NIREOS

Measurements on breast cancerous tissue with HERA VIS-SWIR

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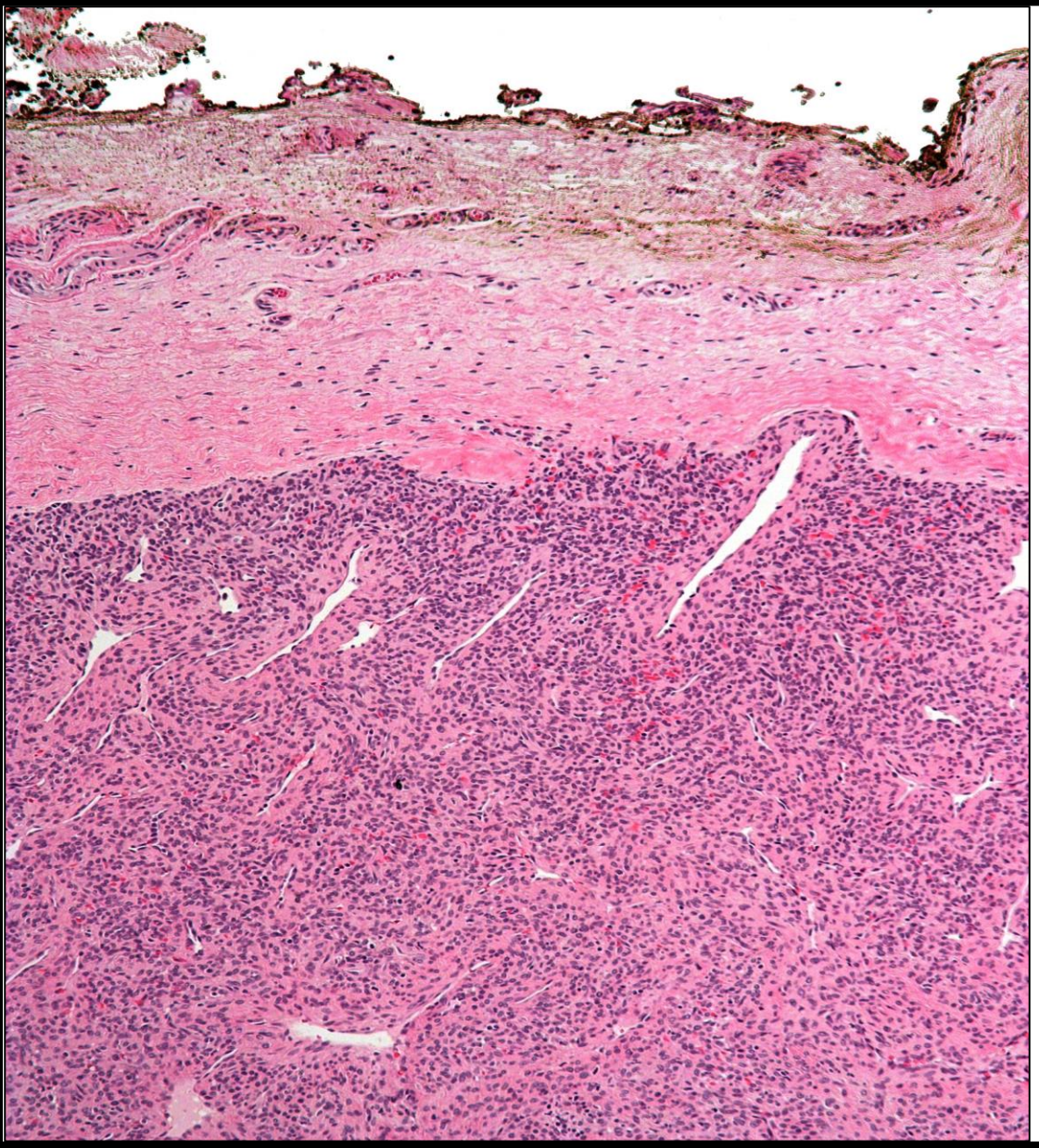
Challenges in intraoperative margin assessment



Up to 39% patients who undergo surgery leave the operating room **without a complete tumor resection** due to positive or close margins.*

