

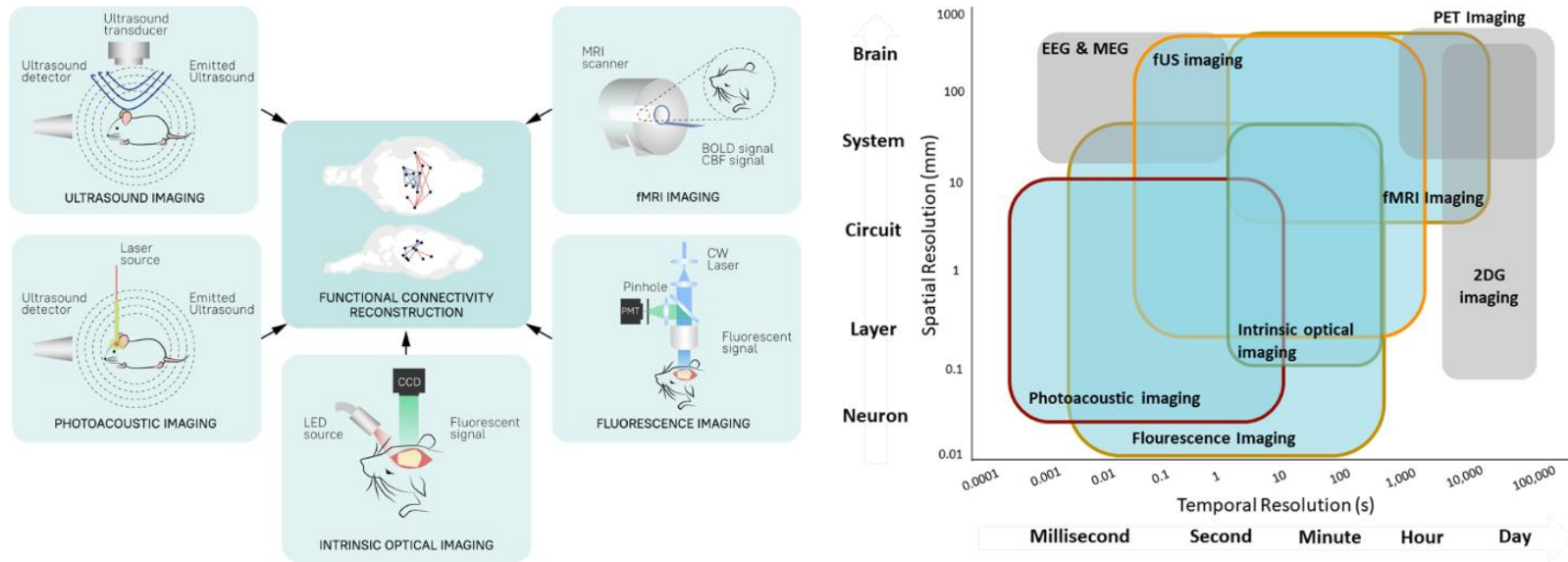


Workflows for 3D rodent brain imaging and registration

EPIC Online Technology Meeting on New
Advancements in Biomedical Imaging

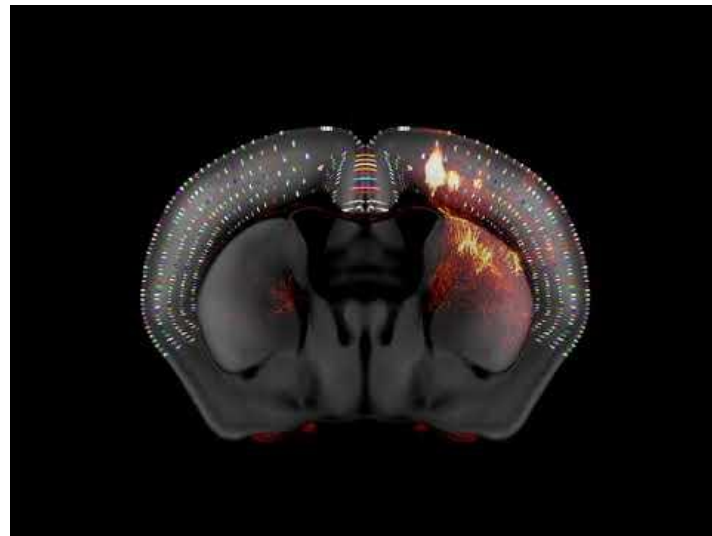
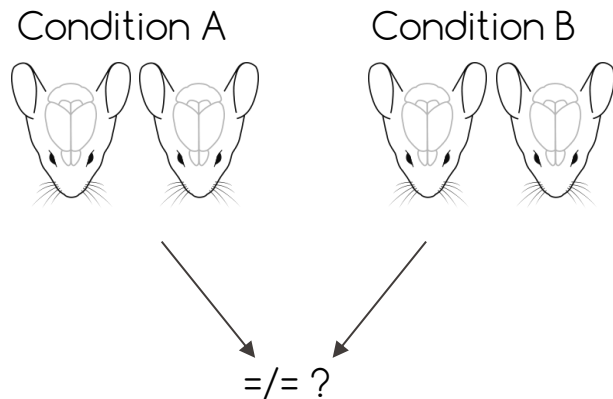
28.09.2026

