Virtual Platform for multimodal data analysis

Immuno Histology vs. Fluorescence as diagnostic aid ?

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Research Engineer
Imagopole (S. Shorte)
Institut Pasteur

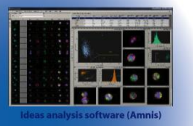
Context

High Content Screening (HCS)

Imaging Cytometry

Cells in suspension


ImageStream (Amnis)


Ideas analysis software (Amnis)

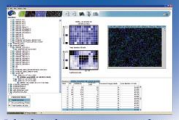
Automated Image Analysis on a Per-Cell basis


diamid treated erythrocytes

Multi-well Plates


Opera (Perkin Elmer)

High Content Analysis (HCA)

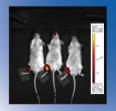

Columbus: data management software (Perkin Elmer)

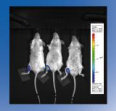
In Vivo Optical Analysis

Whole Animal


IVIS100 (Caliper)

Correlative Analysis


Fluorescence


Bioluminescence

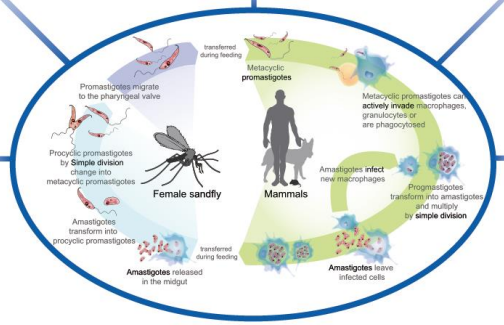
Cytometry

Cell Phenotype


Gallios (Beckman Coulter)


Kaluza analysis software (Beckman Coulter)

Cell population Analysis by Scatter Plots

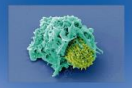


Electronic microscopy

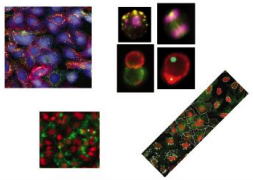
Ultra Structural Level


MEB JSM 6700F (Jeol)

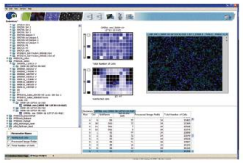
High Resolution Imaging


Macrophage and Lymphocytes infected by HIV

Quantification

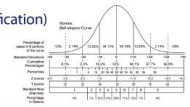


Analysis by readout model

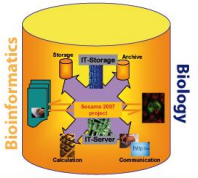


Validation & Statistics

- b Correlation analysis (Realtionship between HCA readouts)
- b Normalisation
- b Multivariate Analysis
- b Cluster analysis of readouts (Classification)
- b Hit/Event selection Procedure



Data Storage & Management



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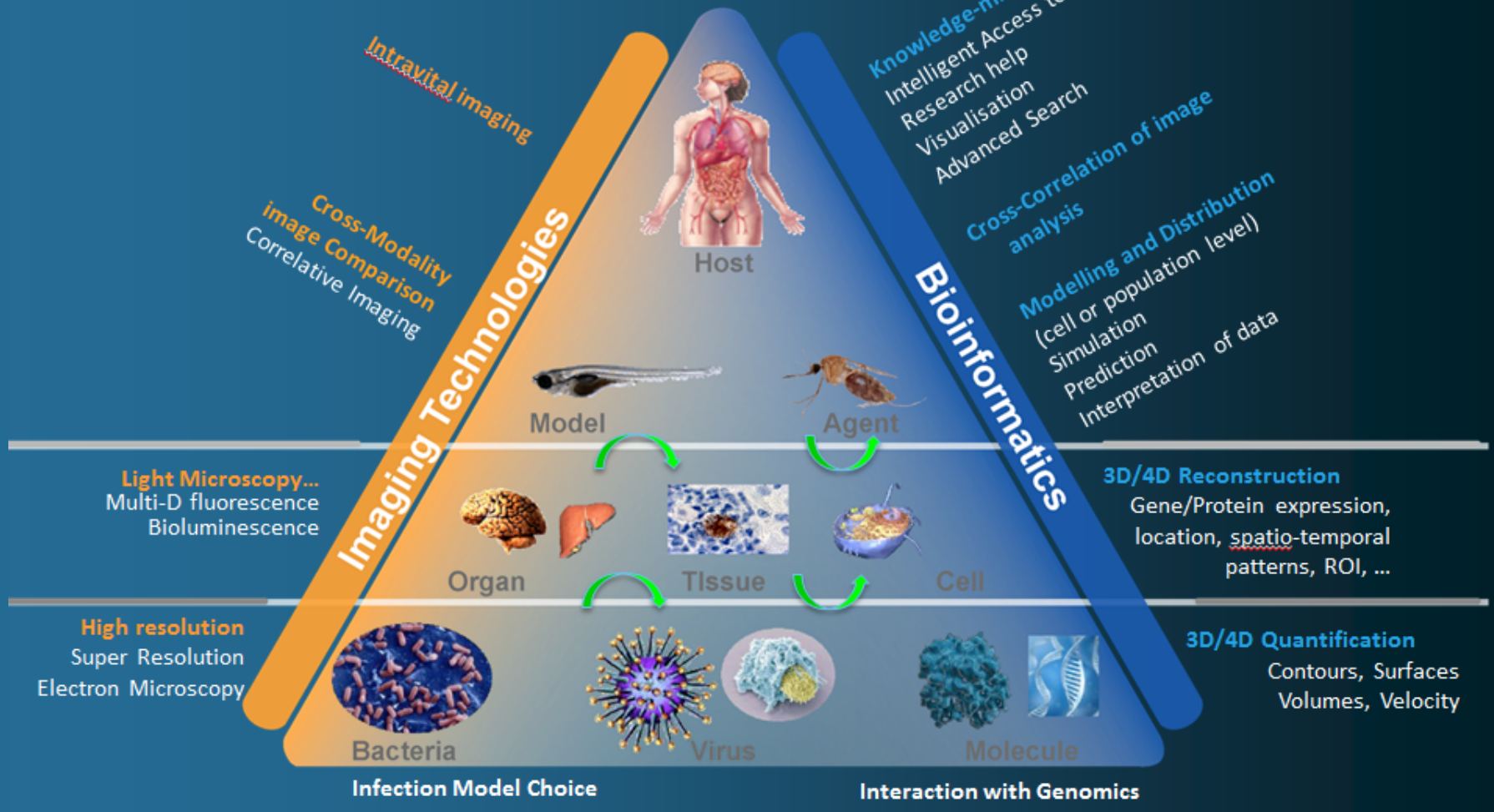
Context


Intravital imaging
Cross-Modality
image Comparison
Correlative Imaging
Imaging Technologies
Light Microscopy…
Multi-D fluorescence
Bioluminescence
High resolution
Super Resolution
Electron Microscopy
Host
Model
Agent
Organ
TIssue
Cell
Bacteria
Virus
Molecule
Bioinformatics
Knowledge-mining tool
Intelligent Access to data
Research help
Visualisation
Advanced Search
Cross-Correlation of image analysis
Modelling and Distribution
(cell or population level)
Simulation
Prediction
Interpretation of data
3D/4D Reconstruction
Gene/Protein expression,
location, spatio-temporal
patterns, ROI, …
3D/4D Quantification
Contours, Surfaces
Volumes, Velocity
Infection Model Choice
Interaction with Genomics

Context

Automatic Multimodal Analysis

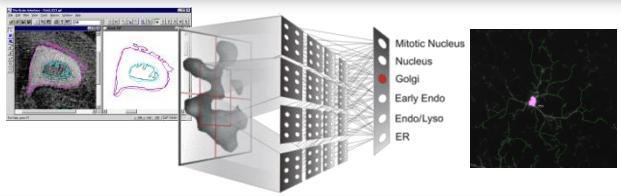
Sample

Treatment

Technology

IMAGES

Analysis



Information Extraction

Pattern Classification

Morphology/ Phenotype

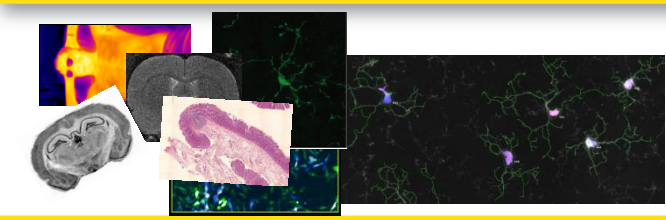
Validation

Multivariate

Hit/event Distribution

Representativeness

Visualisation



Pre diagnostic Score

Cartography

Scientific Hypothesis

Scientific Interpretation

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PTR391 Institut Pasteur 2012-2014

lead by Dr E. Labruyere, IP

Real time imaging of connective tissues to analyse the molecular mechanisms connecting fibrillar collagen remodelling, inflammation and cell migration

Imagopole implication (WP4)

1. Detection of SHG Signal in mucosa on embedded human colon biopsies
2. Comparaison of the scaffold organization in healthy and pathological colonic tissue

Computer-assisted characterization of the collagen fiber architecture on colonic tissue through the SHG signal.