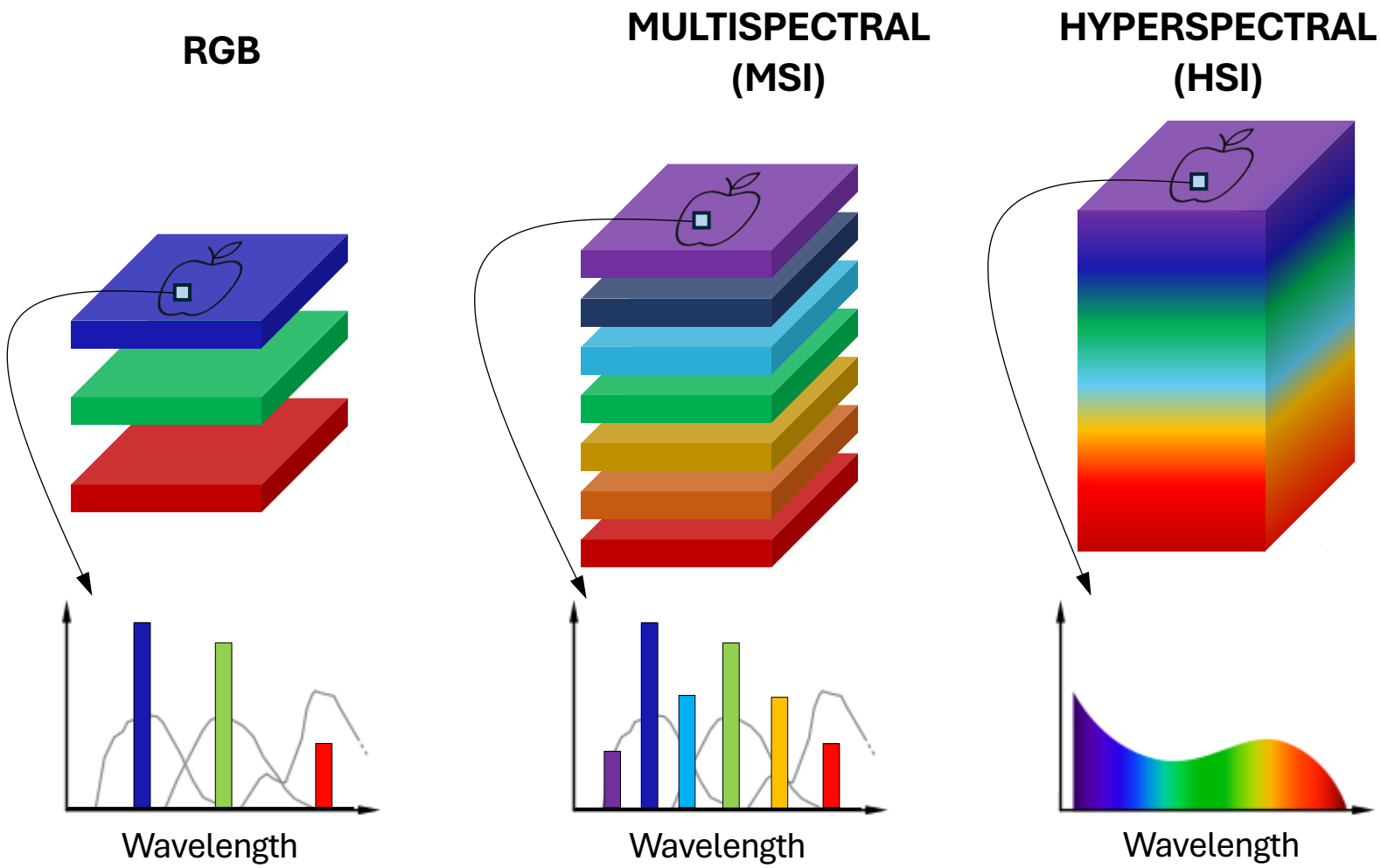
A **complete resection** is associated with **3-5 times improvement in the patient survival rate***

The current standard involves coordination between surgical room and pathology department → **usually not feasible** or hard to do

Q: Could the surgeon make a **quick** and **reliable** assessment of margins *inside* the operating room?