Mouse brain imaging



Markicevic, M., Savvateev, I., Grimm, C. *et al.* Emerging imaging methods to study whole-brain function in rodent models. *Transl Psychiatry* **11**, 457 (2021). <https://doi.org/10.1038/s41398-021-01575-5>

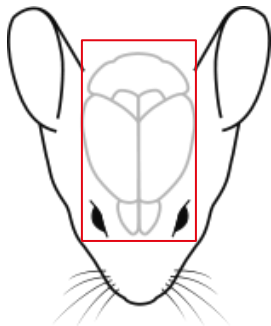
Comparing experimental data



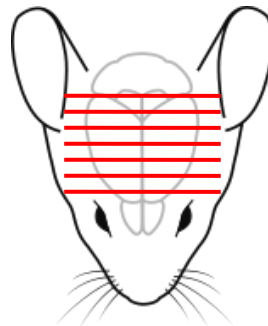
How to compare animals between conditions ?

- Align animal brain data in a common reference
- Allen Brain Common Coordinates Framework v3 (and regions)

3D Data alignment challenges



Aligning 3D data to atlas:
The overall brain structure is preserved

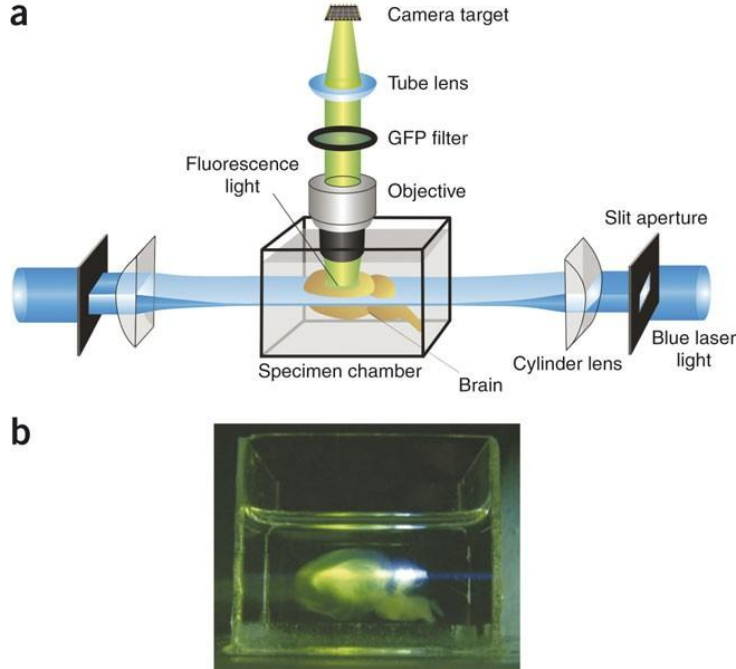


Aligning sections to atlas:
The brain is in many pieces.

