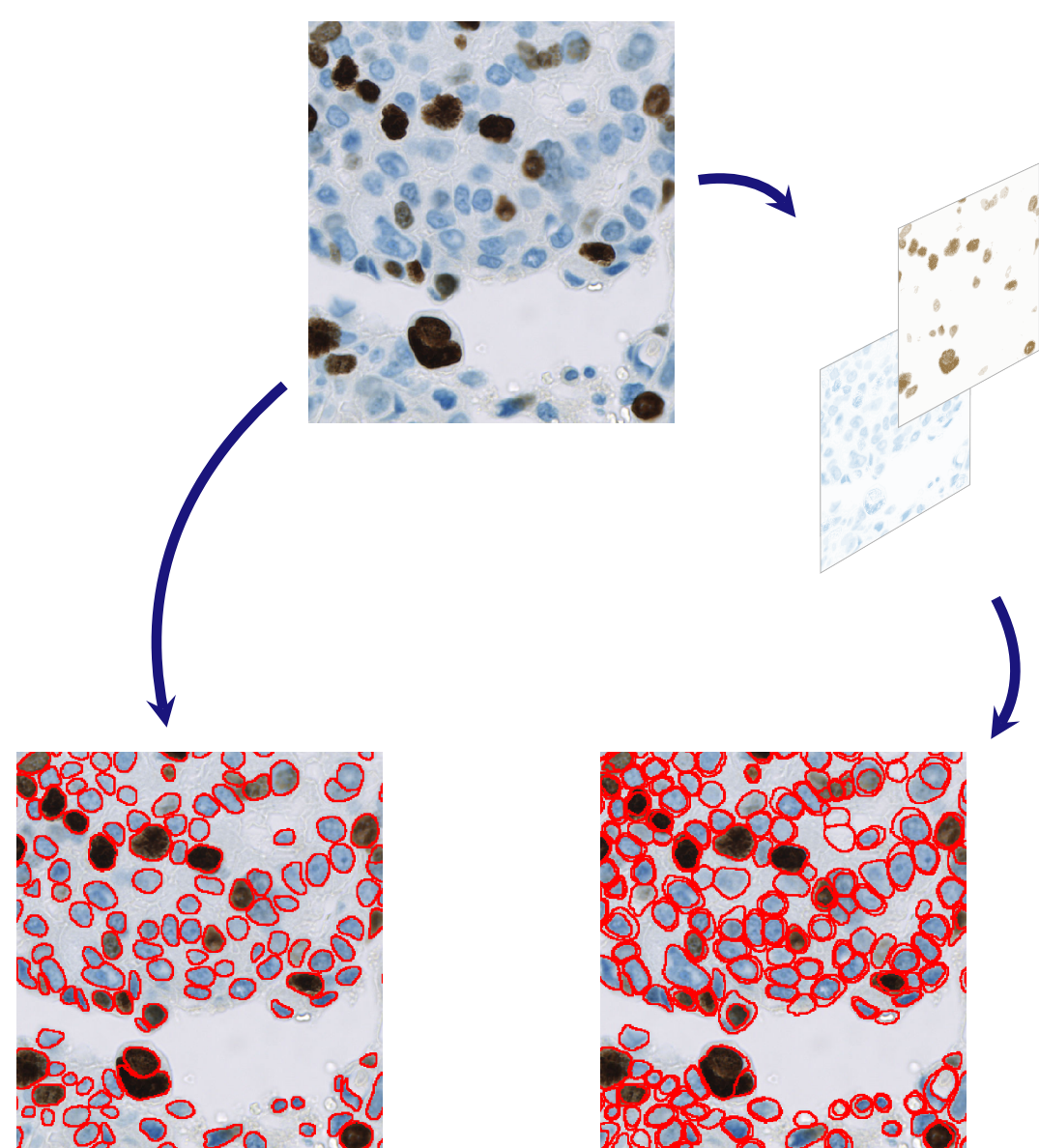
- Enables easy reading and writing of QuPath data
- Direct communication between python and QuPath
- Thorough unit testing
- Opens QuPath's powerful tools (image reading, annotation, visualization) for easy use in deep learning workflows
 - Image features (detections, annotations)
 - Labelled images
 - Convenient wrapping of image readers
 - ICC profiles

InstanSeg is powerful and flexible

For IHC images, InstanSeg can be used directly to segment RGB images or supplied with color deconvolution stains.

