
Computational Pathology

Challenges in the A/D conversion process

Gabriel Jimenez, Mehdi Ounissi, Daniel Racoceanu

Histopathology → Digital Pathology



Microscopic examination of a biopsy or surgical specimen by a pathologist

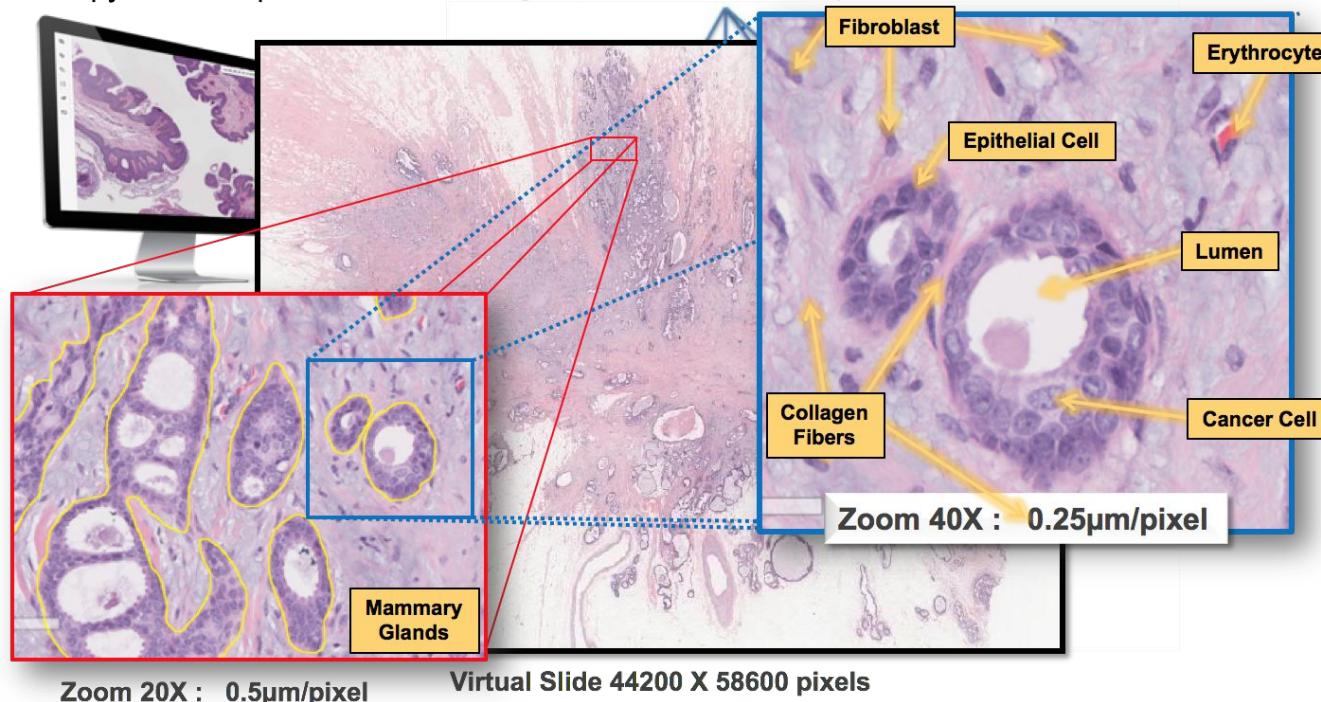


The management and interpretation of pathology information generated from a digitized glass slide

Digital Pathology → Computational Pathology

Whole Slide Imaging:

Tiled pyramidal representation → Image tiles on demand at a specific location and different resolutions



DICOM

DICOM is the International Standard for Medical Imaging and related information:

- images,
- waveforms,
- derived measurements and assessments,
- image presentation controls,
- and workflow management in the imaging department



Published as NEMA PS3 and as ISO 12052

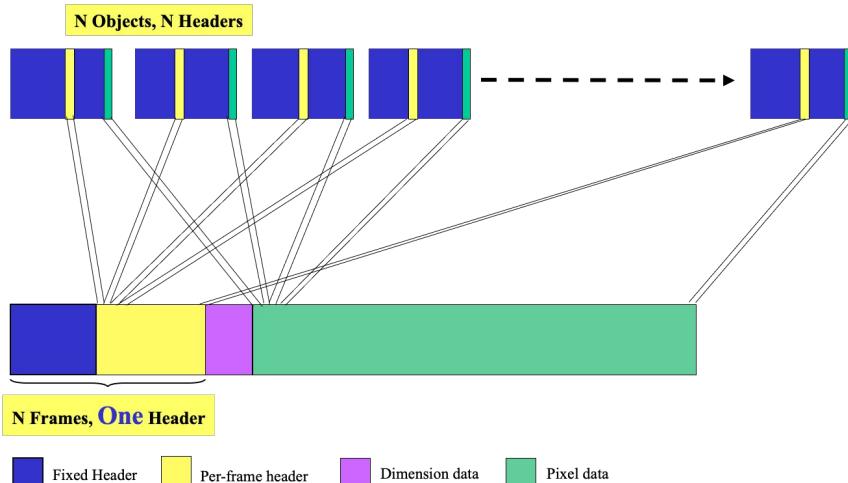
DICOM Images

Multiframe objects to support 1000+ slice studies

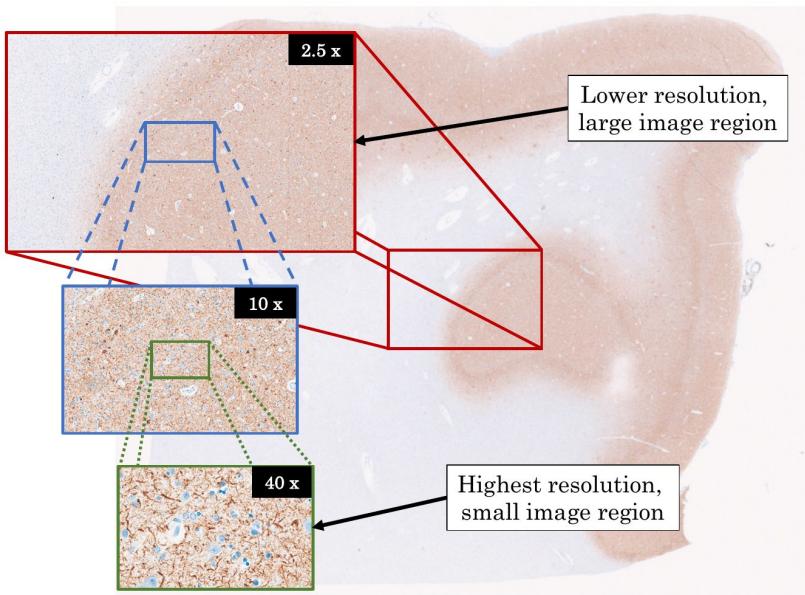
- Image header supports functional group attributes changing during acquisition
- Dimensions allow multiple views of data