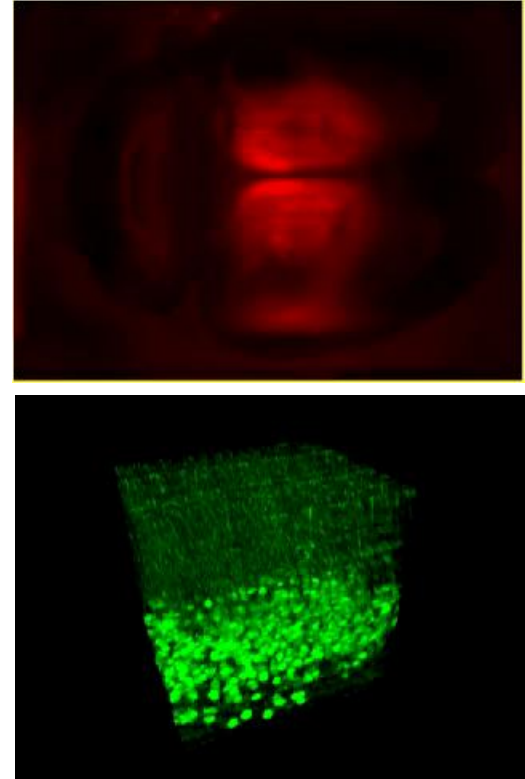


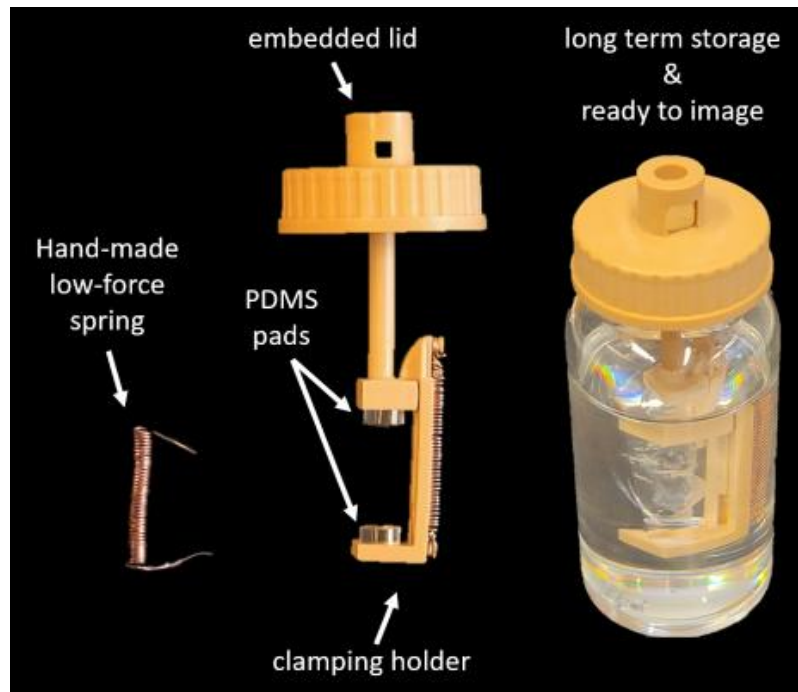
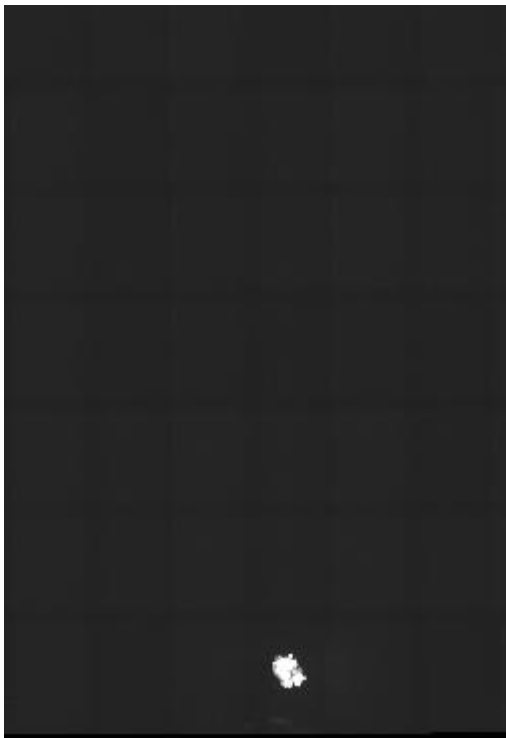
Workflow for 3D rodent whole-brain imaging and registration

Ultramicroscopy



Dodt, H. U., Leischner, U., Schierloh, A., Jährling, N., Mauch, C., Deininger, K., Deussing, J. M., Eder, M., Zieglgänsberger, W. and Becker, K. (2007). Ultramicroscopy: three-dimensional visualization of neuronal networks in the whole mouse brain. *Nat. Methods* **4, 331-336.**