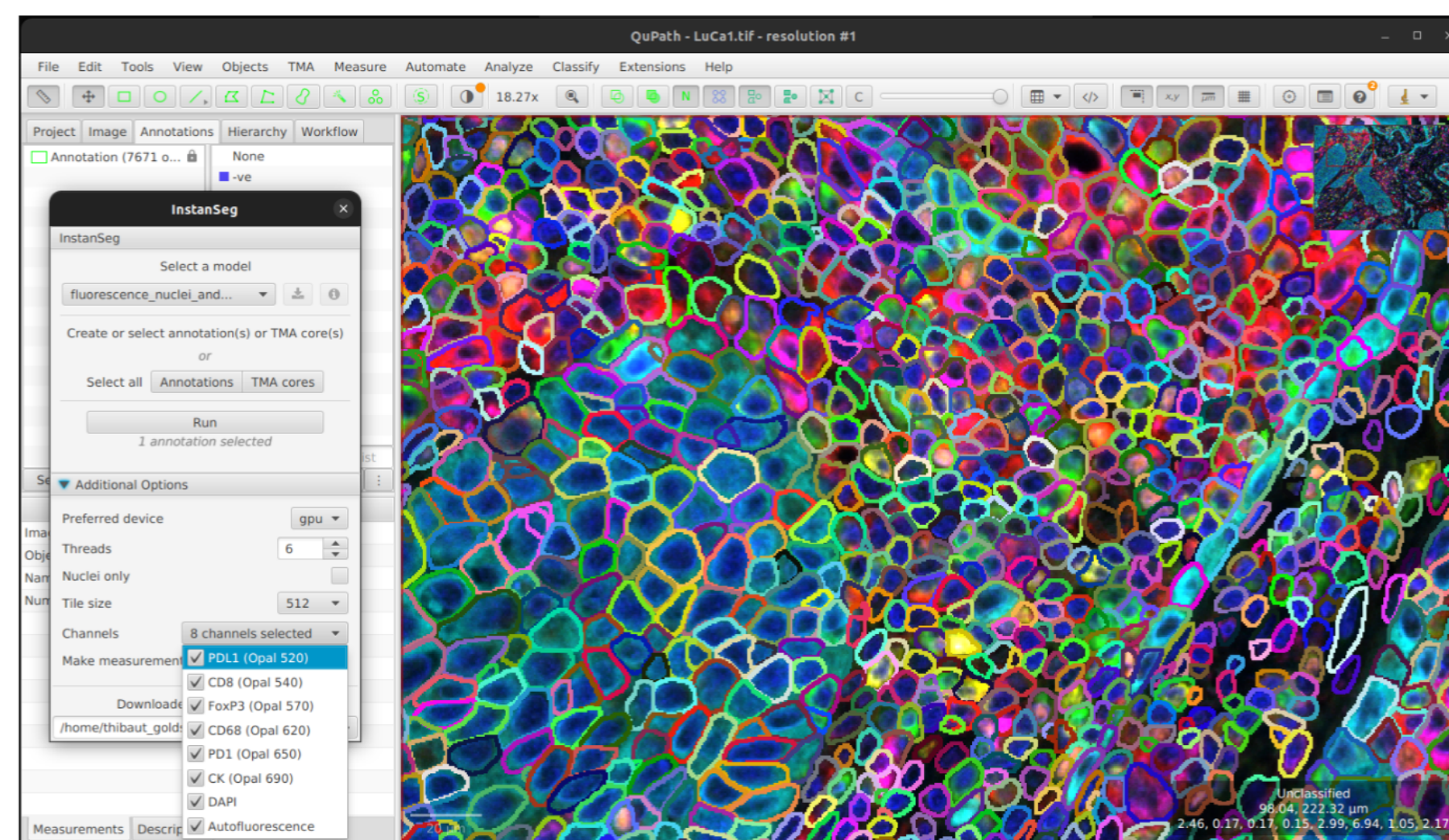
This enables rough estimation of nuclear and cytoplasmic boundaries.

Results vary hugely depending on the stains and tissue.