Common structure used for all new image IODs

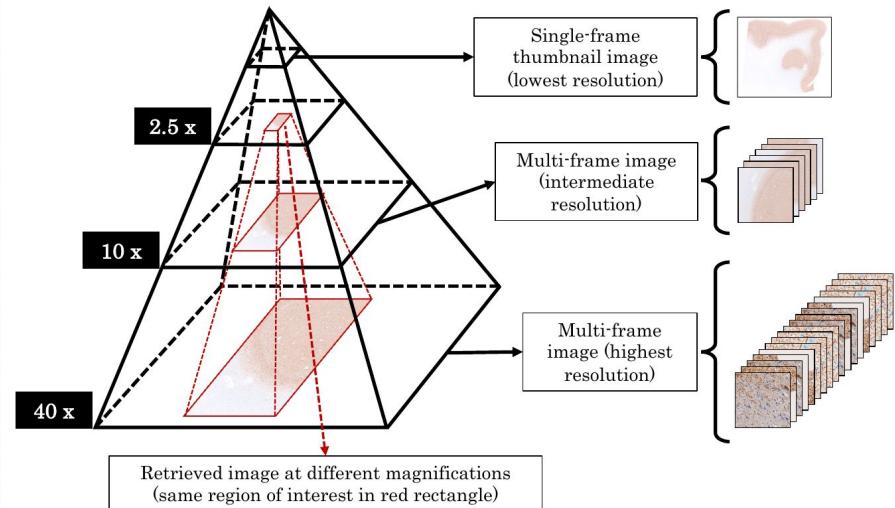
- Sup 141 Enhanced MR Color (First Read)
- Sup 139 Enhanced XA/XRF Informative Annex (Public Comment 2008)
- Sup 43 Enhanced Ultrasound (in Public Comment)
- Sup 125 Breast Tomosynthesis (Adopted August 2008)
- Sup 117 Enhanced PET (Adopted January 2008)
- Sup 110 Ophthalmic Coherence Tomography (Adopted August 2007)
- Sup 116 3-D X-ray (Adopted January 2007)
- Sup 83 Enhanced XA/XRF Image (Adopted 2004)
- Sup 58 Enhanced CT (Adopted 2003)
- Sup 49 Multiframe MR (Adopted 2001)
- **Sup 145 Whole Slide Microscopic Image (2010)**
- **Sup 122 Specimen Module and Revised Pathology (2008)**



Whole Slide Images



Rapid zooming issue when accessing lower resolution images: large amount of data need to be loaded into memory. In this example, the image size at the highest resolution (221 nm/pixels) is 82432×80640 pixels.



Pyramidal organization of Whole Slide Images. In this example, the image size at the highest resolution (221 nm/pixels) is 82432×80640 pixels. The compressed (JPEG) file size is 2.22 GB, whereas the uncompressed version is 18.57 GB.

*Jiménez, G., Racoceanu, D. (2023). Computational Pathology for Brain Disorders. In: Colliot, O. (eds) Machine Learning for Brain Disorders. Neuromethods, vol 197. Humana, New York, NY. https://doi.org/10.1007/978-1-0716-3195-9_18

Why is the standard not being used?

[J Pathol Inform](#). 2021; 12: 21.

Published online 2021 May 11. doi: [10.4103/jpi.jpi_88_20](https://doi.org/10.4103/jpi.jpi_88_20)

PMCID: PMC8274303

PMID: [34267986](https://pubmed.ncbi.nlm.nih.gov/34267986/)

Dicom_wsi: A Python Implementation for Converting Whole-Slide Images to Digital Imaging and Communications in Medicine Compliant Files

[Qiangqiang Gu](#),¹ [Naresh Prodduturi](#),¹ [Jun Jiang](#),¹ [Thomas J. Flotte](#),² and [Steven N. Hart](#)¹

[► Author information](#) [► Article notes](#) [► Copyright and License information](#) [Disclaimer](#)

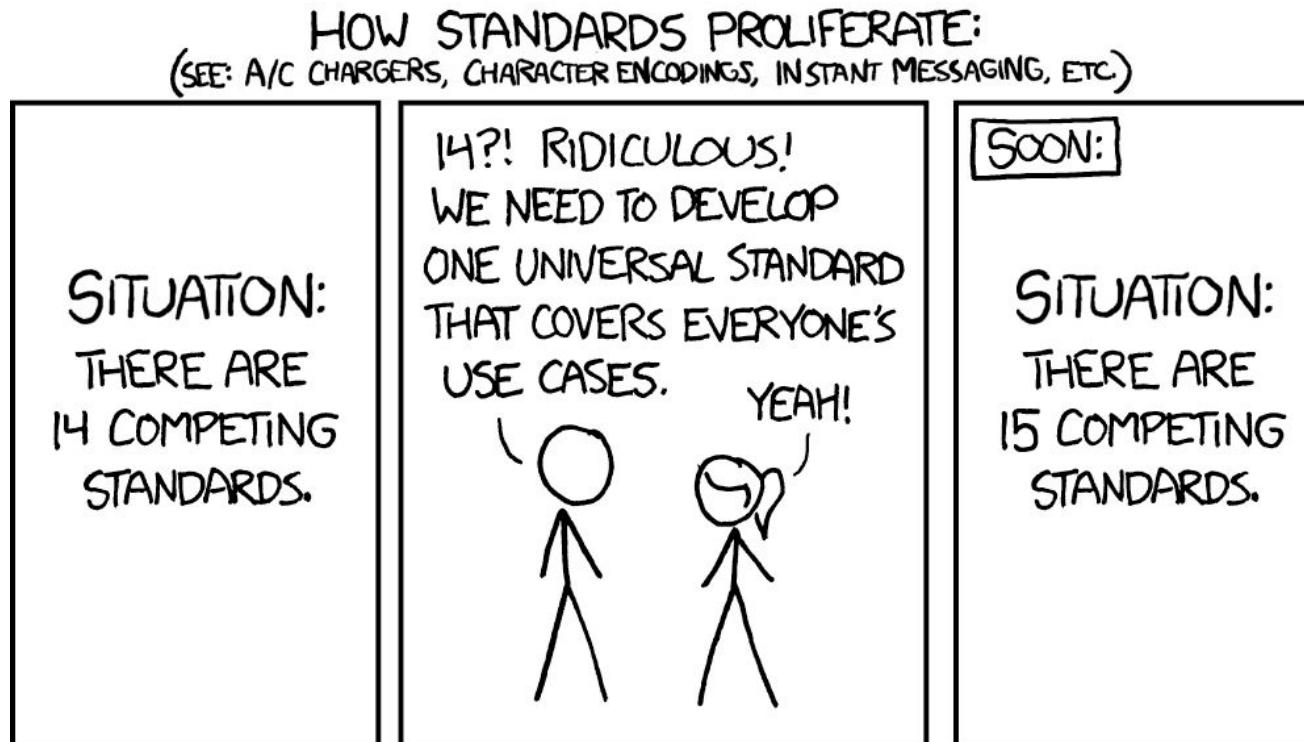
Abstract

Go to:

Background:

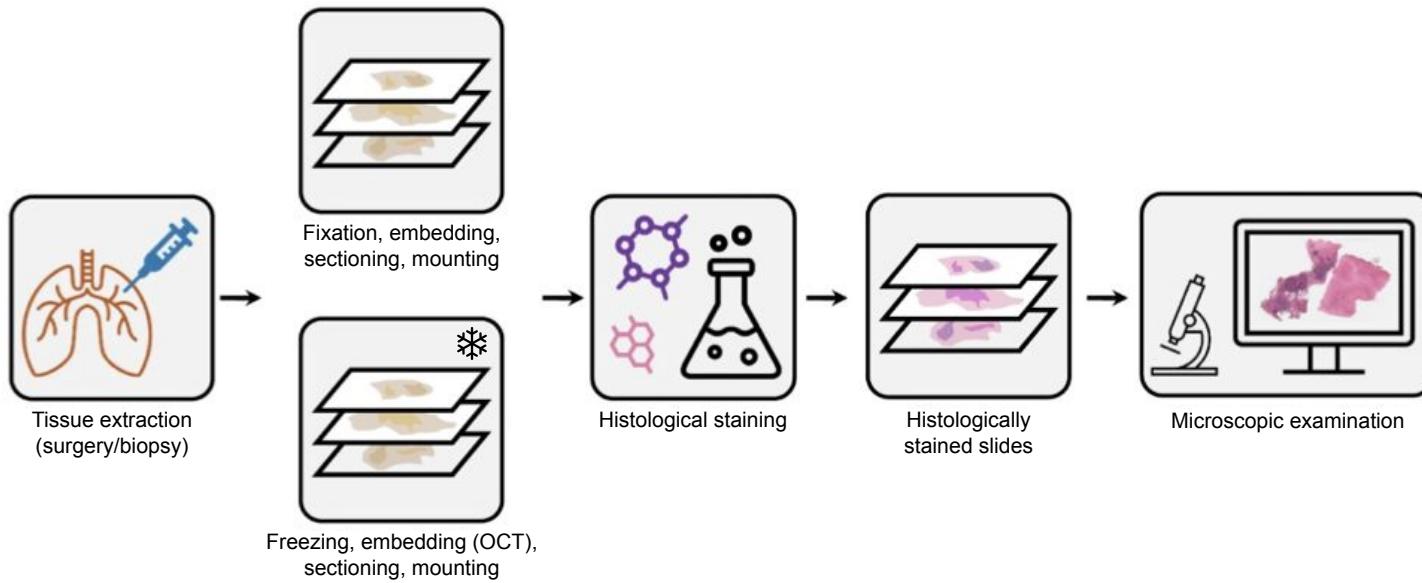
Adoption of the Digital Imaging and Communications in Medicine (DICOM) standard for whole slide images (WSIs) has been slow, despite significant time and effort by standards curators. One reason for the lack of adoption is that there are few tools which exist that can meet the requirements of WSIs, given an evolving ecosystem of best practices for implementation. Eventually, vendors will conform to the specification to ensure enterprise interoperability, but what about archived slides? Millions of slides have been scanned in various proprietary formats, many with examples of rare histologies. Our hypothesis is that if users and developers had access to easy to use tools for migrating proprietary formats to the open DICOM standard, then more tools would be developed as DICOM first implementations.

Why is the standard not being used?



<https://xkcd.com/927/>

The standard histological staining process



*Bai, B., Yang, X., Li, Y. et al. Deep learning-enabled virtual histological staining of biological samples. Light Sci Appl 12, 57 (2023)

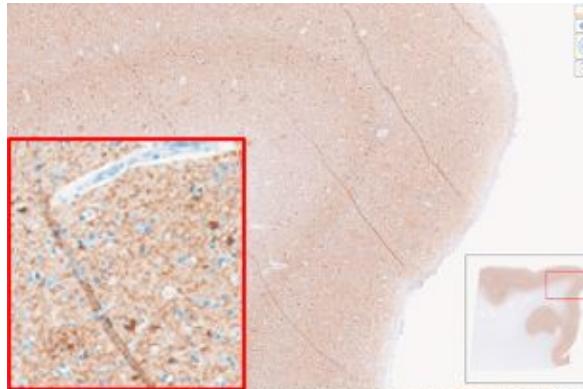
Mechanical and hardware related errors?



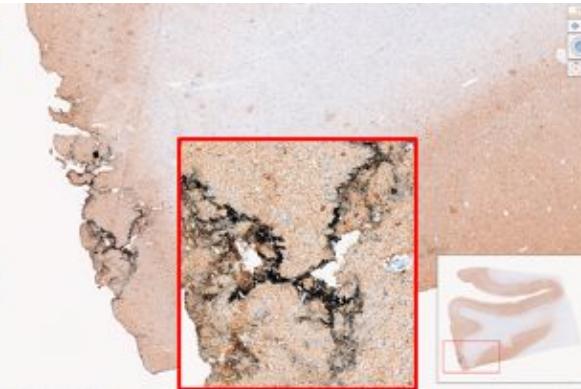
Every WSI scanner has six components: (a) a microscope with lens objectives, (b) light source (bright field and/or fluorescent), (c) robotics to load and move glass slides around, (d) one or more digital cameras for capture, (e) a computer, and (f) software to manipulate, manage, and view digital slides

Imaging artifacts

Folding artifact (floatation and mounting related artifact)



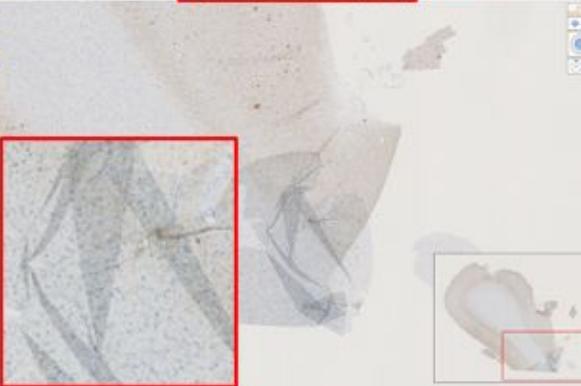
Marking fixation process (fixation artifact)



Breaking artifact (microtome-related artifact)



Overlaying tissue (mounting artifact).



*Jiménez, G., Racoceanu, D. (2023). Computational Pathology for Brain Disorders. In: Colliot, O. (eds) Machine Learning for Brain Disorders. Neuromethods, vol 197. Humana, New York, NY. https://doi.org/10.1007/978-1-0716-3195-9_18

Imaging artifacts

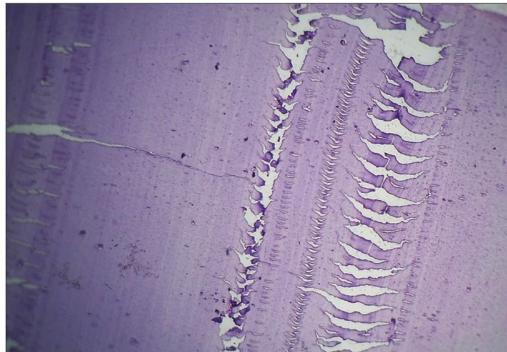


Figure 12: Histopathological image shows venetian blind artifact due to vibration of knife edge (H&E, $\times 10$)

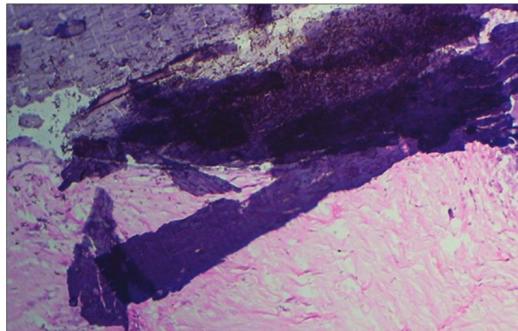


Figure 9: Histopathological image shows bone trabeculae stained strongly with hematoxylin due to incomplete decalcification (H&E, $\times 10$)