Dynamic GUI (demo)

```
bigstitcher:
  xml_file:
  reader: fast
  fusion: fast
  channel_alignment:
    pairwise_shifts_downsamples:
      x: 4
      y: 4
      z: 2
    filter_min_r: 0.7
  tile_alignment:
    use_channel: 0
    pairwise_shifts_downsamples:
      x: 4
      y: 4
      z: 2
    filter_min_r: 0.7
  icp_refinement:
    icp_refinement_type: Simple (all together)
    downsampling: Downsampling 8/8/4
    interest: Average Threshold
    icp_max_error: Normal Adjustment (<5px)
  reorientation:
    raw_orientation: IPL
    reorient_to_asr: true # Choose false
  fusion:
    fusion_type: bigstitcher
```



Select your custom configuration

General **Bigstitcher** Brainreg Run

Xml File

Reader

Channel Alignment

Pairwise Shifts Downsamples

X

Y

Z

Filter Min R

Tile Alignment

Use Channel

Pairwise Shifts Downsamples

X

Y

Z

Filter Min R

Icp Refinement

Icp Refinement Type

Downsampling

Interest

Icp Max Error

Reorientation

Raw Orientation

Reorient To Asr ☒

Fusion

Fusion Type

Downsampling

Preserve Original

Produce

Fused Image

Fusion Method

Fuse Dir

A ligning

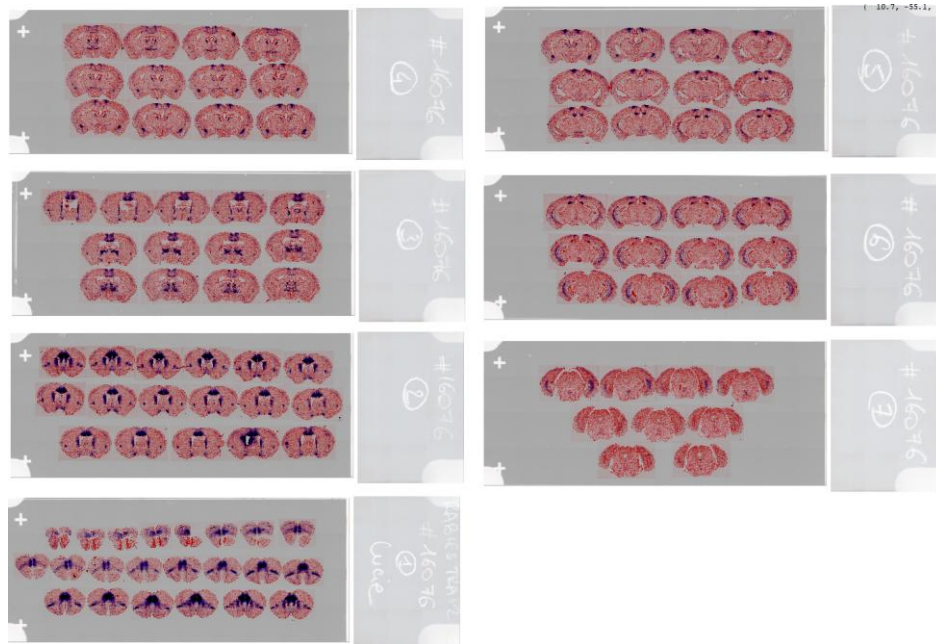
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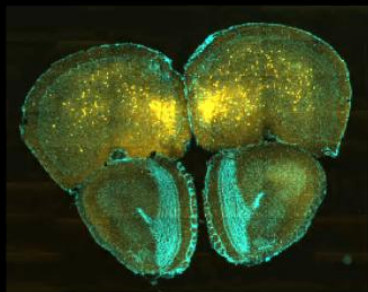
A tlases