A: **Optical Methods** (RGB, Multispectral or Hyperspectral, Raman,…) + **Machine Learning**

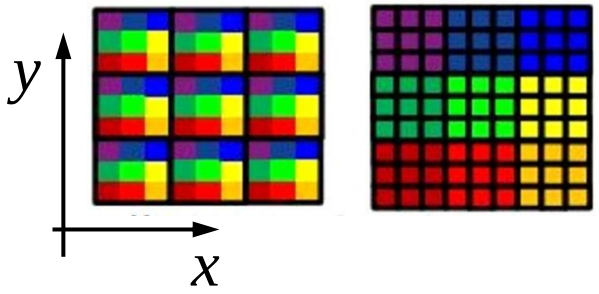
** Fei B, et al., J Biomed Opt. 2017 Aug;22(8):1-7*

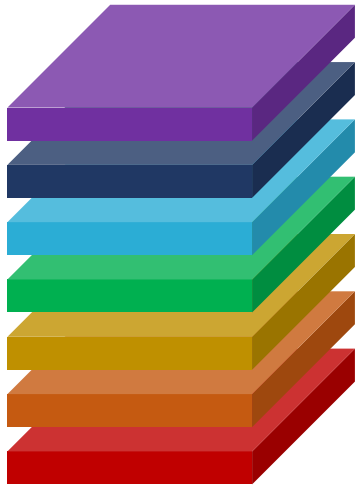


HSI provide a combination of **SPECTRAL** and **SPATIAL** information, paving the way to the creation of **computer-aided diagnostic tools** for both **stained** and **unstained** histological specimens.

Improvement in the detection of tumors compared to traditional RGB.

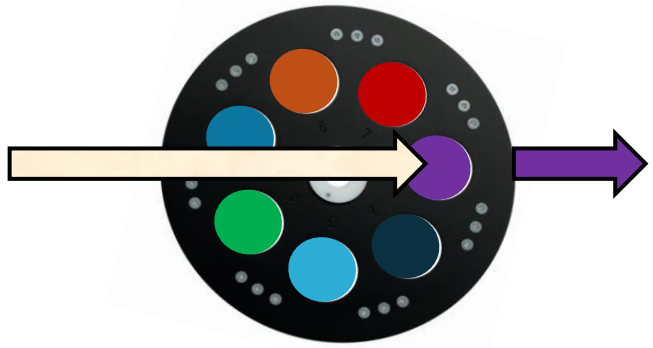
SNAPSHOT IMAGING

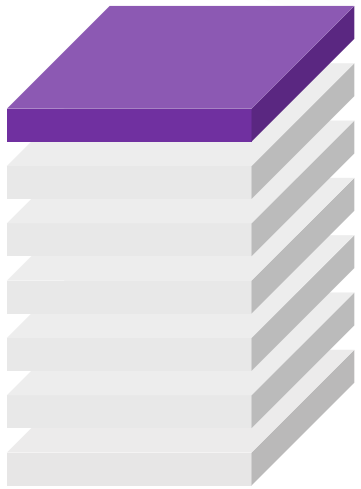




- Fast
- Compromise Spatial & Spectral res.