Collaboration

Pathological samples choice and expertise:

- Human pathologist specialist on digestive tract diseases
  Jeanne Tran Van Nhieu (CHU H. Mondor,Créteil)
- Histology and histopathology
  Fabrice Chrétien (IP Unité d'Histopathologie humaine et modèles animaux)

Image analysis : Vanary Meas Yedid (IP Unité d'Analyse d'Images Quantitative)

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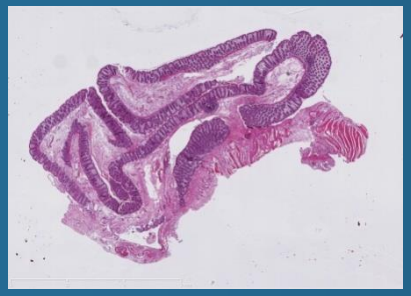
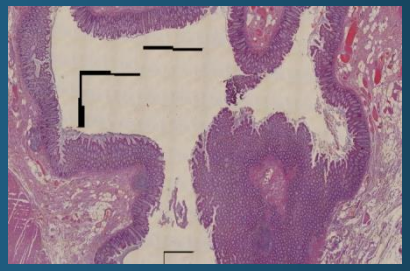
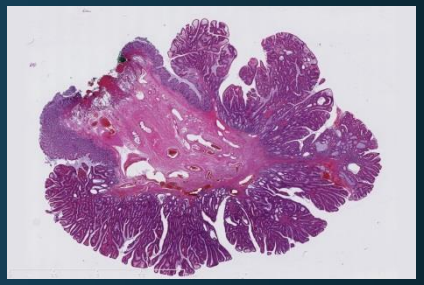
Samples and pathology



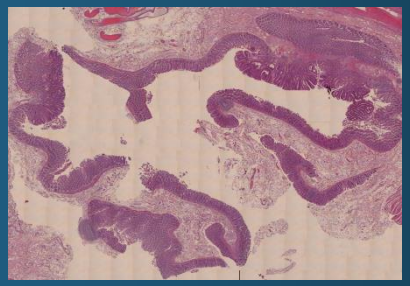
Normal



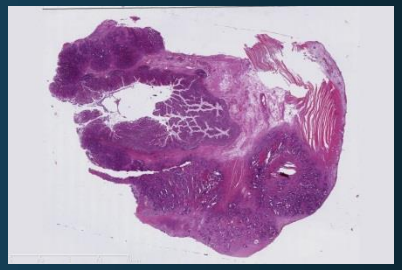
Normal/Polyp



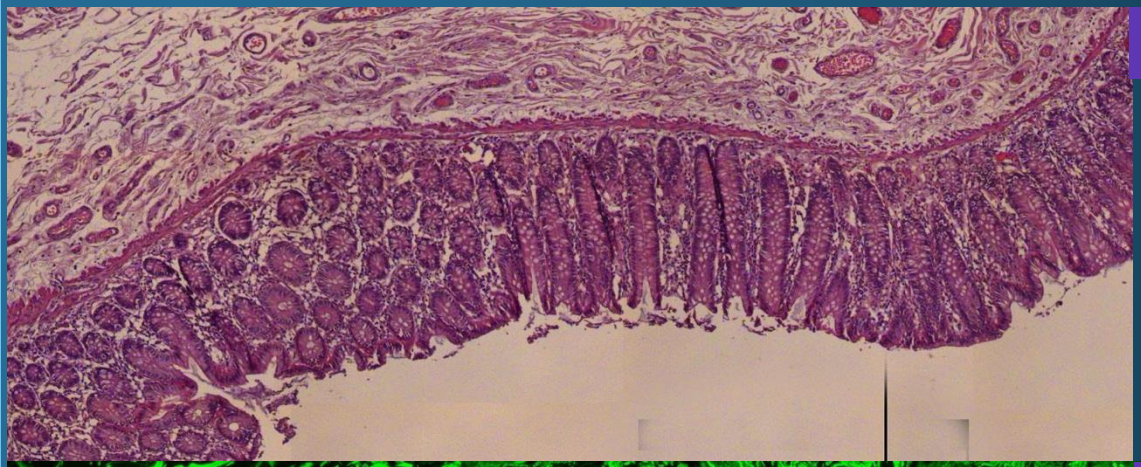
Sessile polyp



Normal/Polyp

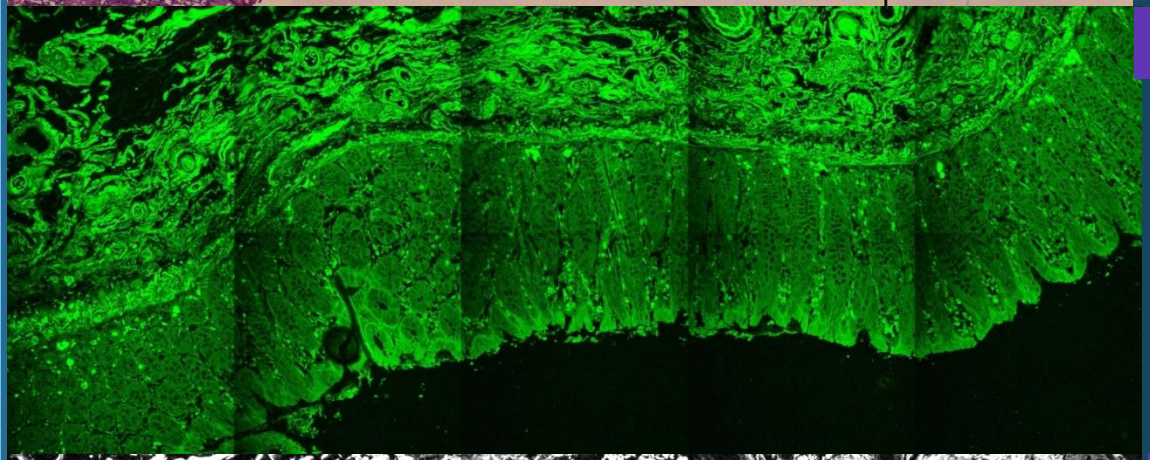


Adenoma carcinome liberkühmien



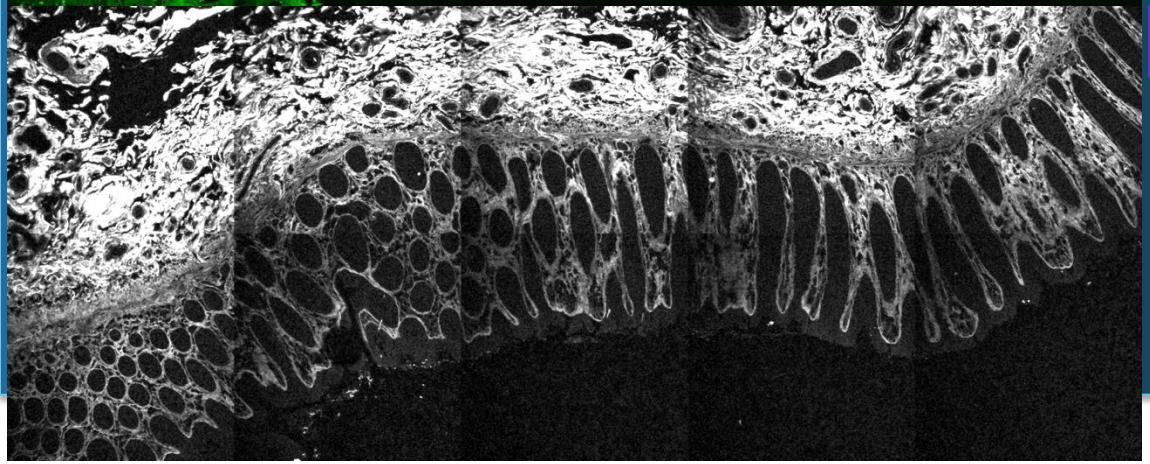
HE-Histological Staining

Tissu/Cell Morphology
and architecture.
Visual expertise for ROI
and pre diagnostic



Autofluorescence @527-556 nm

Functional state of the cells
: biochemical, enzymatic, …



SHG @410-820nm

3D Structural tissue information
(collagen)

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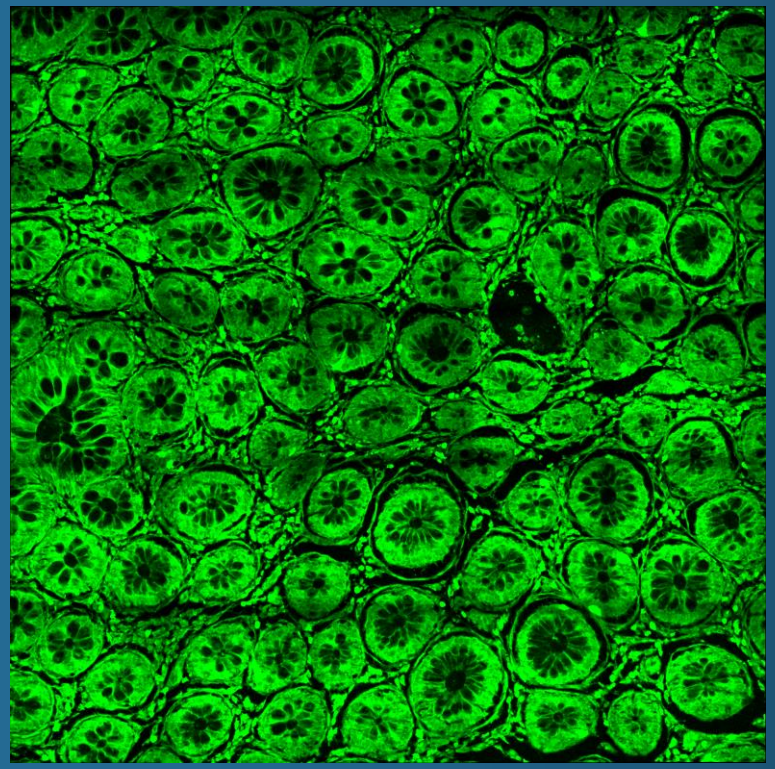
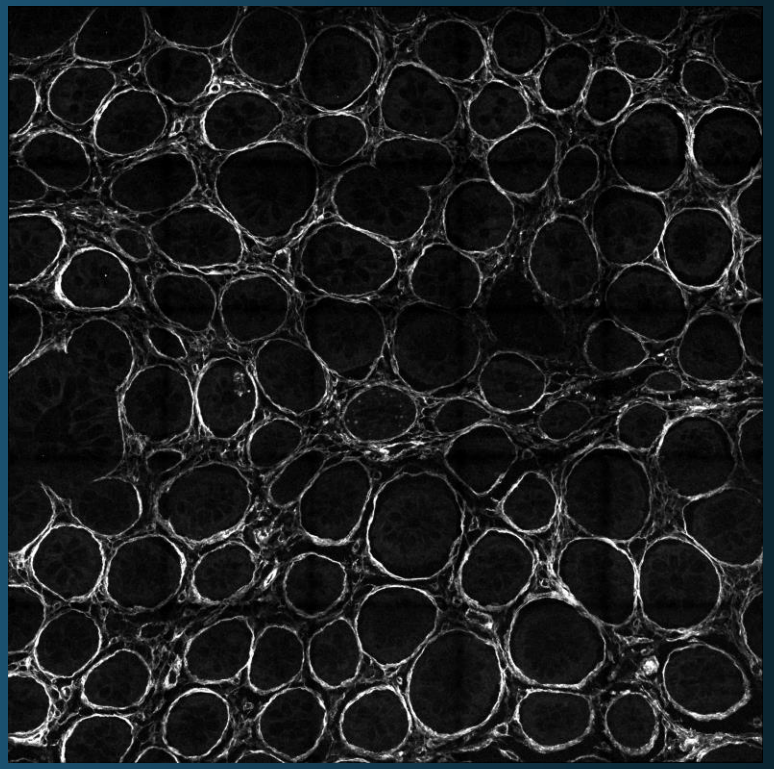
Collagen structure in mucosa