Serial coronal sections

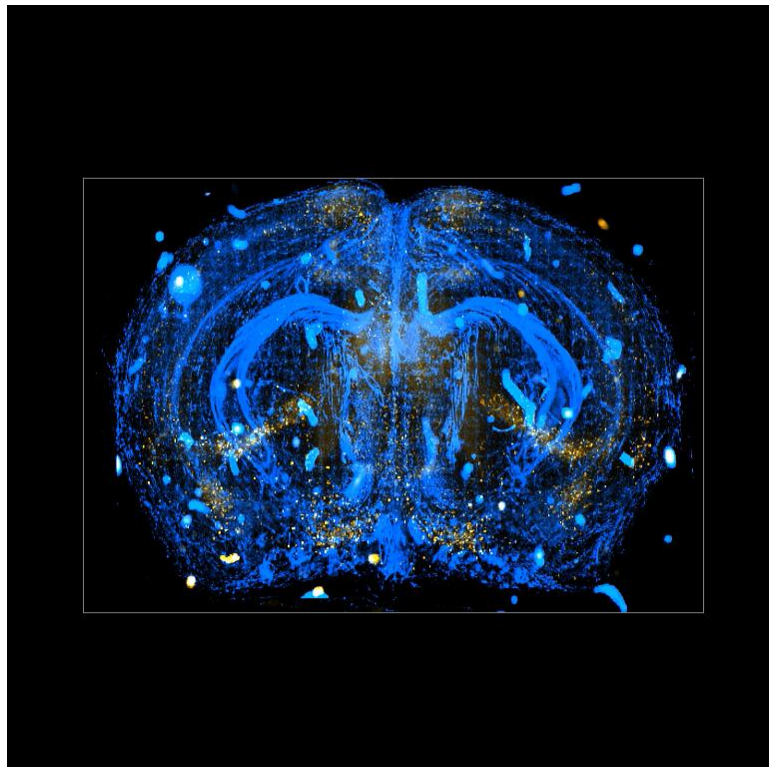
- Stack sections along the slicing axis in 3D
- Move (translate, rotate) each section to match the atlas in-plane
- Warp (with moderation) sections to match the atlas even better
- Detect and classify cells of interest
- Position detected cells within the atlas
- Allow for user interaction