QuPath + InstanSeg

- Streamlined and highly optimized workflow
- GPU-accelerated (CUDA and MPS)
- Integrated directly with QuPath
- Easy selection of inputs (e.g., fluorescence channels, color deconvolution, pixel sizes...)
- Comprehensive output options for downstream analysis or machine learning



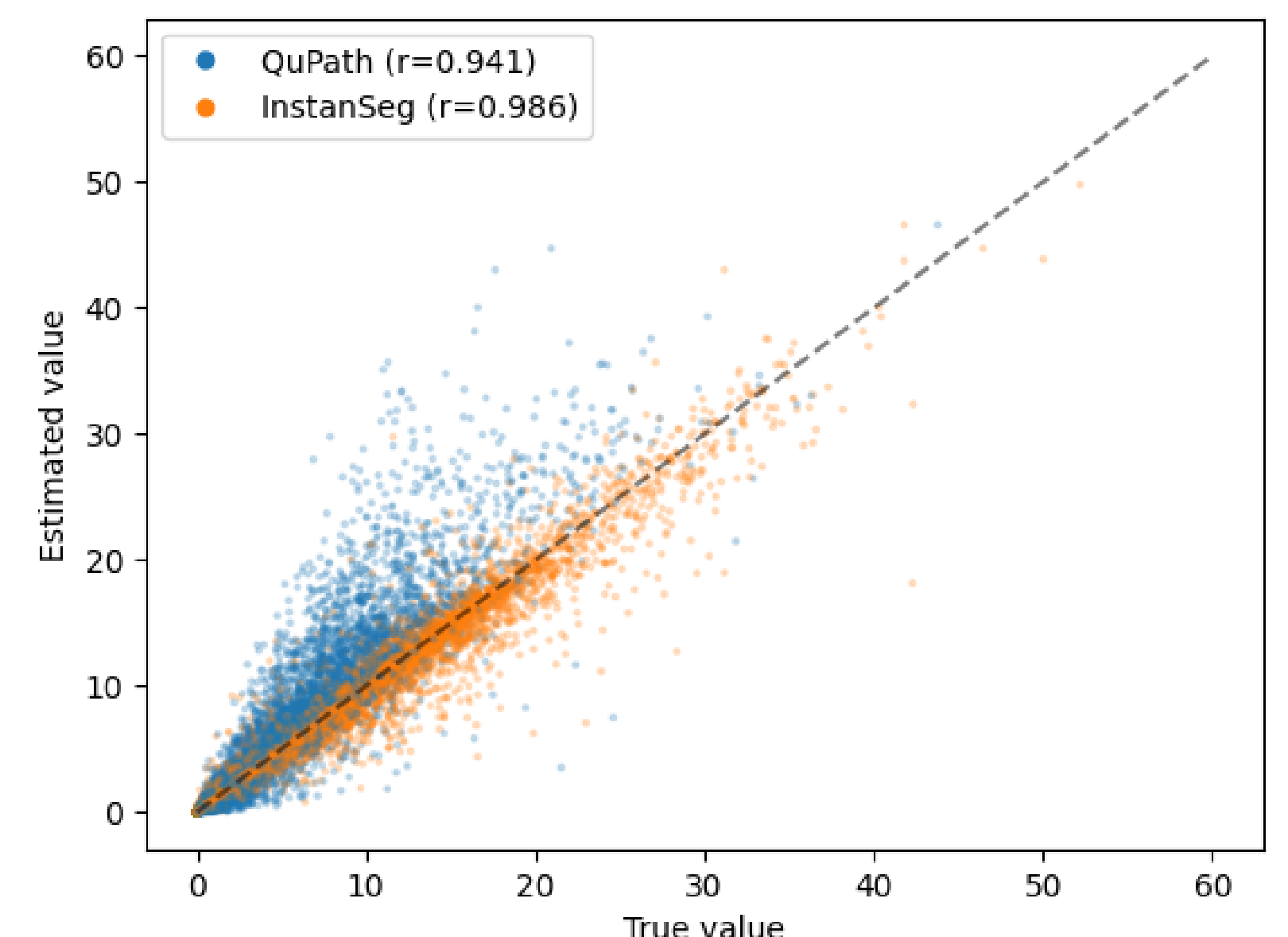
More accurate cell phenotyping

Accurate segmentation is key to accurate cell phenotyping.

InstanSeg gives more precise cell counts (see above).

The underlying cells and tissue are more accurately phenotyped.

Based on ground truth labels from Aleynick *et al.* (2023).