Preserved structure



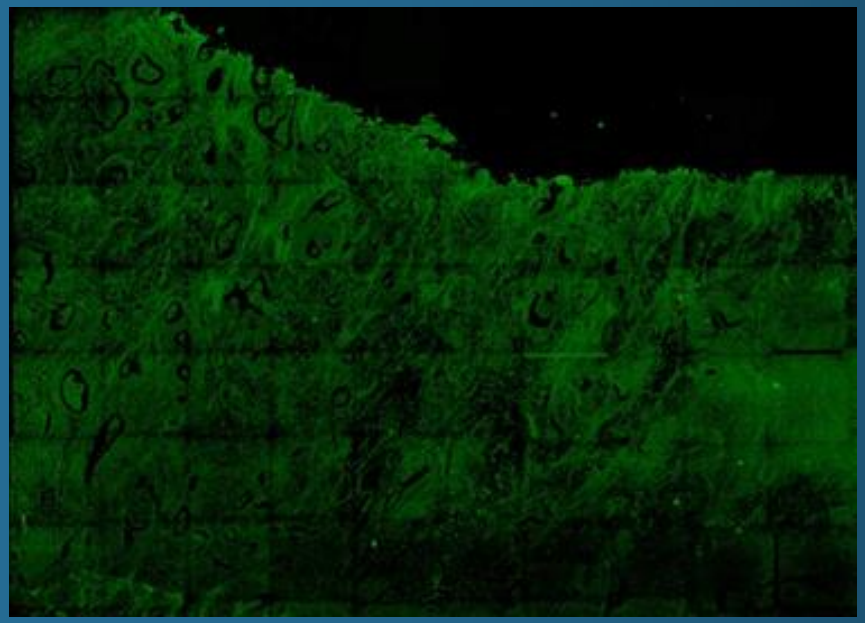


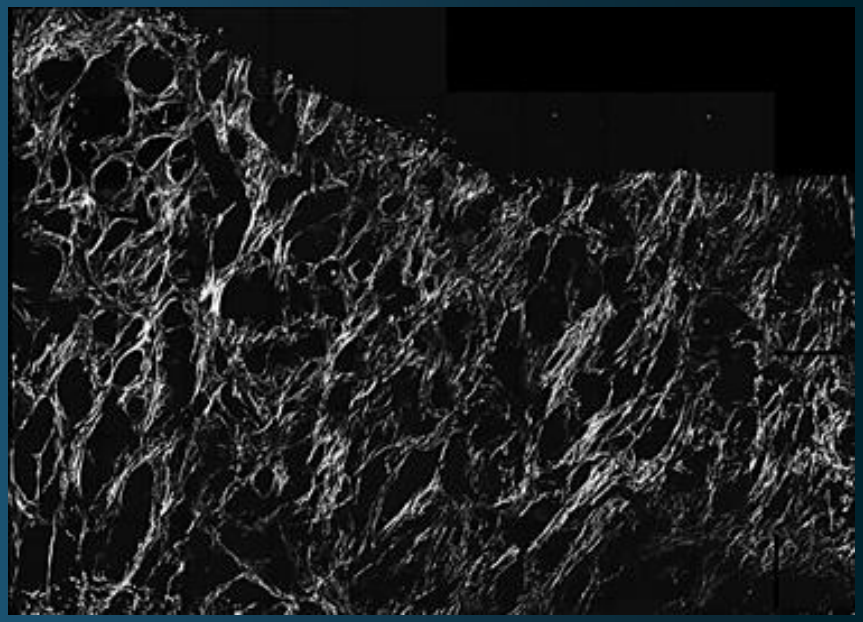
XLPLN 60X W NA:0.9

25 Fields,
211µmx211µm each

Collagen structure in mucosa

Altered structure





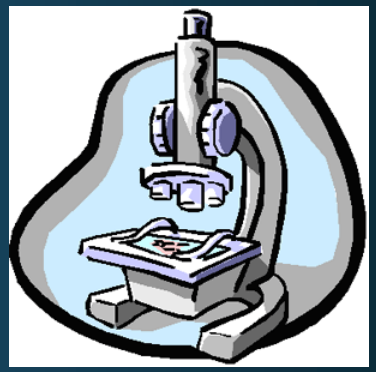
XLPLN 25X W NA:1.05

70 Fields,
509µmx509µm each

A dedicated image analysis must be developed on SHG signals.

Expertise scheme

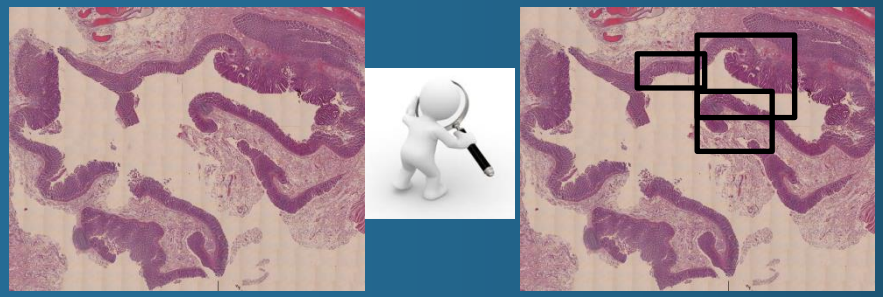
2D

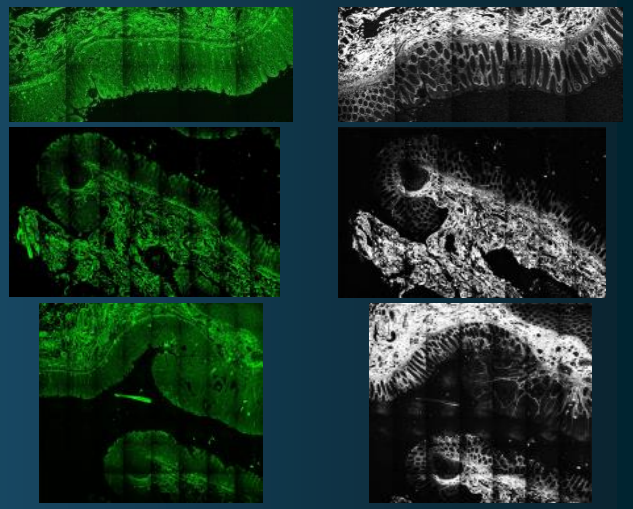


3D

Histo staining slice

intrinsic signals
Slice without staining





HE

Autofluorescence & SHG

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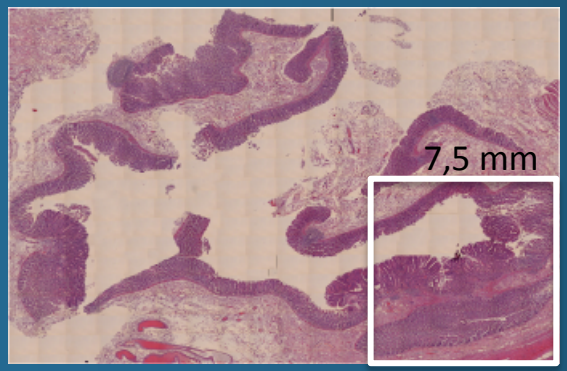
Data Acquisition