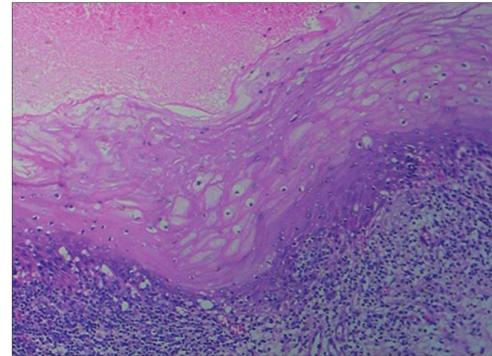


Figure 7: Histopathological image shows tissue autolysis due to delayed fixation (H&E, $\times 10$)

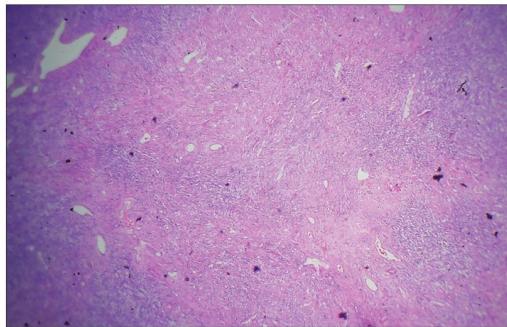


Figure 17: Histopathological image shows stain deposit due to formation of fluorescent sheen in hematoxylin solution (H&E, $\times 10$)

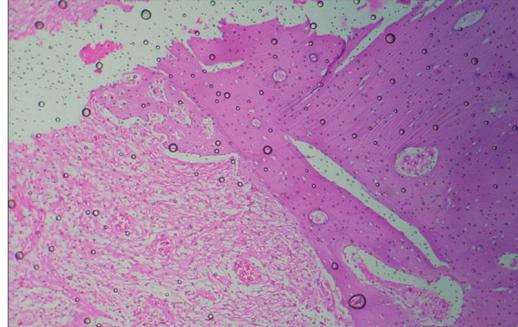


Figure 18: Histopathological image shows air bubbles formed during mounting procedure (H&E, $\times 10$)



Figure 16: Histopathological image shows wrinkles and folds due to uneven stretching of tissue sections (H&E, $\times 10$)

*Taqi, Syed Ahmed, Syed Abdus Sami, Lateef Begum Sami, and Syed Ahmed Zaki. 2018. "A Review of Artifacts in Histopathology." Journal of Oral and Maxillofacial Pathology: JOMFP 22 (2): 279.

StratiflAD project

Interpretable Deep Learning in Computational Histopathology for Alzheimer Disease Patient Stratification Refinement

Gabriel Jimenez, Mehdi Ounissi, Daniel Racoceanu
Pathology department: Lev Stimmer, Benoit Delatour (ICM)

• Understanding Alzheimer's Disease

Alzheimer disease is caused by

- Misfolding and accumulation of Amyloid-beta peptides and **tau proteins**.

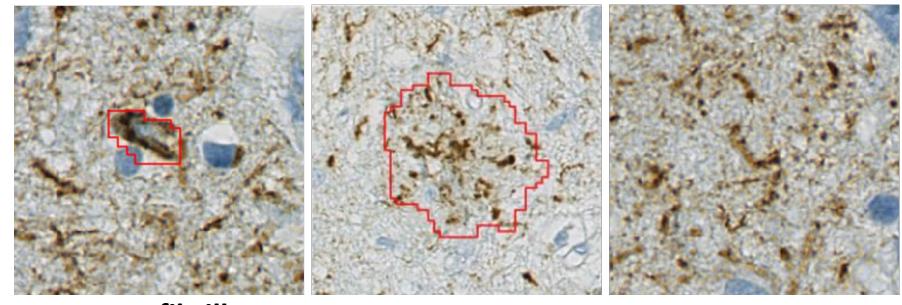
Findings by Duyckaerts, et al.*:

- Tau pathologies (tauopathies) are highly linked to clinical manifestations of AD.

The problem:

- The clinical presentation of the patients is very heterogeneous.
- In vivo imaging methodologies are not able to reach the resolution obtained by microscopy.

Tau proteins (located in neurons)