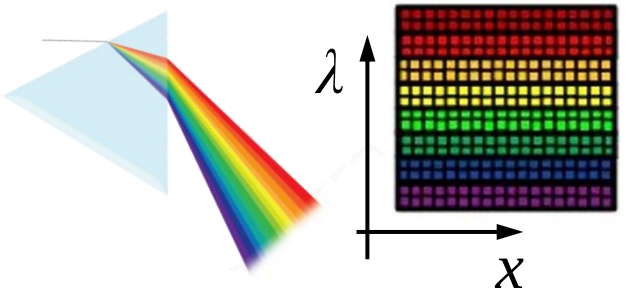
STARING IMAGING



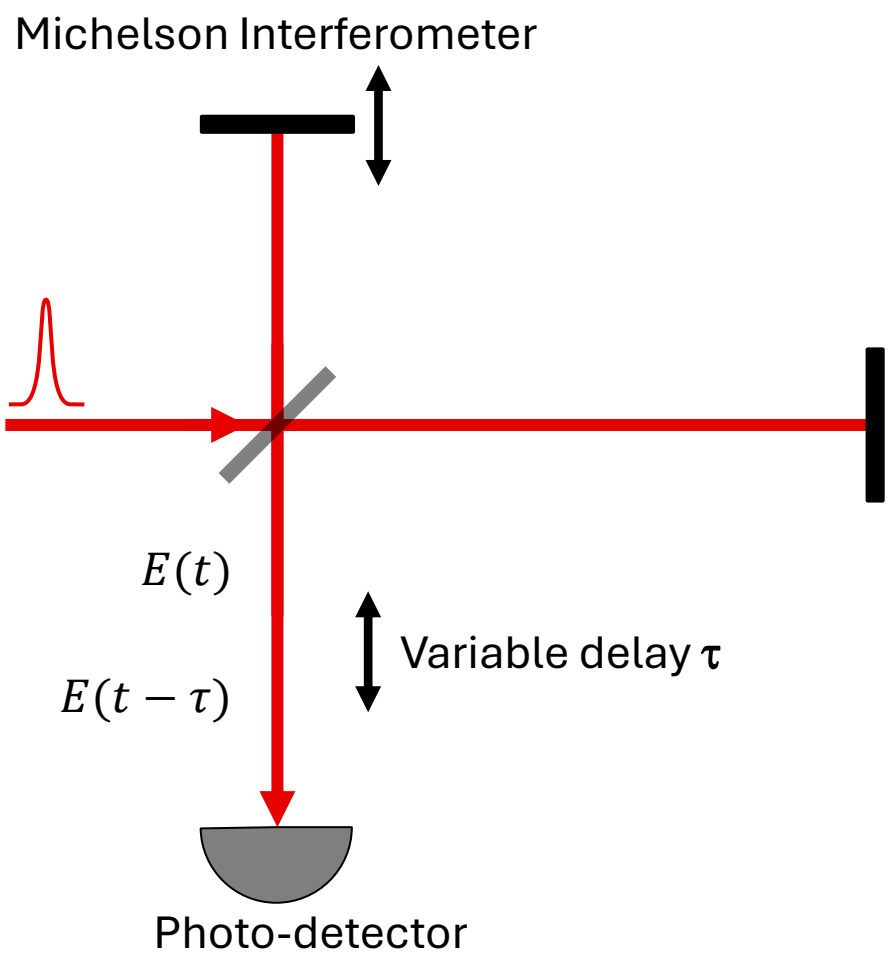


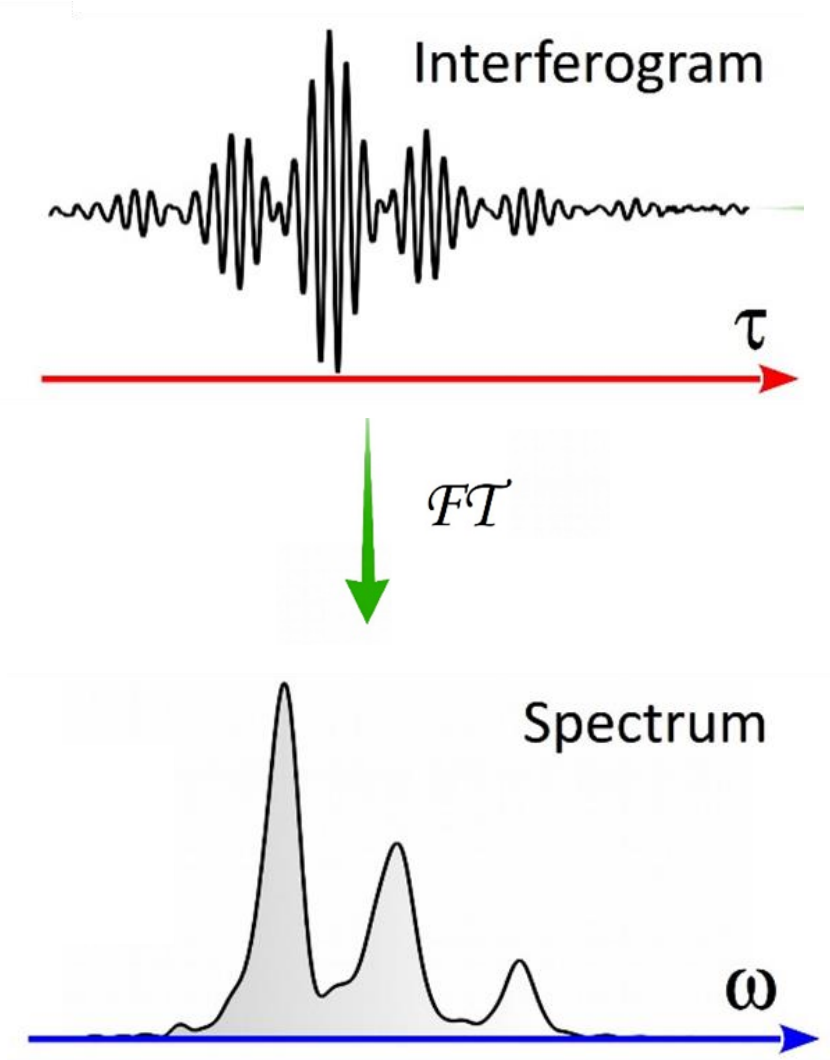
- Good Spatial Resolution
- Low number of bands