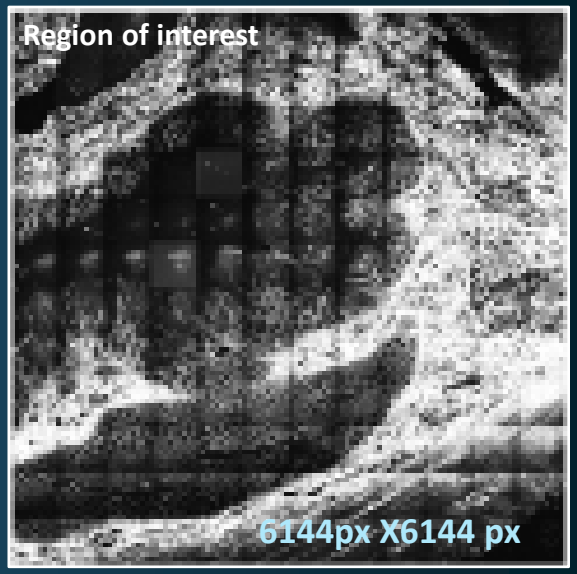
HE-Histological Staining



Plan Apo 10X/NA 0,3

Collagen SHG
Em 410 nm

Slice without staining



Plan Apo 20X/NA1,0 Exc@820nm

Microscopy acquisition and automation : Pascal Roux, Imagopole

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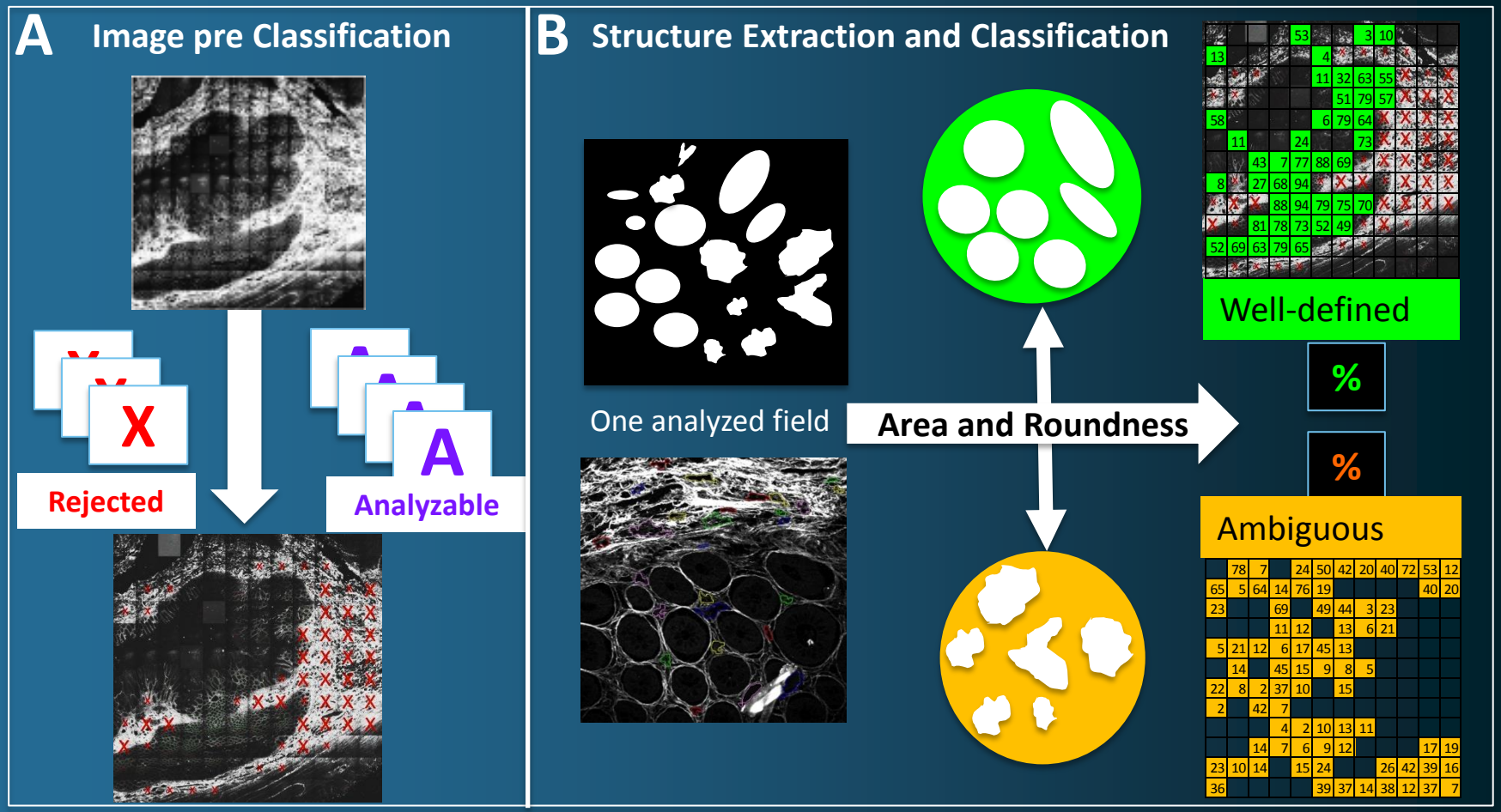
Method