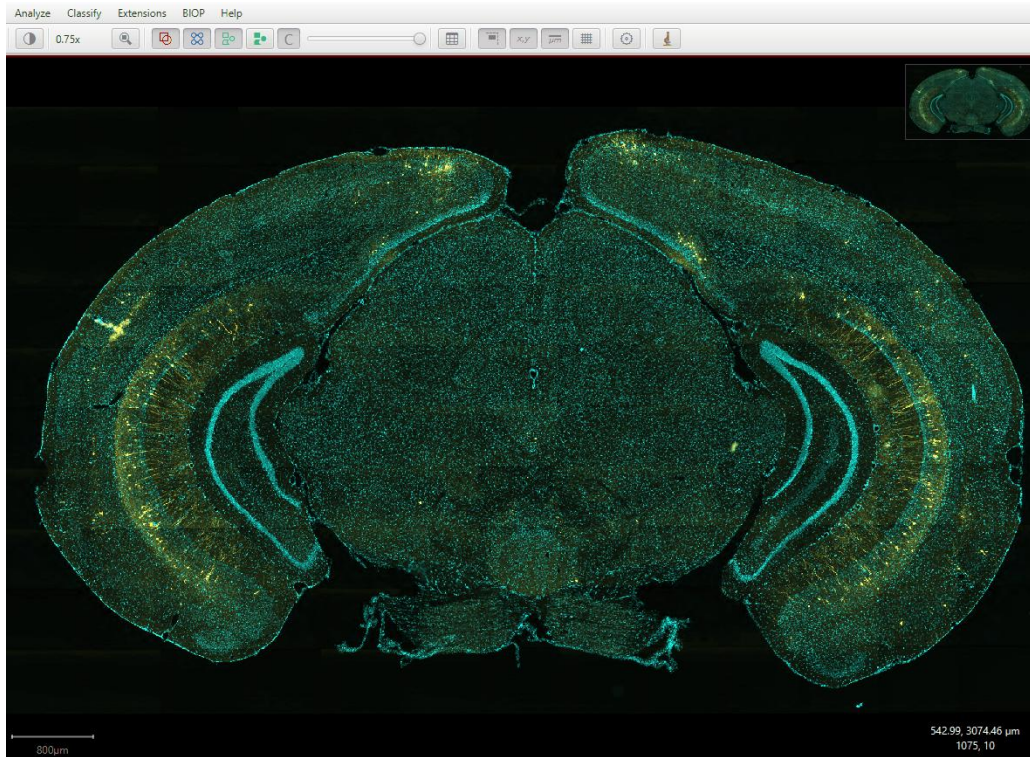
Visualizing aligned dataset in 2D



Visualizing aligned dataset in 3D



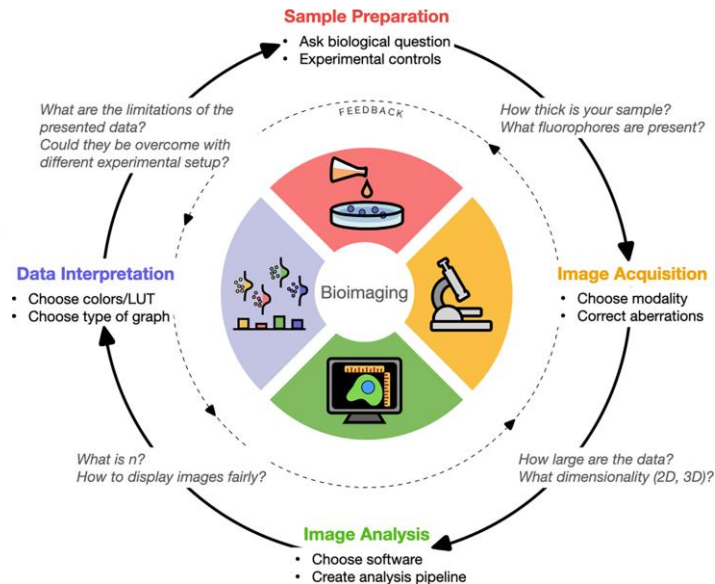
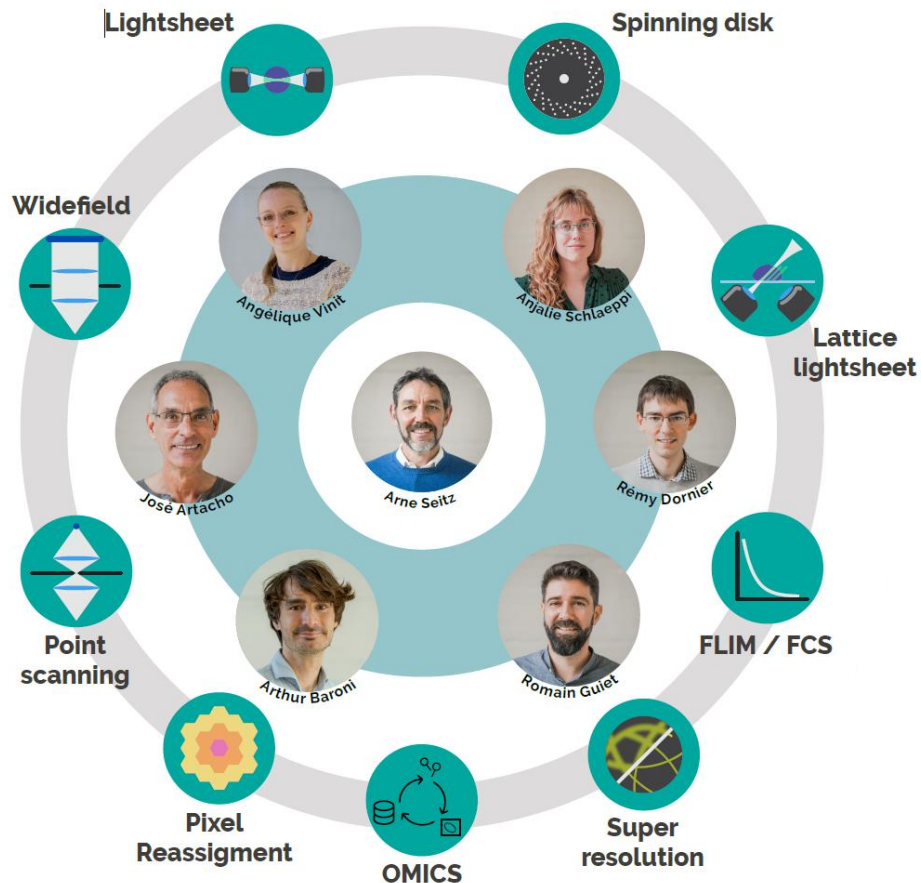
Importing results into QuPath



QuPath is the tool of choice for analyzing WSI dataset.

You can import the registered Regions in QuPath, and then **use your favorite scripts on a fully registered dataset.**

Biolmaging & Optics Platform



Senft RA, Diaz-Rohrer B, et al. (2023)

A biologist's guide to planning and performing quantitative bioimaging experiments.

PLoS Biol 21(6): e3002167. <https://doi.org/10.1371/journal.pbio.3002167>

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