PUSHBROOM IMAGING



- Good Spatial & Spectral res.
- Line Scan: movement required

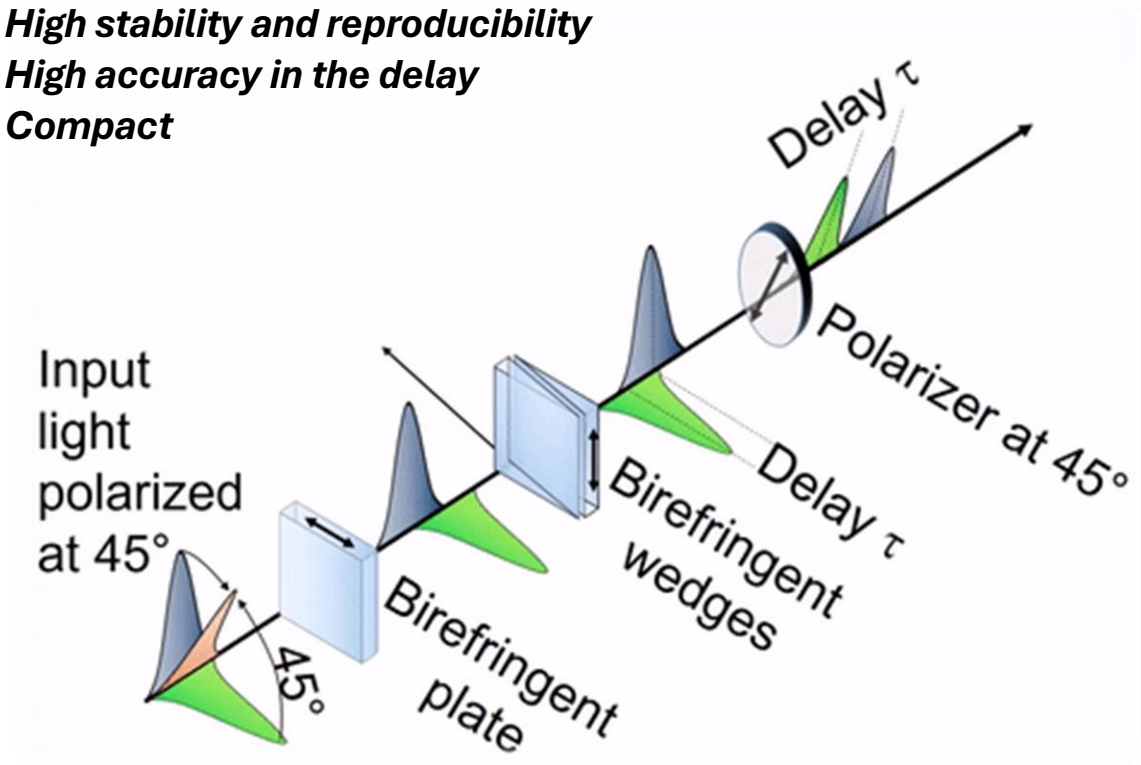

Michelson Interferometer
$E(t)$
$E(t-\tau)$
Variable delay τ
Photo-detector

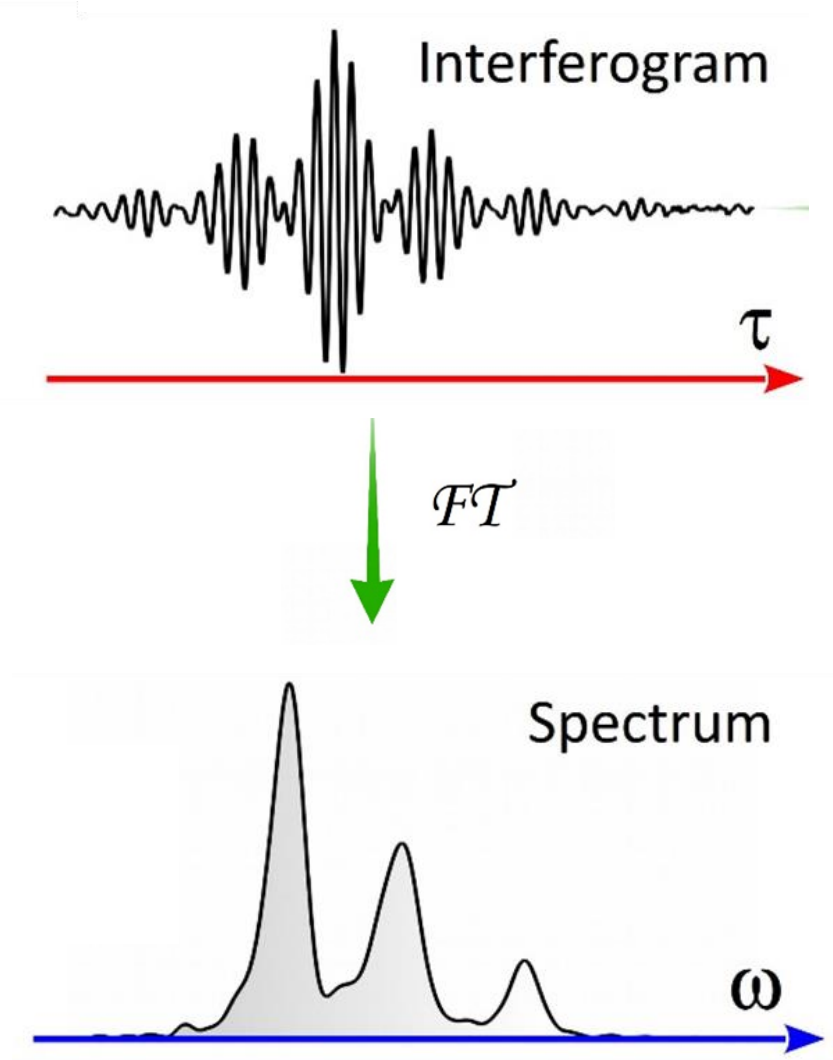

Interferogram
τ
$\mathcal{FT}$
Spectrum
ω

Fourier transform approach

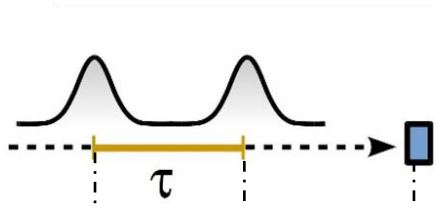