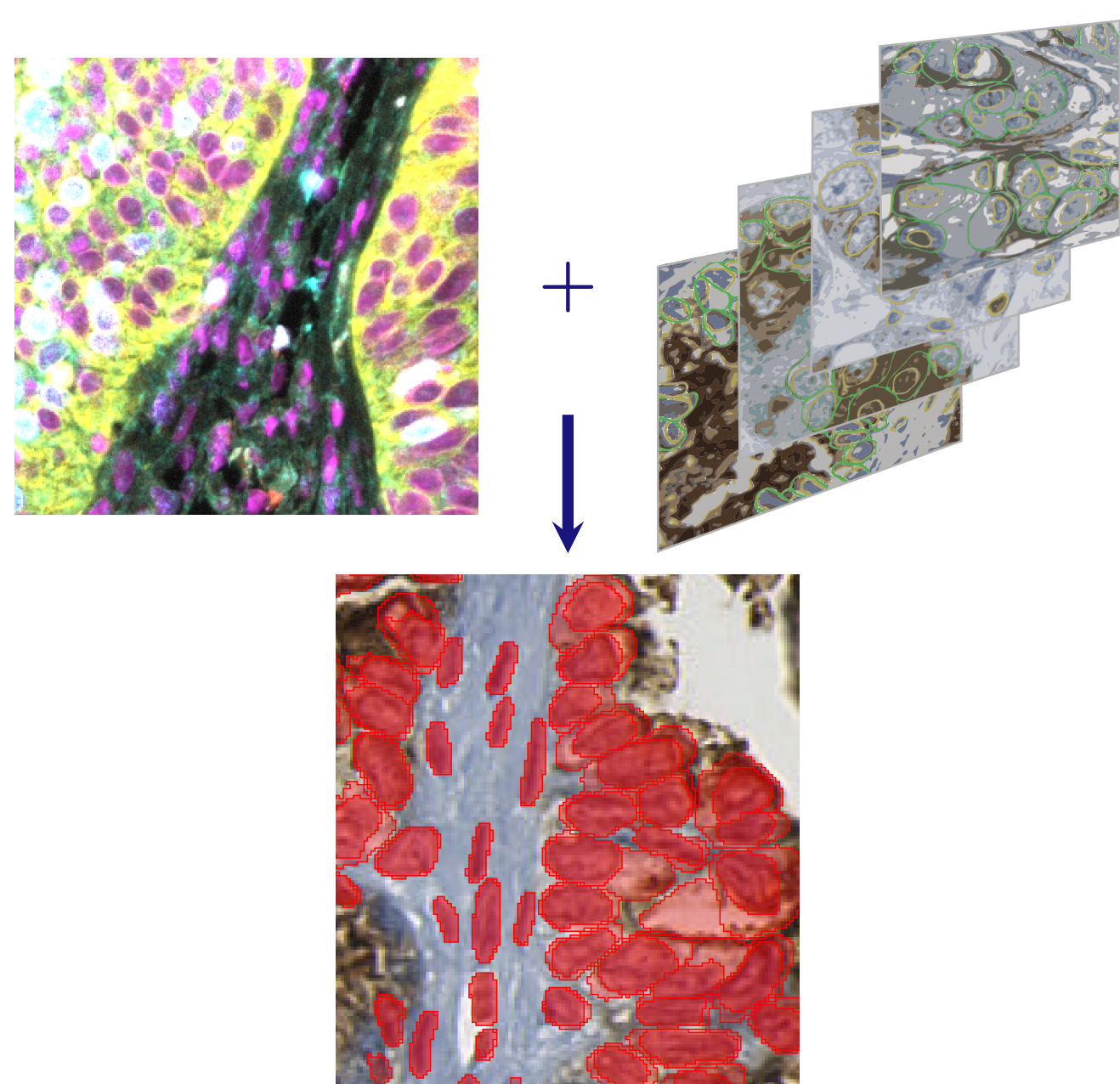


Beyond nuclei in IHC

To quantify membrane staining in ovarian tissue, we created annotations using QuPath, exported these with QuBaLab, to train InstanSeg (+ augmented fluorescence images).