Pre treatment : Maximum Projection by field

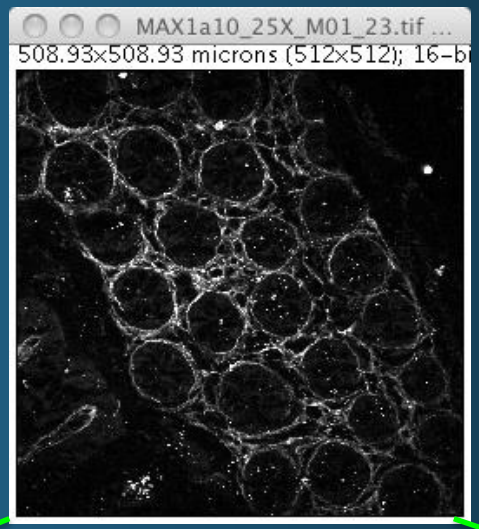


Pre classification

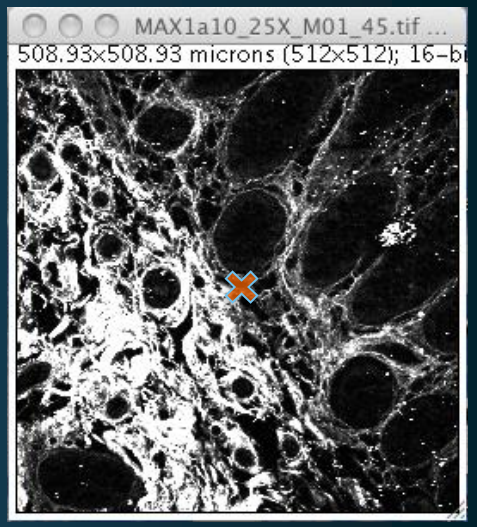
x25

Class 1: Rejected

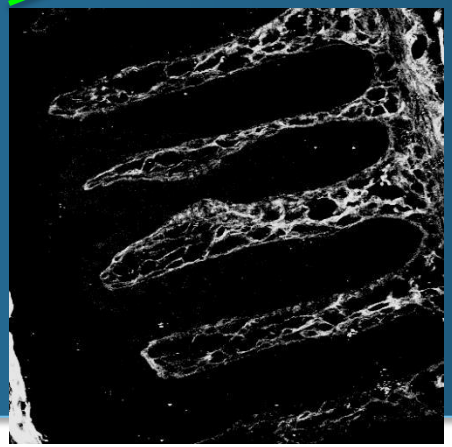
Class 2: Analyzed

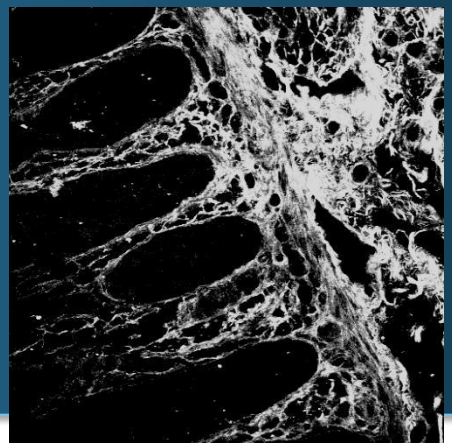


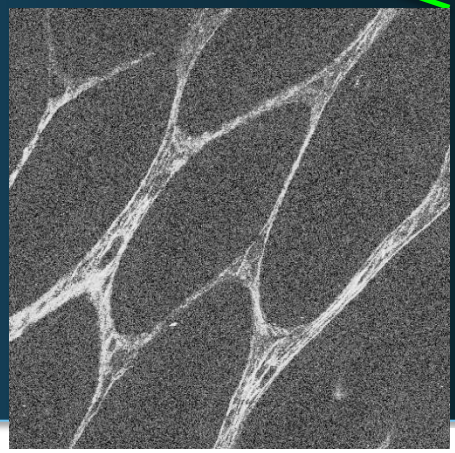
Class 3: ≈Analyzed



Other examples







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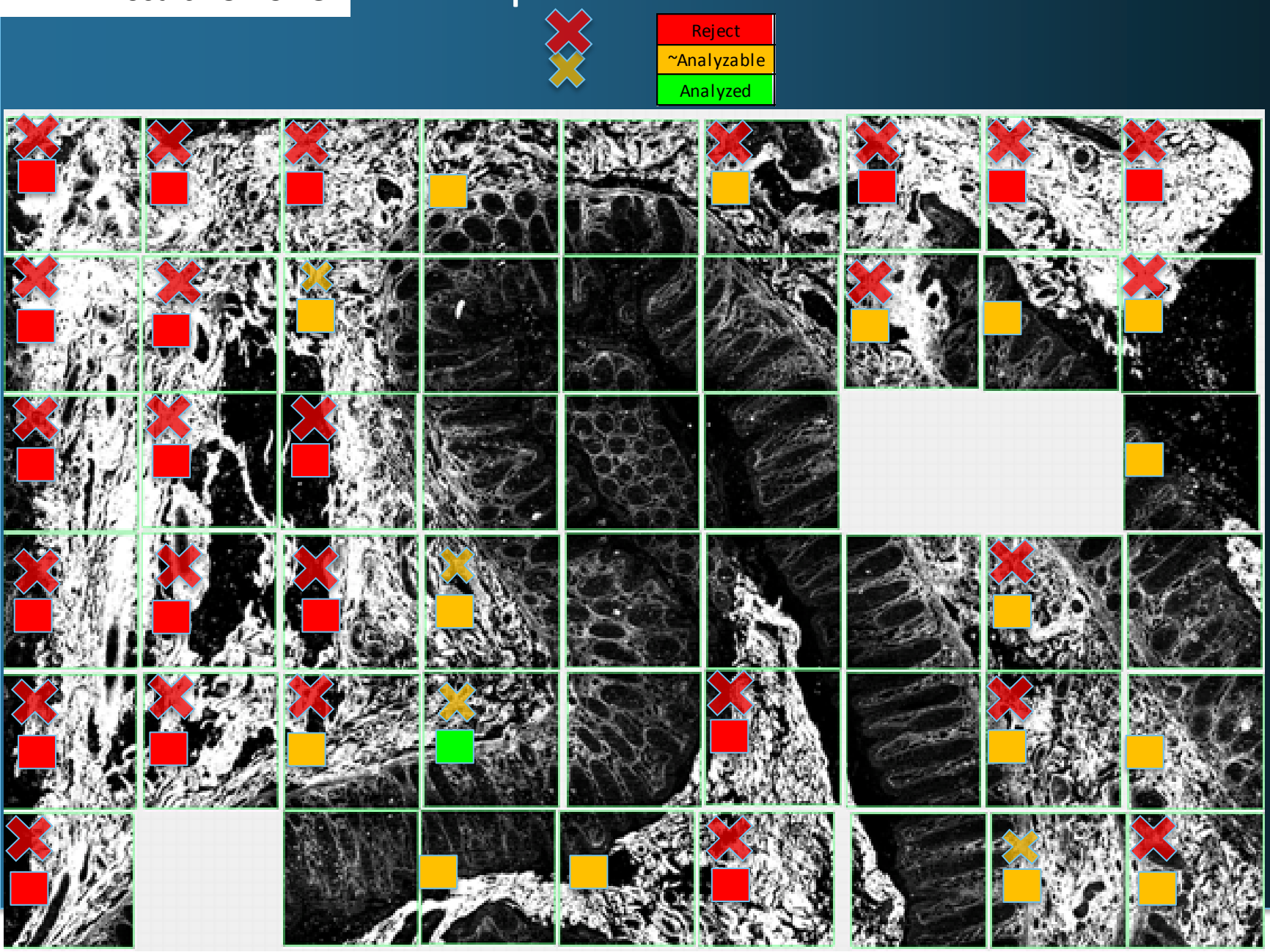
MAX-Mosaic 25X-SHG

Expert v.s Pre classification



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Structure Classification : Details

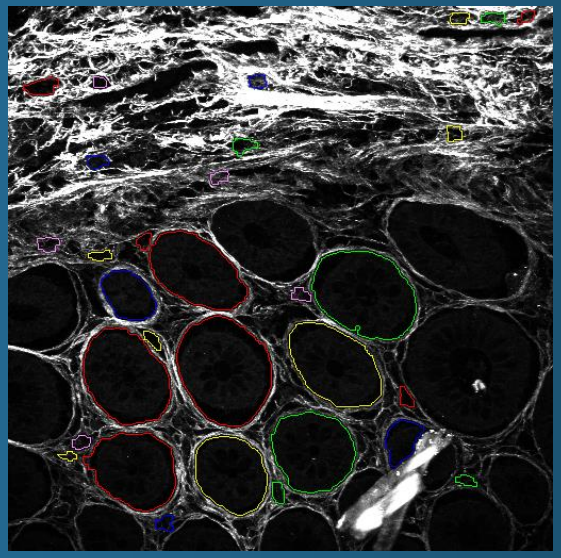
- Tissue structure (closed only) segmentation
- Roundness and area of segmented structures
- Classification
- Artifacts

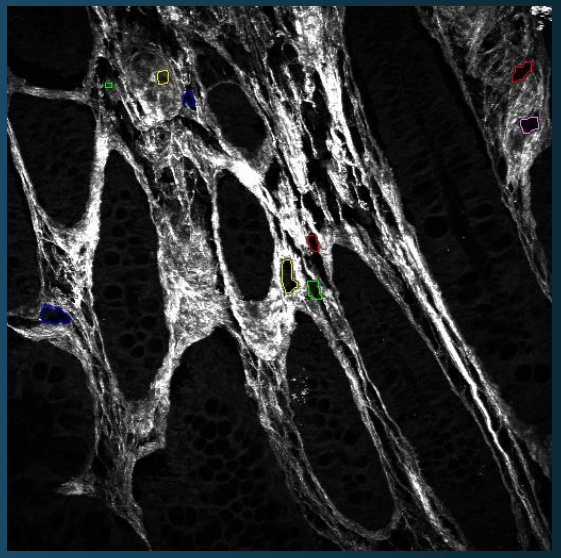
Tissue Structures (Closed)

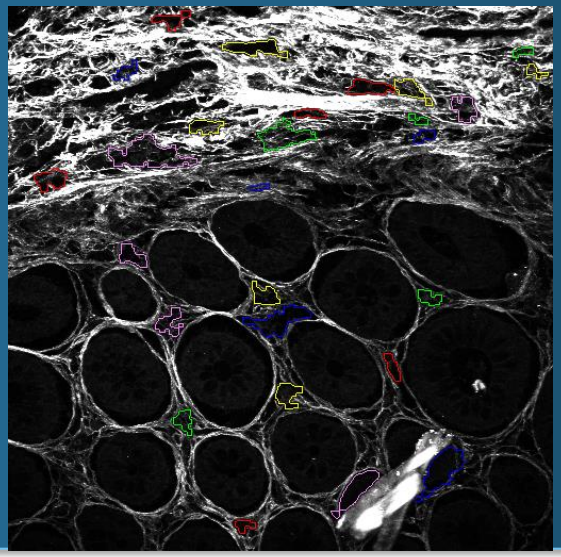
Classified

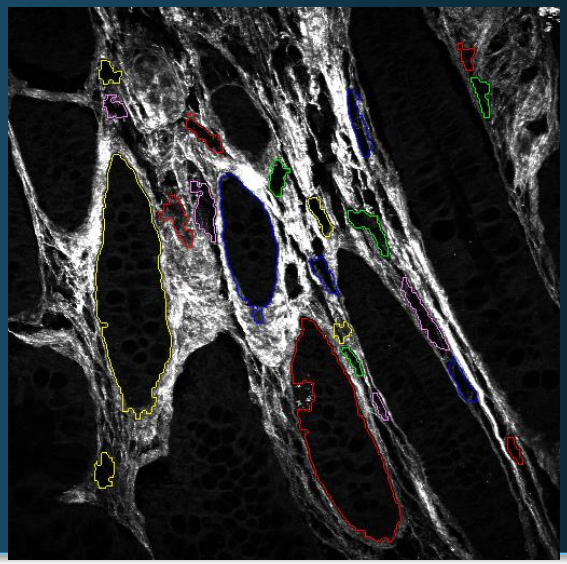
Based on Roundness

Ambiguous

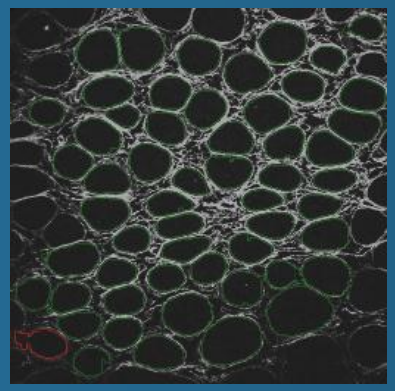


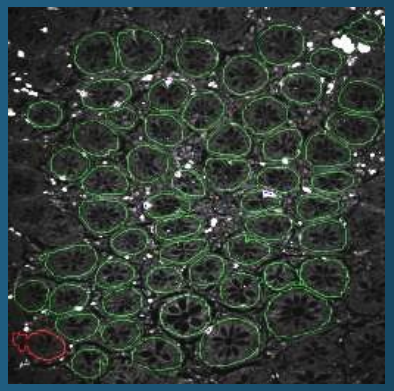


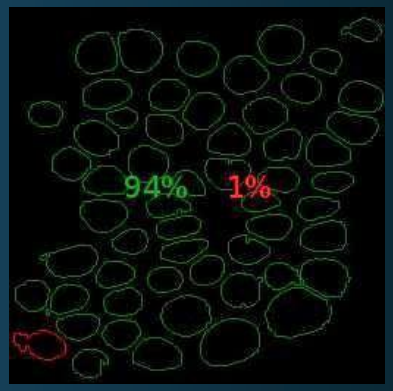




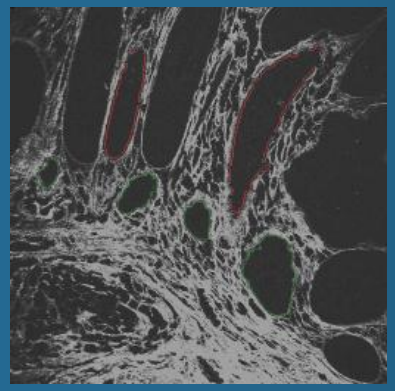
Structure Classification by field

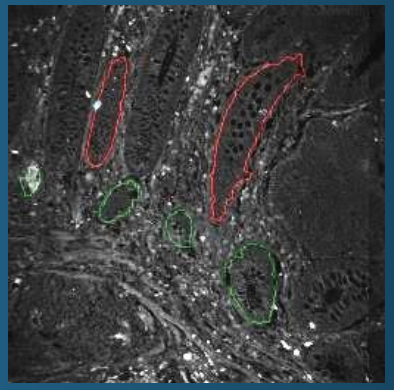






Healthy?





Not healthy?