a. Neurofibrillary tangle

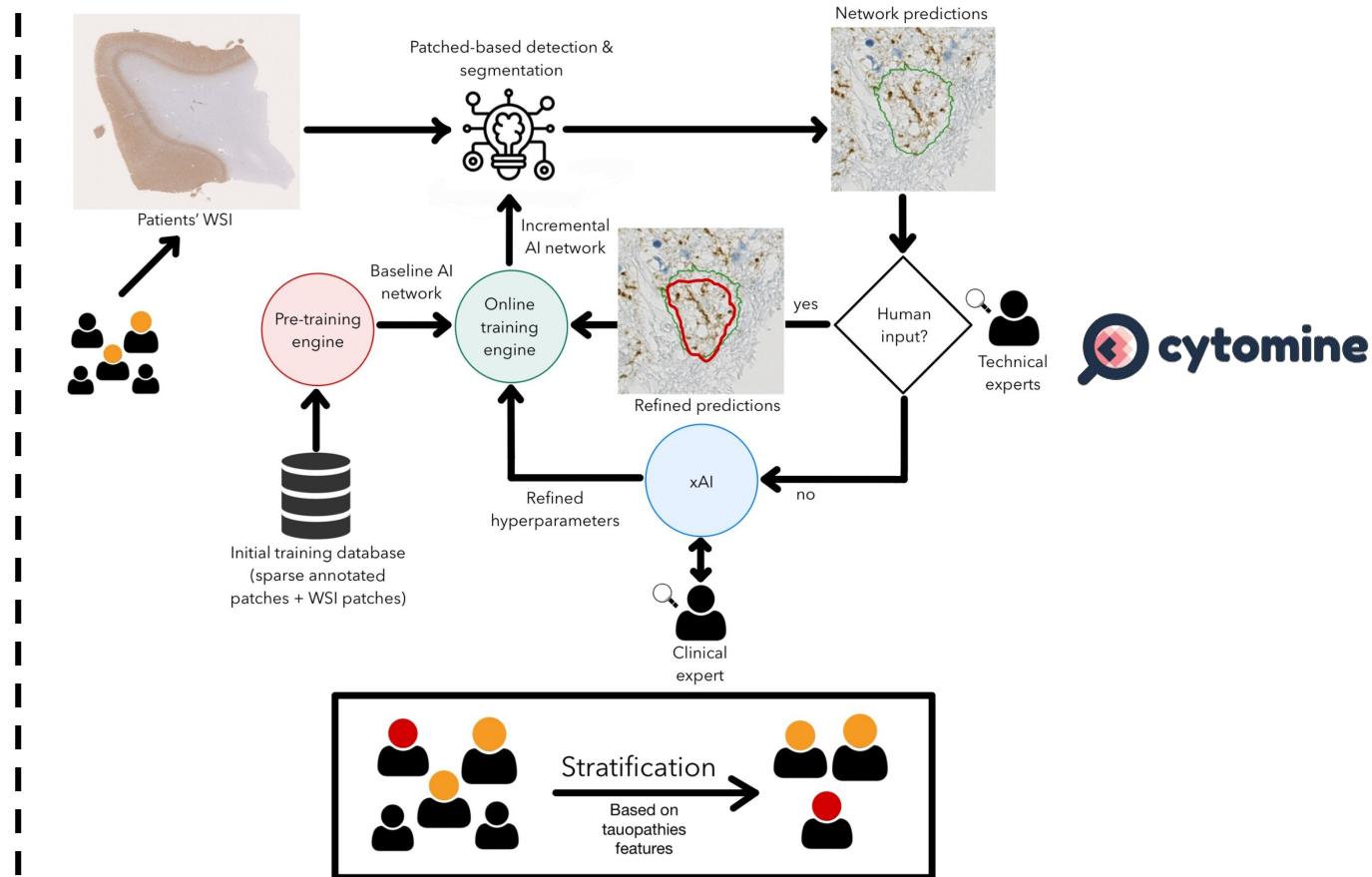
b. Neuritic Plaque

c. Dystrophic Neurites

*Duyckaerts C, Delatour B, Potier MC. Classification and basic pathology of Alzheimer disease. Acta Neuropathol. 2009 Jul;118(1):5-36.

The idea : Annotation Refinement Tool

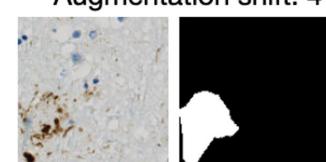
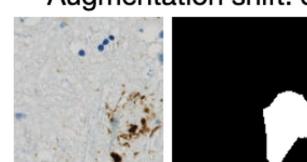
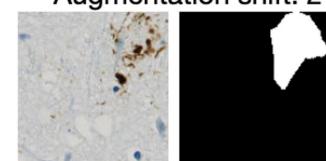
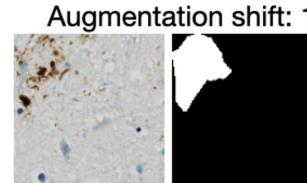
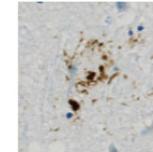
- Automatic **segmentation/detection** of tau protein aggregates from WSI.
- Manual annotation refinement using DL attention mechanisms and visual explainability features from DL models.



- The datasets

Dataset #1*	Dataset #2
6 WSI	8 WSI
NanoZoomer 2.0-RS	NanoZoomer 2.0-RS & S60
ALZ50 antibodies	AT8 antibodies
Manual annotation	Manual annotation
BRS & EDS	ROI-guided sampling
Plaques (NP)	Plaques (NP)
128x128	128x128 & 256x256
20x (plaques) 227 nm/px @ 40x	20x (plaques) 221 nm/px @ 40x

Original image patch and ground truth mask



*Manouskova , K., Abadie, V., Ounissi, M., Jimenez, G., Stimmer, L., Delatour, B., Durrleman, S., Racoceanu, D. (2022) Tau protein discrete aggregates in Alzheimer's disease: neuritic plaques and tangles detection and segmentation using computational histopathology, SPIE Medical Imaging, San Diego, USA

• The networks

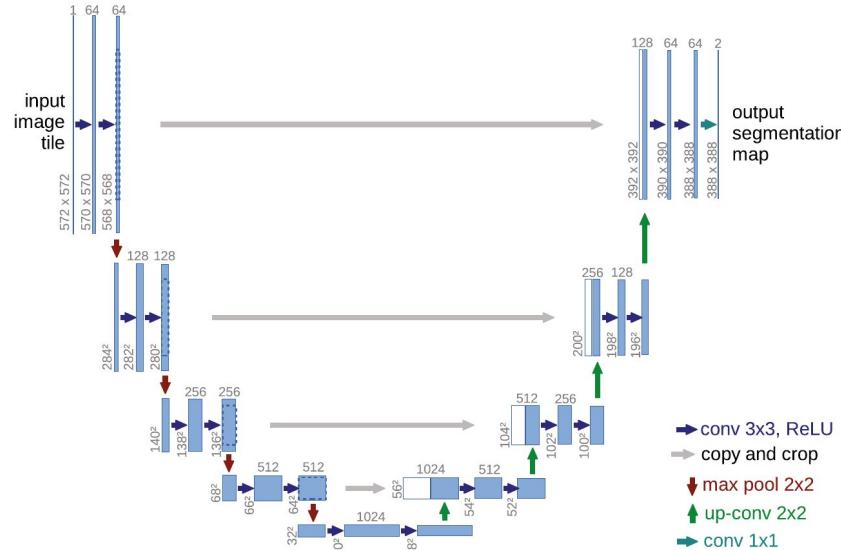


Fig. 1. U-net architecture (example for 32x32 pixels in the lowest resolution). Each blue box corresponds to a multi-channel feature map. The number of channels is denoted on top of the box. The x-y-size is provided at the lower left edge of the box. White boxes represent copied feature maps. The arrows denote the different operations.

*Olaf Ronneberger, et al. U-Net: Convolutional Networks for Biomedical Image Segmentation. MICCAI, 2015

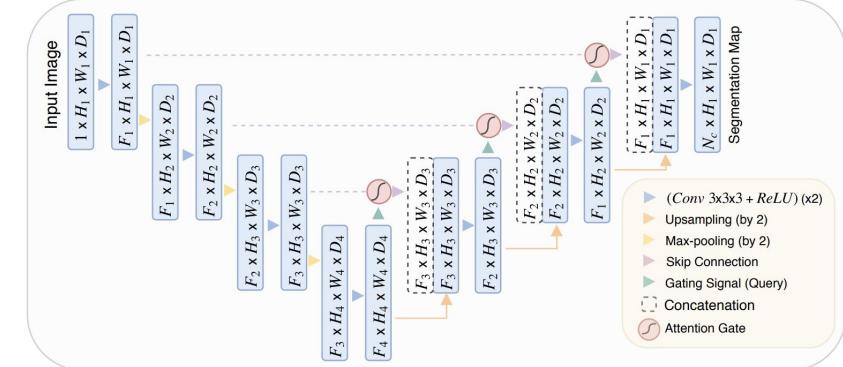


Figure 1: A block diagram of the proposed Attention U-Net segmentation model. Input image is progressively filtered and downsampled by factor of 2 at each scale in the encoding part of the network (e.g. $H_4 = H_1/8$). N_c denotes the number of classes. Attention gates (AGs) filter the features propagated through the skip connections. Schematic of the AGs is shown in Figure 2. Feature selectivity in AGs is achieved by use of contextual information (gating) extracted in coarser scales.

*Ozan Oktay, et al. Attention U-Net: Learning Where to Look for the Pancreas. MIDL, 2018

Results

- Dataset #1: detection and segmentation. Architecture: UNet*

Task	Normalization	F1 (cross-val)	F1 (cross-test)
Det.	Macenko	99.8 %	81.3 %
Seg.	Vahadane LS	81.5 %	78.2 %

- Dataset #2 (8 WSI): segmentation results.