Based on the patents
N°: US9182284B2 (2013)
N°: 102018000008171 (2018)
N°: 102023000005346 (2023)

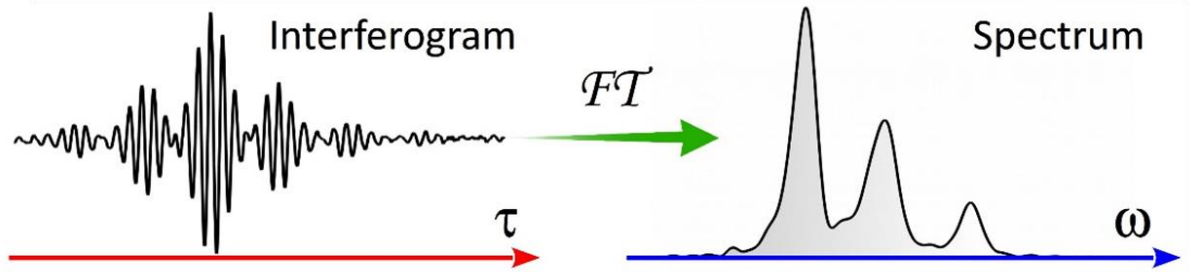
- ***High stability and reproducibility***
- ***High accuracy in the delay***
- ***Compact***



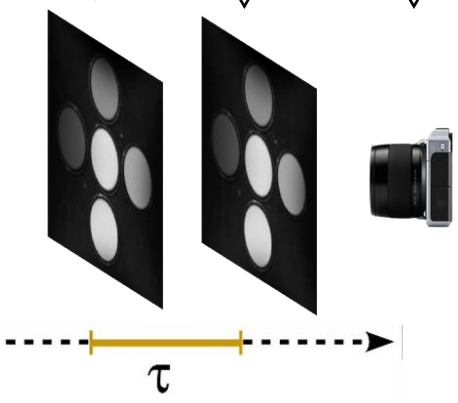


FTIR spectrometer
(**single pixel detector**)