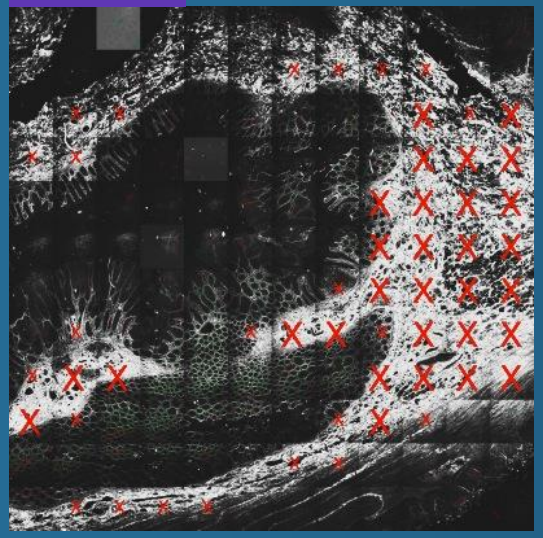
SHGMAX

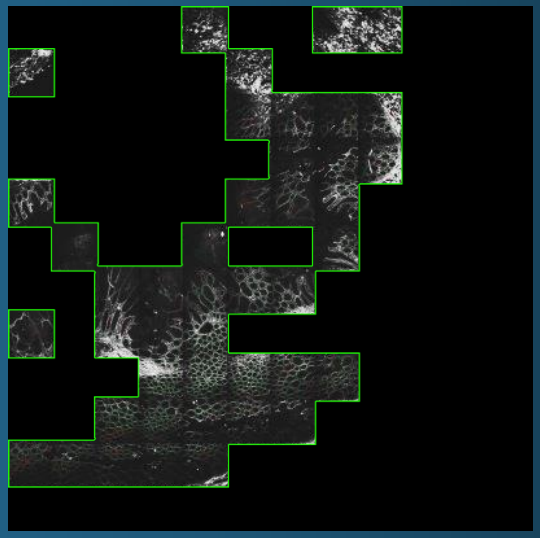
FLUOMAX

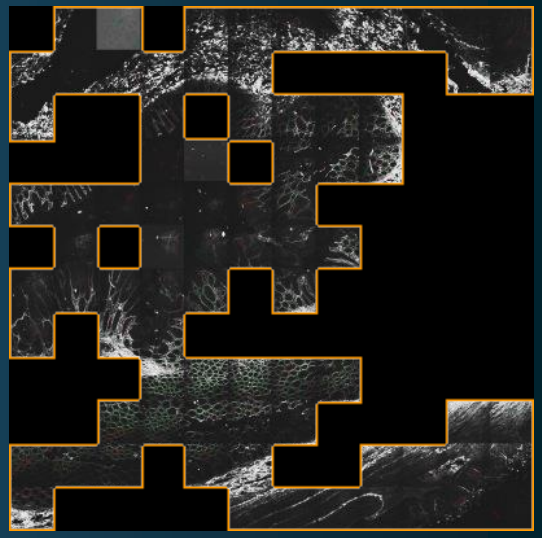
Output

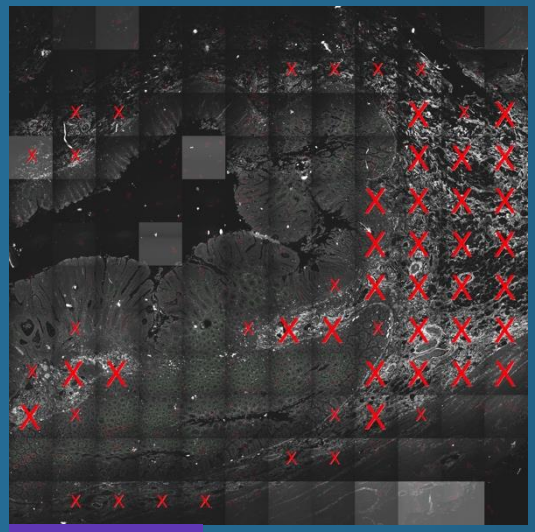
Pre score by mosaic

SHGMAX









FLUOMAX

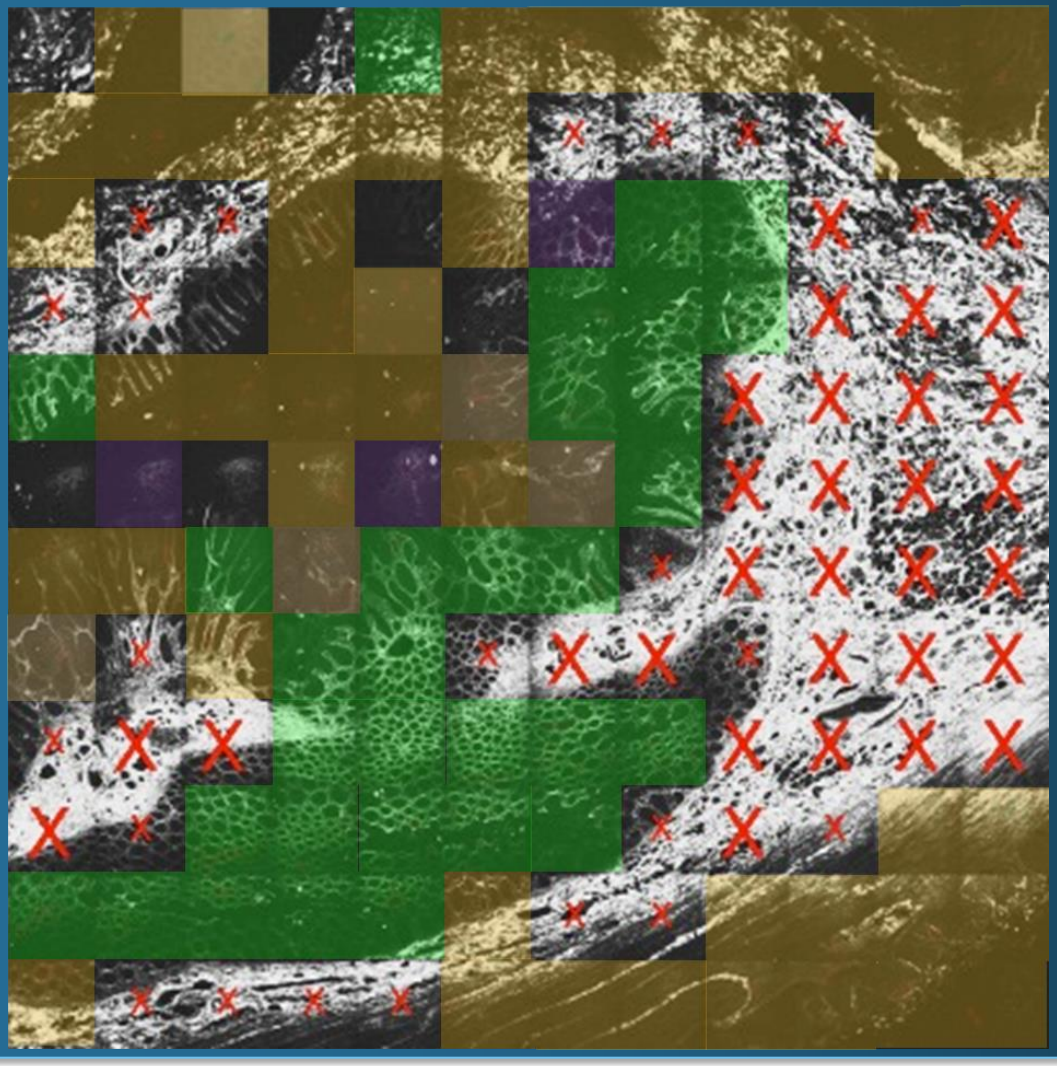
				53			3	10			
13					4						
					11	32	63	55			
						51	79	57			
58					6	79	64				
	11			24			73				
		43	7	77	88	69					
8		27	68	94							
			88	94	79	75	70				
		81	78	73	52	49					
52	69	63	79	65							

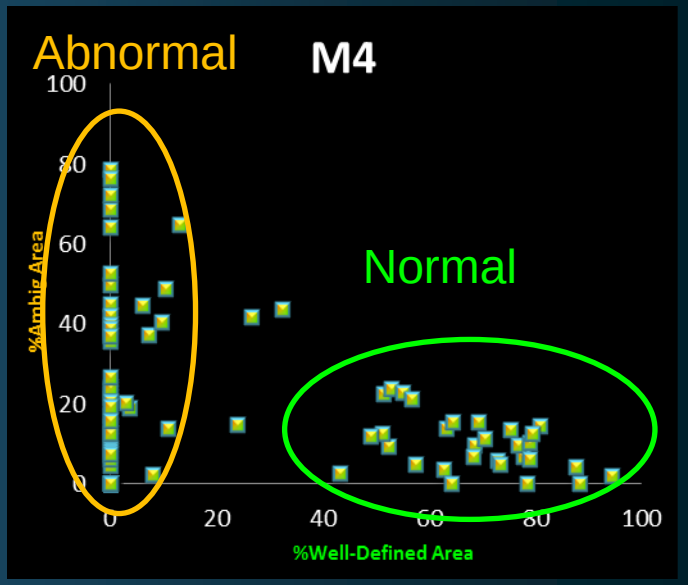
% Well-defined Area

	78	7		24	50	42	20	40	72	53	12
65	5	64	14	76	19					40	20
23			69		49	44	3	23			
			11	12		13	6	21			
5	21	12	6	17	45	13					
	14		45	15	9	8	5				
22	8	2	37	10		15					
2		42	7								
			4	2	10	13	11				
		14	7	6	9	12				17	19
23	10	14		15	24			26	42	39	16
36					39	37	14	38	12	37	7