Architecture: UNet

Patch size	Normalization	F1 (cross-val)	F1 (cross-test)
128x128	Macenko	$69.5 \pm 2.9\%$	$68.5 \pm 2.6\%$
256x256	Macenko	$66.0 \pm 4.2\%$	$64.6 \pm 3.3\%$

Architecture: Attention UNet

Patch size	Normalization	F1 (cross-val)	F1 (cross-test)
128x128	Macenko	$75.2 \pm 3.3\%$	$69.20 \pm 2.5\%$
256x256	Macenko	$69.31 \pm 4.5\%$	$63.42 \pm 3.0\%$

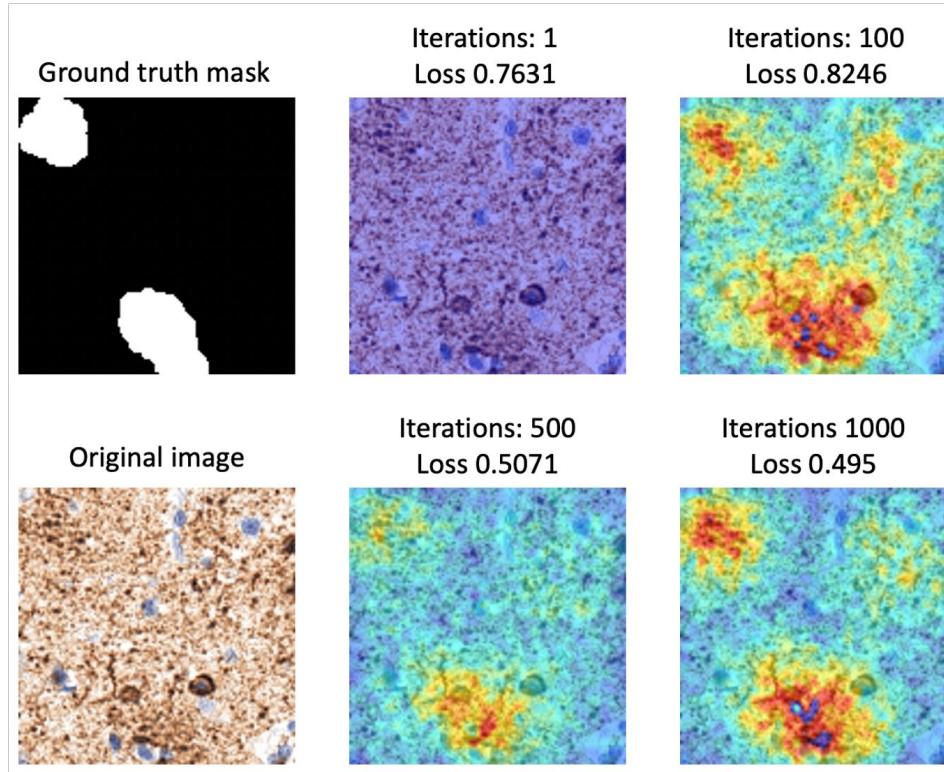
- Dataset #2 (8 WSI): analysis different resolution and normalization. Architecture: UNet.

Scanner	Patch size	Normalization	F1 (cross-test)
NanoZoomer 2.0-RS	128x128	Macenko	$73.4 \pm 0.6\%$
NanoZoomer S60	128x128	Macenko	$63.5 \pm 2.4\%$

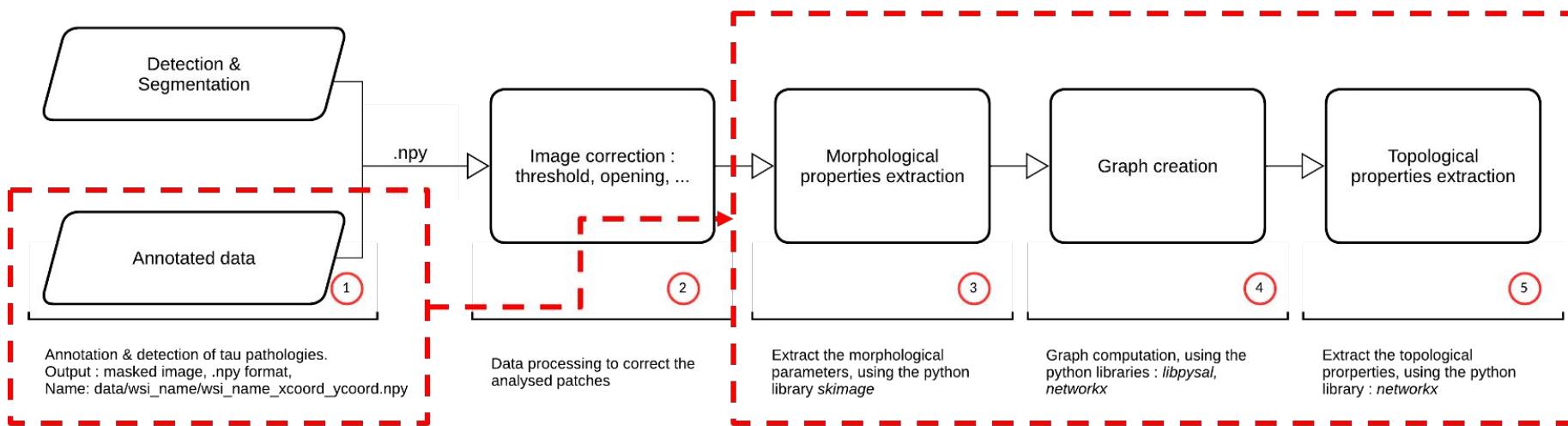
Normalization	Patch size	F1 (cross-test)
Macenko	128x128	72.5 %
Vahadane	128x128	70.9 %

*Jimenez, G. et al. (2022). Visual Deep Learning-Based Explanation for Neuritic Plaques Segmentation in Alzheimer's Disease Using Weakly Annotated Whole Slide Histopathological Images. In: Wang, L., Dou, Q., Fletcher, P.T., Speidel, S., Li, S. (eds) Medical Image Computing and Computer Assisted Intervention – MICCAI 2022. Lecture Notes in Computer Science, vol 13432. Springer, Cham. https://doi.org/10.1007/978-3-031-16434-7_33