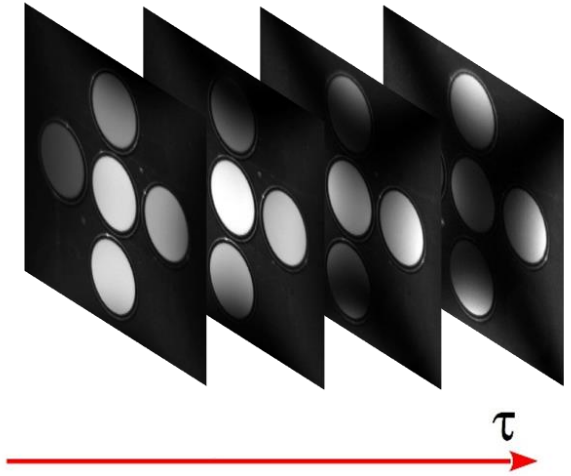




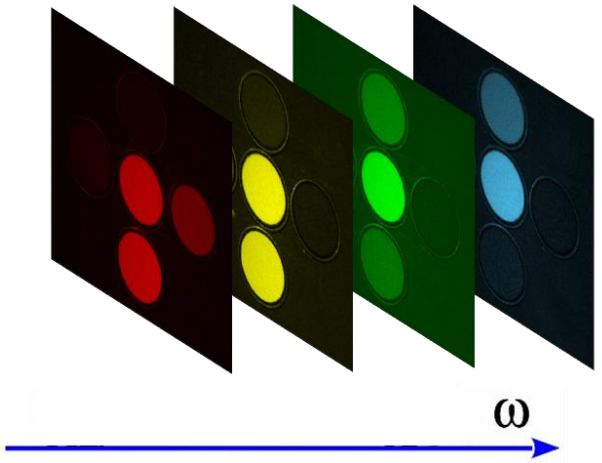
FTIR Hyperspectral camera
(**2D detector**)



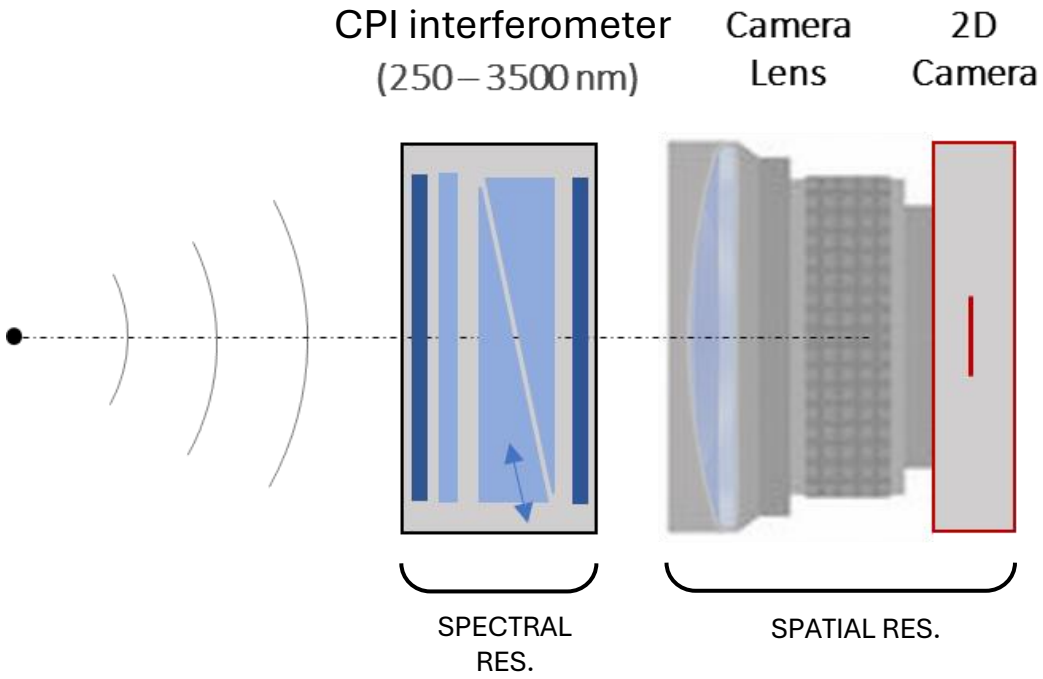
Time domain data-cube



Hyperspectral data-cube



Fourier transform Hyperspectral camera



Spectral range (limited by the sensor):

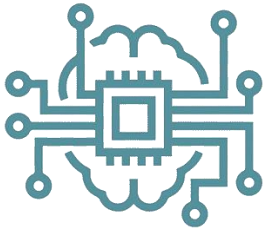
- 400 – 1000 nm (Si detector)
- 900 – 1700 nm (InGaAs detector)
- 1200 – 2300 nm (T2SL detector)
- **400 – 1700 nm (Si + InGaAs)** NEW