%Ambiguous Area

Data Visualisation and Annotation by Mosaic

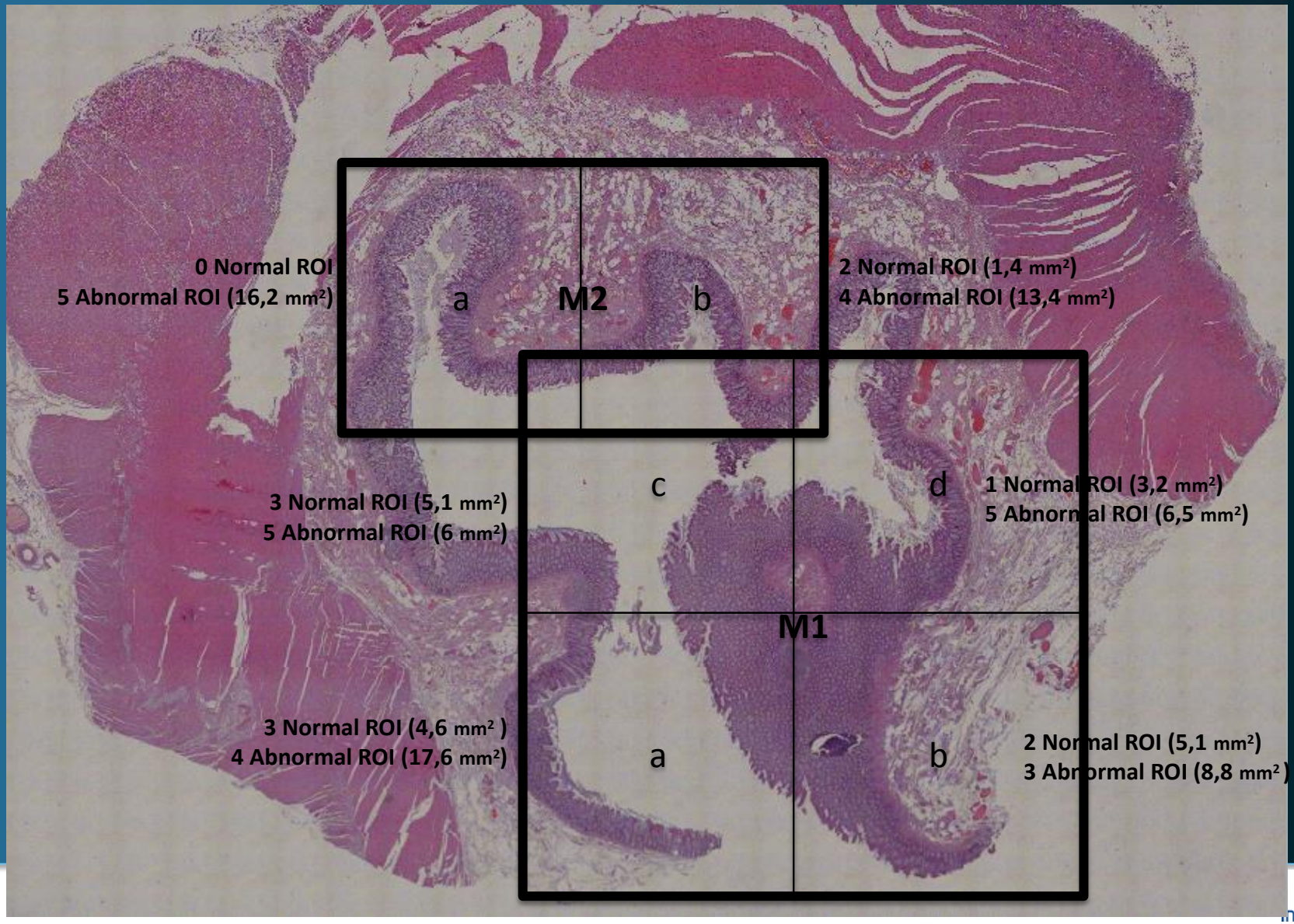




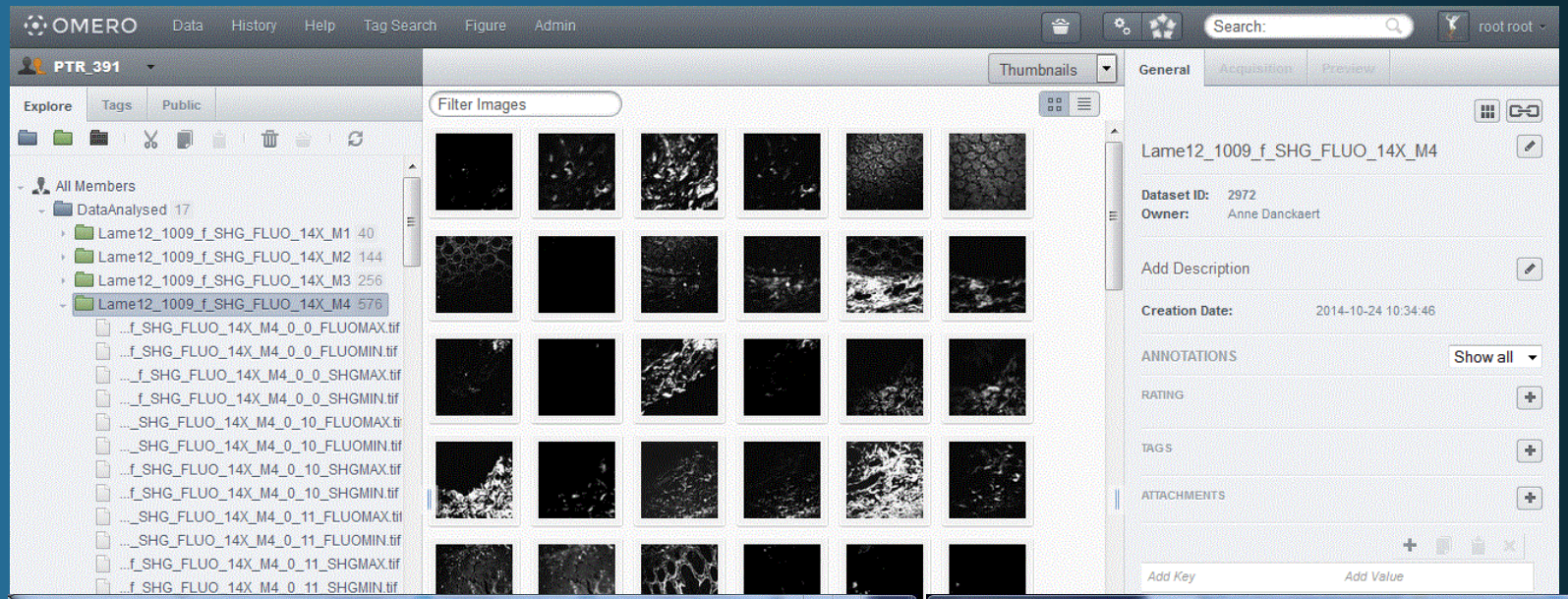
Cartography

- Automatic ROI location
- ROI annotation (5N 3AN)

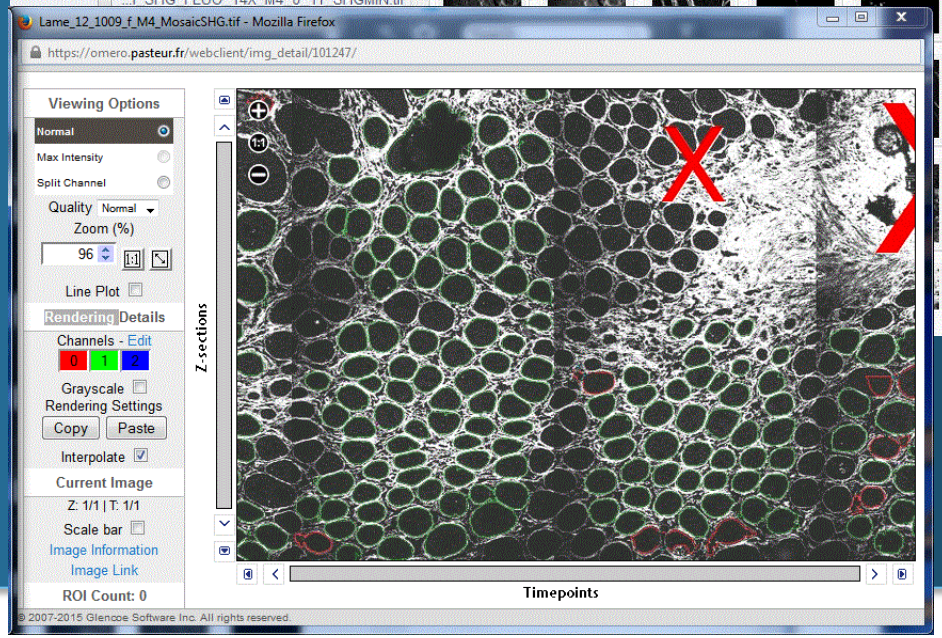
Resume of Pre score in immuno histological slice : Normal/Abnormal

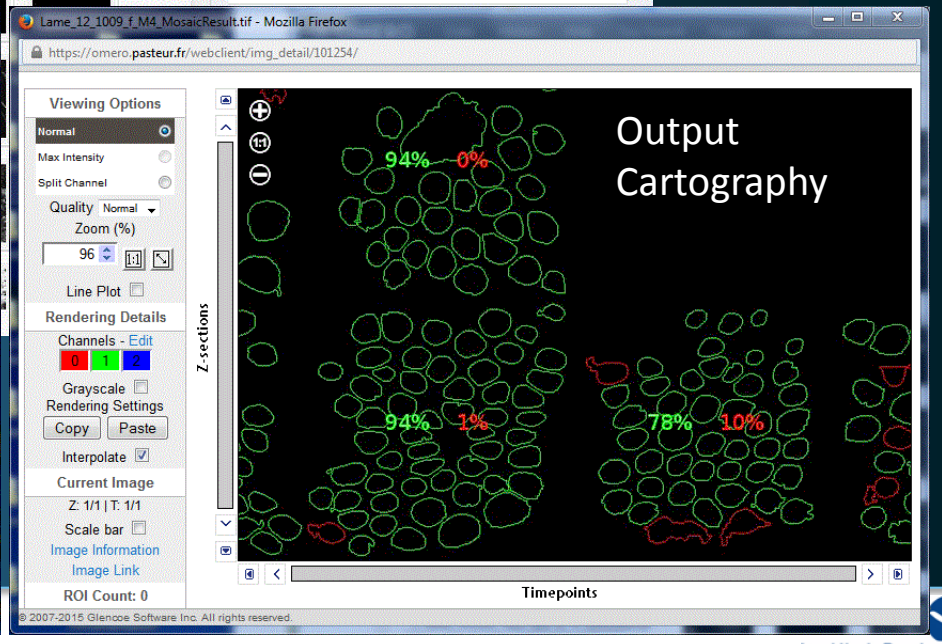


Data Organisation (OMERO)