- Visual explanation properties of DL model → improving annotations

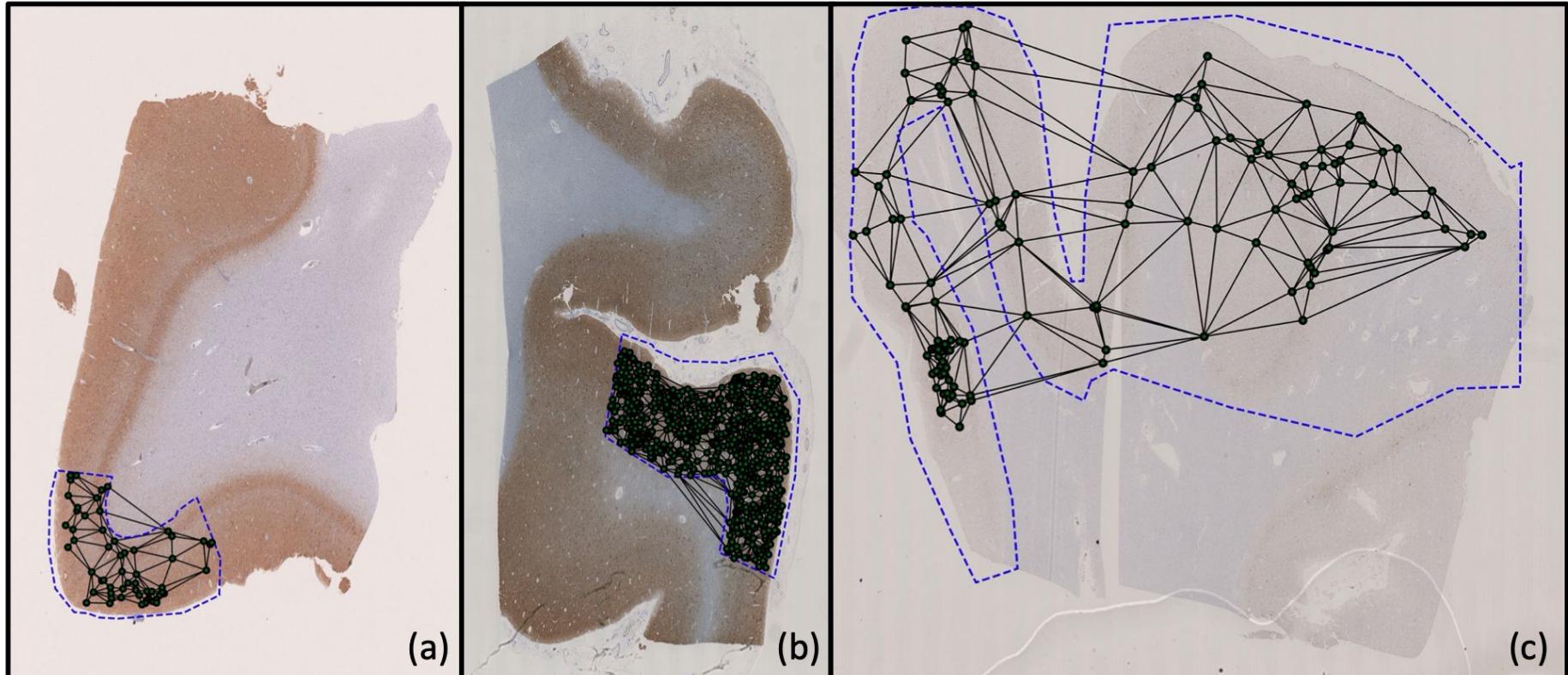


- What about global information of the WSI?

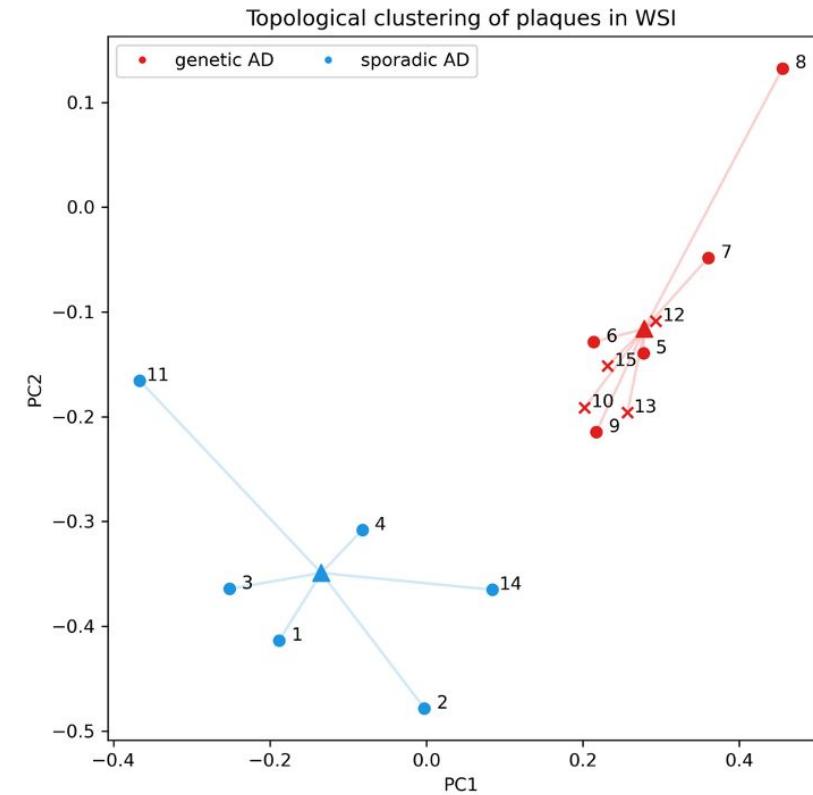
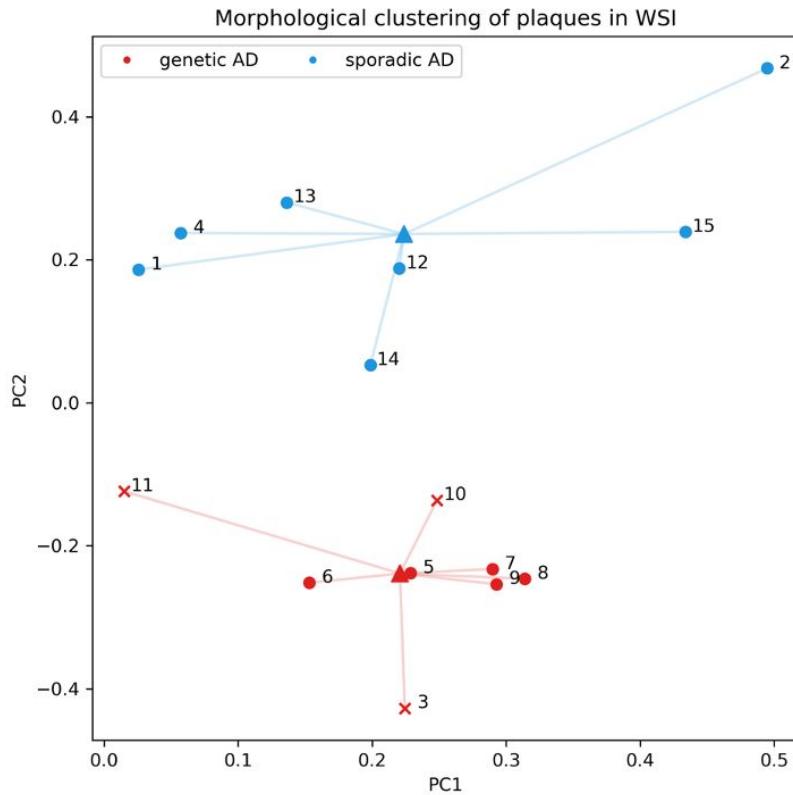