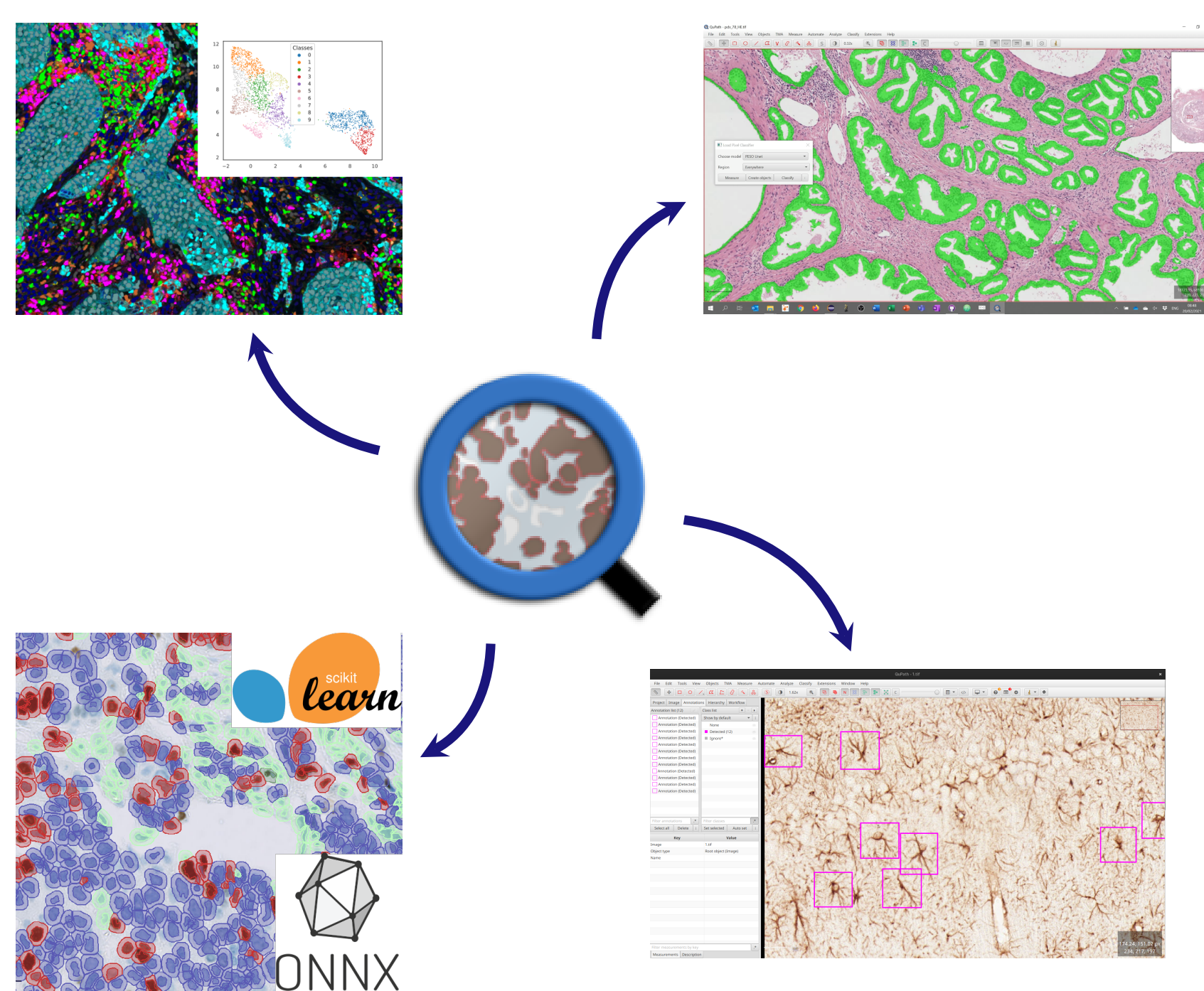
This enables H-scoring of membraneous stains in IHC imaging.

We are developing tools to facilitate generation of open ground truth datasets.



The future of ML in QuPath

- Clustering using SOM, UMAP, KNN
- U-Net semantic segmentation
- scikit-learn and ONNX object classification workflows
- Detection and segmentation of diverse objects using masks or just bounding boxes



Summary

InstanSeg is a powerful tool for nucleus and cell segmentation (and beyond!) in brightfield and fluorescence images.

QuPath and QuBaLab provide powerful tools for annotation and training of machine learning models.

We are training and deploying deep learning models in QuPath and developing tools so you can, too!

References

Goldsborough, Thibaut, Alan O'Callaghan, Fiona Inglis, et al. 'A Novel Channel Invariant Architecture for the Segmentation of Cells and Nuclei in Multiplexed Images Using InstanSeg'. Preprint, 8 September 2024. <https://doi.org/10.1101/2024.09.04.611150>.

Goldsborough, Thibaut, Ben Philps, Alan O'Callaghan, et al. 'InstanSeg: An Embedding-Based Instance Segmentation Algorithm Optimized for Accurate, Efficient and Portable Cell Segmentation'. arXiv:2408.15954. Preprint, arXiv, 28 August 2024. <https://doi.org/10.48550/arXiv.2408.15954>.

Aleynick, N., Li, Y., Xie, Y. *et al.* Cross-platform dataset of multiplex fluorescent cellular object image annotations. *Sci Data* **10**, 193 (2023). <https://doi.org/10.1038/s41597-023-02108-z>

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