Raw Data
+
Result Cartography
+
Annotations





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Perspectives

Multimodal Data Spaces

□

Images + Metadata

- Colour space
- Intensity of response
- Texture
- Region location

…

○

Annotations

- Clinical
  age, sex, weight, species,…
- Biological
  genomics, proteomics, transcriptomics, …
- Sample
  organ, tissue, cell, …

△

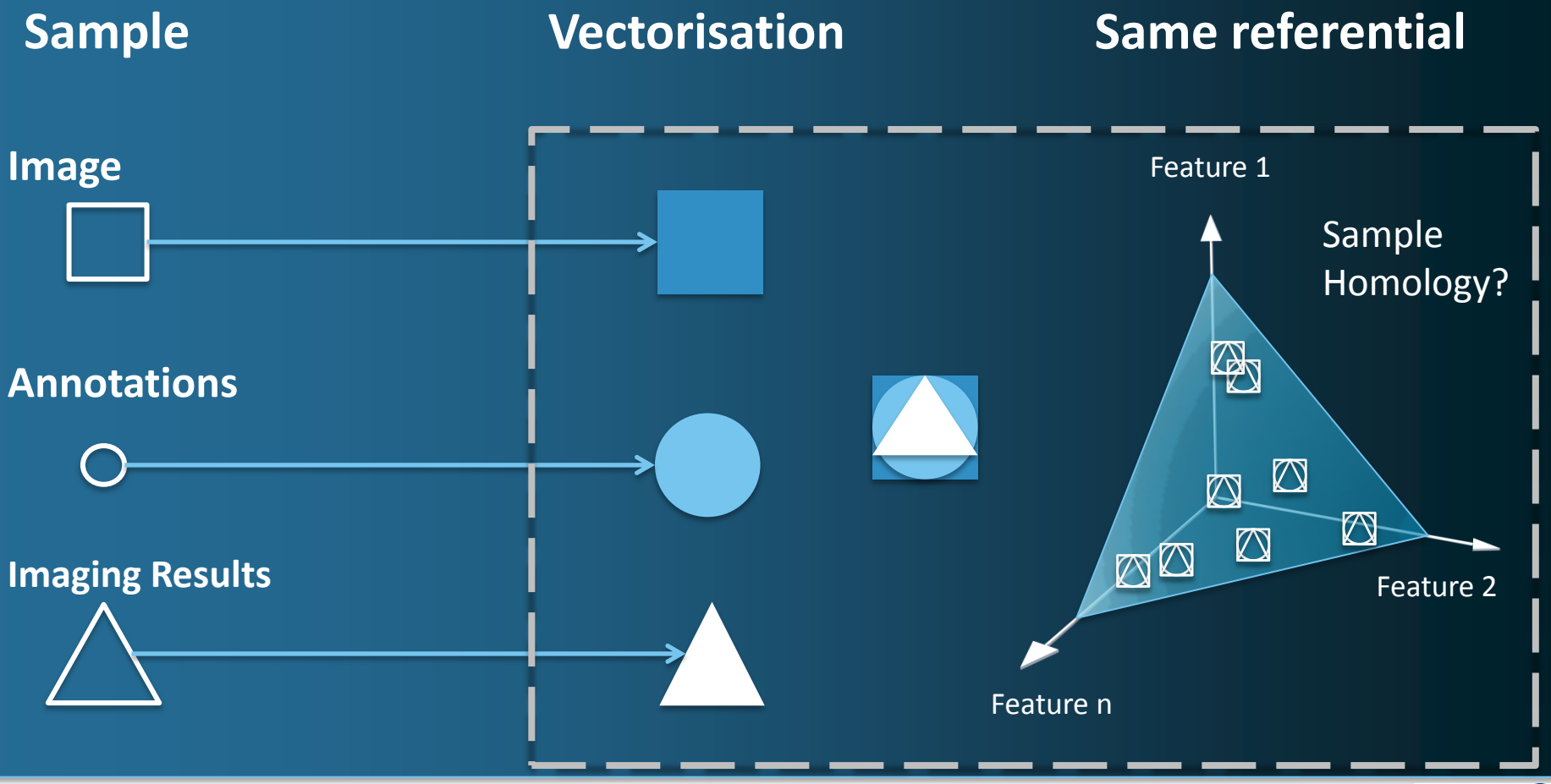
Results

- Morphology
  roundness, deformation, surface, …
- Quantitative Data
  density, surface, volume, velocity, …
- Meta-information
  cluster, cartography, …

Perspectives

Virtual Platform for multimodal data analysis

Cross Correlation of Data Analysis (Concept)



Based on Costa Pereira et al. Method (2014) IEEE Transactions on Pattern Analysis and Machine intelligence

Virtual Platform for multimodal data analysis

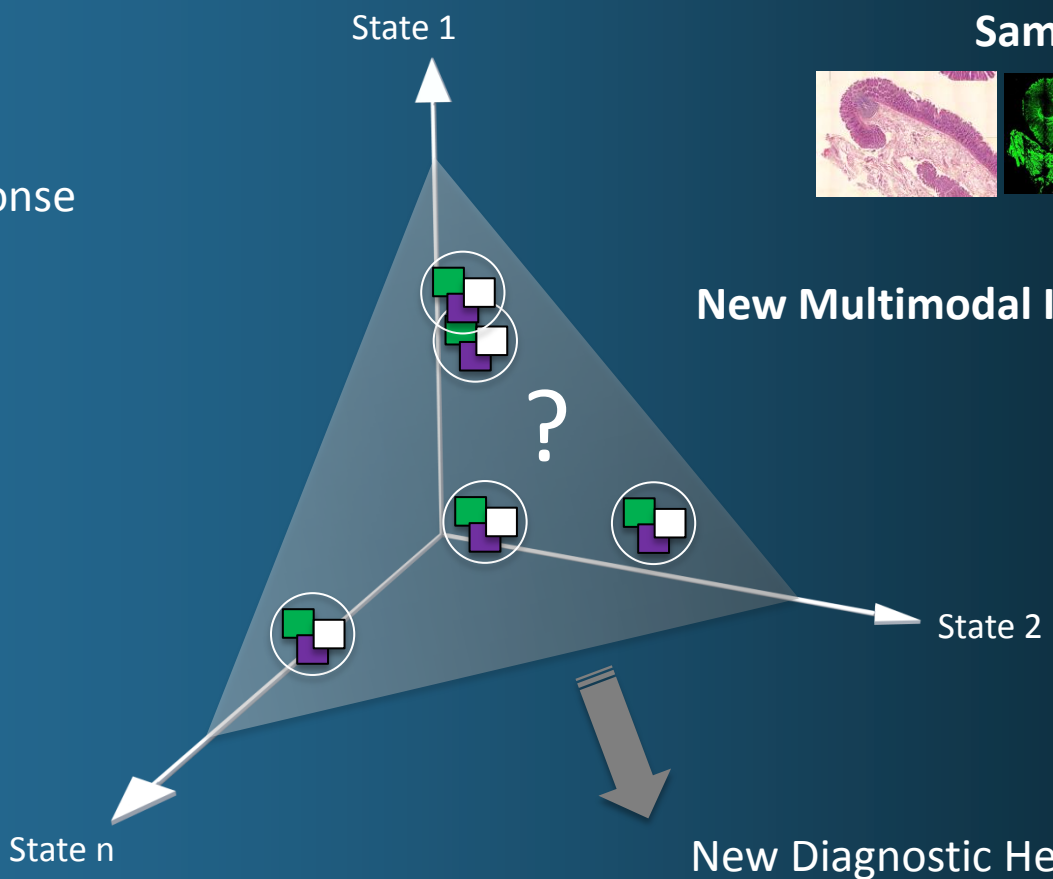
Deviation to Multimodal Imaging

Images

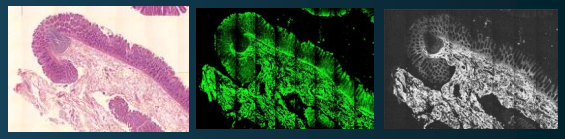
colour histogram
intensity of response
texture
…

Annotations

Clinical Data
Visual Expertise
Quantitative Score
Pathology
…



Same ROI



New Multimodal Imaging Referential

New Diagnostic Help ?

Cross Correlation Imaging Signal Space

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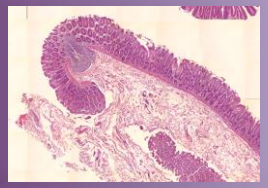
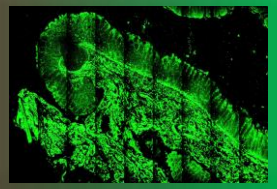
1



Multimodal imaging



Visual expertise Data **Images with associated Meta Data**

2

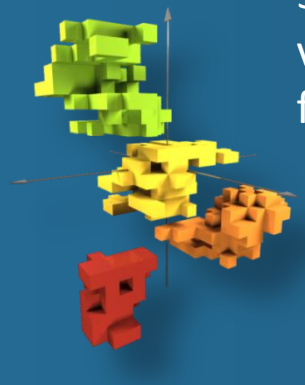
Feature Extraction with associated method parameters

color histogram, texture, morphology, pattern classification, location …
density of nuclei, volume, intensity …

3

Similarity measures

3D virtual voxel space for feature



clustering by homology

Cross-correlation Image Analysis & Statistics

4

Comparative space

- biological images retrieval and data resources integration
- clustering of objects sharing analogous qualitative and quantitative features
- diagnosis of a new image by similarity search
- comparison of specific features between structures, regions, models, samples...

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