Results

- Graph creation: 1 WSI per patient → 1 graph per patient



- Clustering using morphological and topological features



Results

- Tangle and plaques in layers of the brain

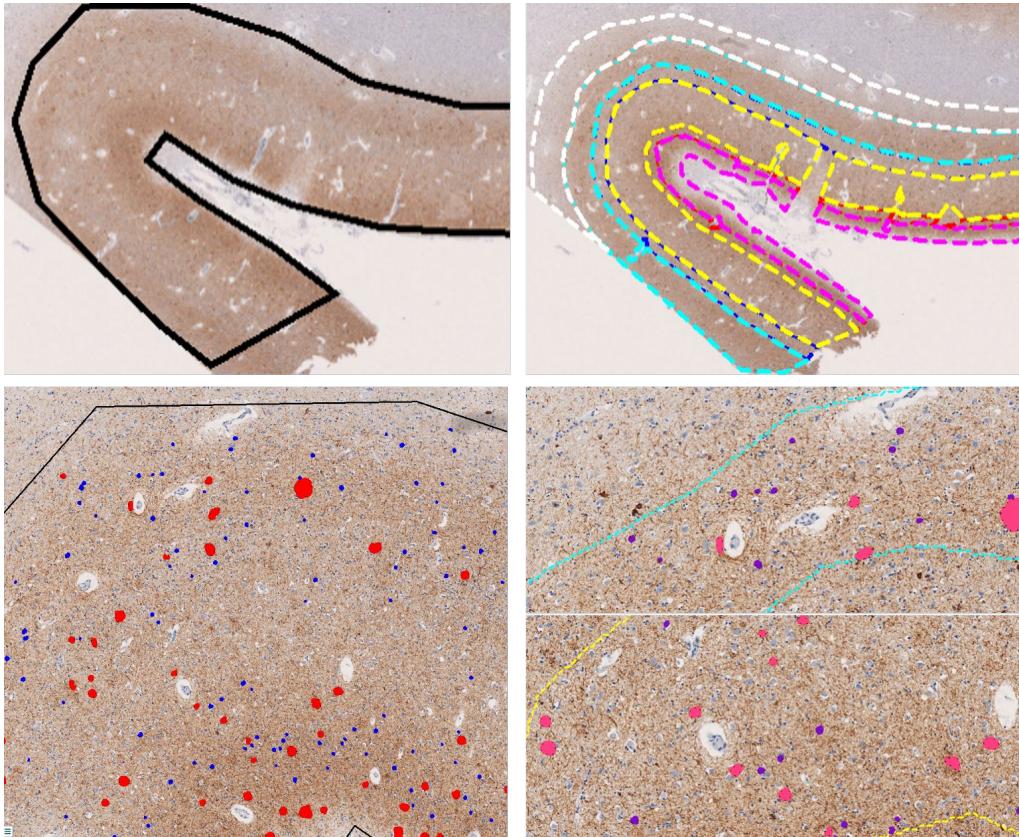


Fig. Example of a WSI and its annotations on the Region of Interest (ROI) on the left and the 6 layers delimitation on the right. **Plaques** are shown in a **red** mask, and **tangles** in a **blue** mask.

Results

Fig. Comparison of clustering method for raw data points and GNN-based embeddings. The left graph presents clustered nodes based on their **spatial coordinates**. In contrast, the right graph leverages **GNN embeddings** to cluster nodes.

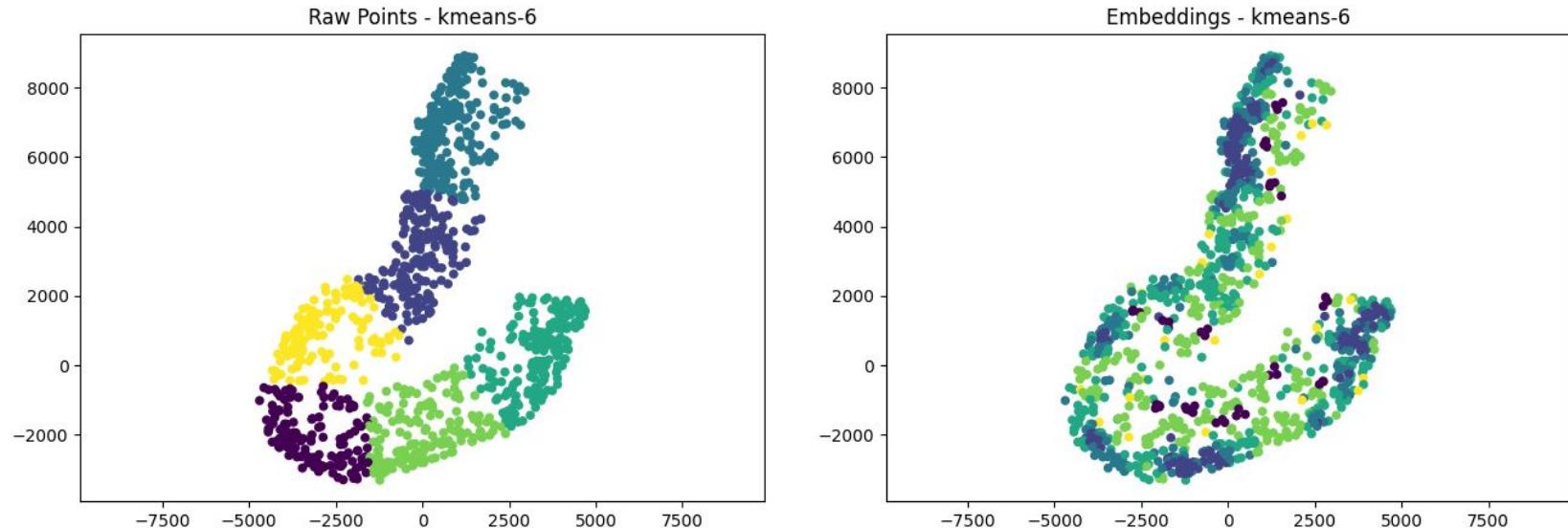


Table. Cross-validation accuracy and standard deviation of the RF classification between cAD and rpAD classes for plaques and tangles layers.

Layers	1	2	3	4	5	6
Plaques	0.62 ± 0.23	0.72 ± 0.18	0.83 ± 0.16	0.67 ± 0.16	0.62 ± 0.26	0.61 ± 0.23
Tangles	0.57 ± 0.16	0.55 ± 0.29	0.64 ± 0.25	0.72 ± 0.23	0.65 ± 0.22	0.65 ± 0.25

*Jimenez, G. et al. Unravelling the Topographical Organization of Brain Lesions in Variants of Alzheimer's Disease Progression. MICCAI 2024 (rev.)

- **The key ideas from the study**

1. Understanding the impact of WSI acquisition procedures in DL models.
2. Graphs can integrate local and global information of WSI.
3. GNN and GNN embeddings are a powerful tool to deeply analyze patterns of tangles and plaques present in brain tissue layers.

Acknowledgement

This research is supported by Mr Jean-Paul Baudecroux and The Big Brain Theory Program - Paris Brain Institute (ICM).

The human samples were obtained from the Neuro-CEB brain bank (BRIF Number 0033-00011), partly funded by the patients' associations ARSEP, ARSLA, "Connaître les Syndromes Cérébelleux", France-DFT, France Parkinson and by Vaincre Alzheimer Foundation, to which we express our gratitude. We are also grateful to the patients and their families.

Contact:

Gabriel JIMENEZ, Doctorant, Sorbonne University, Paris, France
Paris Brain Institute (ICM – Inserm / CNRS / AP-HP / Sorbonne), Inria team “Aramis”
gabriel.jimenez@icm-institute.org