- **High Spatial & Spectral Resolution** (e.g. 1.3 Mpixel // <1.5 nm @400 nm)
- **Staring technique** → Ease of use & **Integration with commercial microscopes**
- No slit, no gratings → **High optical throughput** → **Fluorescence Hyperspectral Imaging**
- **Variable Spectral Resolution** (selectable via software) → Flexibility and versatility

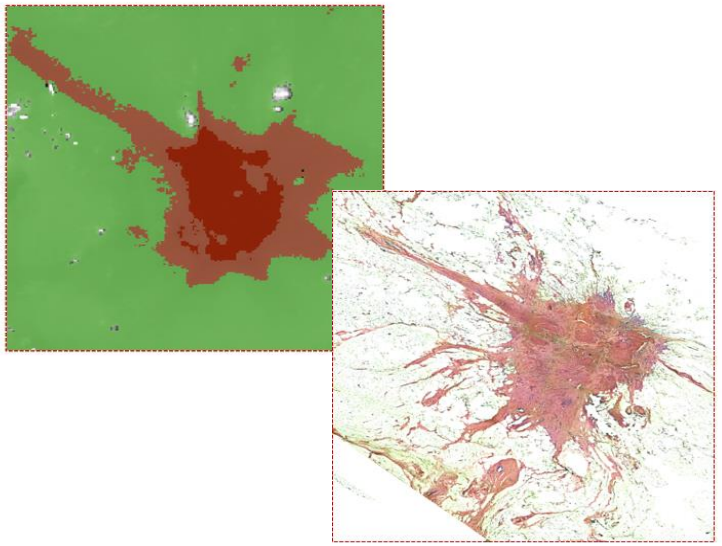
Hyperspectral imaging in pathology - workflow

Step 1: collecting hyperspectral data on fresh biopsies & preprocessing

Step 2: training & validation of an algorithm on known spectra sampled from datacubes



Step 3: test the model on whole datacubes and visualize classified images



Step 4: comparing classified image with H&E-stained image (gold standard) to evaluate the model

Hyperspectral imaging in pathology – experimental setup

Field-of-view (FOV) ≈ 3 x $2.5\ cm$

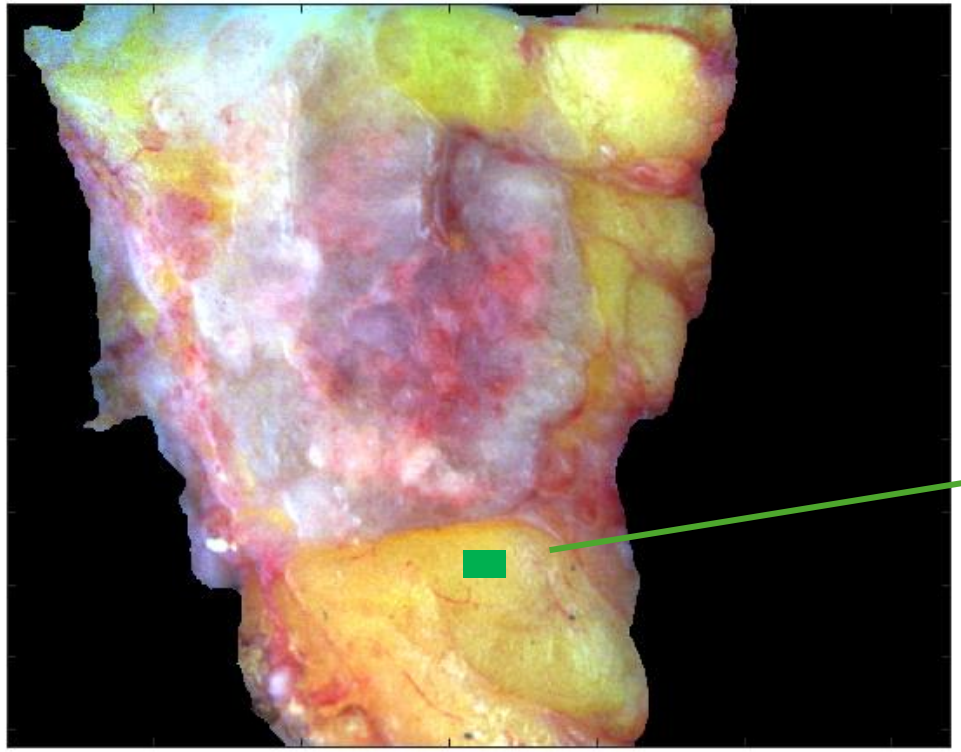
Image size $= 1280$ x $1024\ px$

Spatial resolution $\approx 23\ \mu m/px$

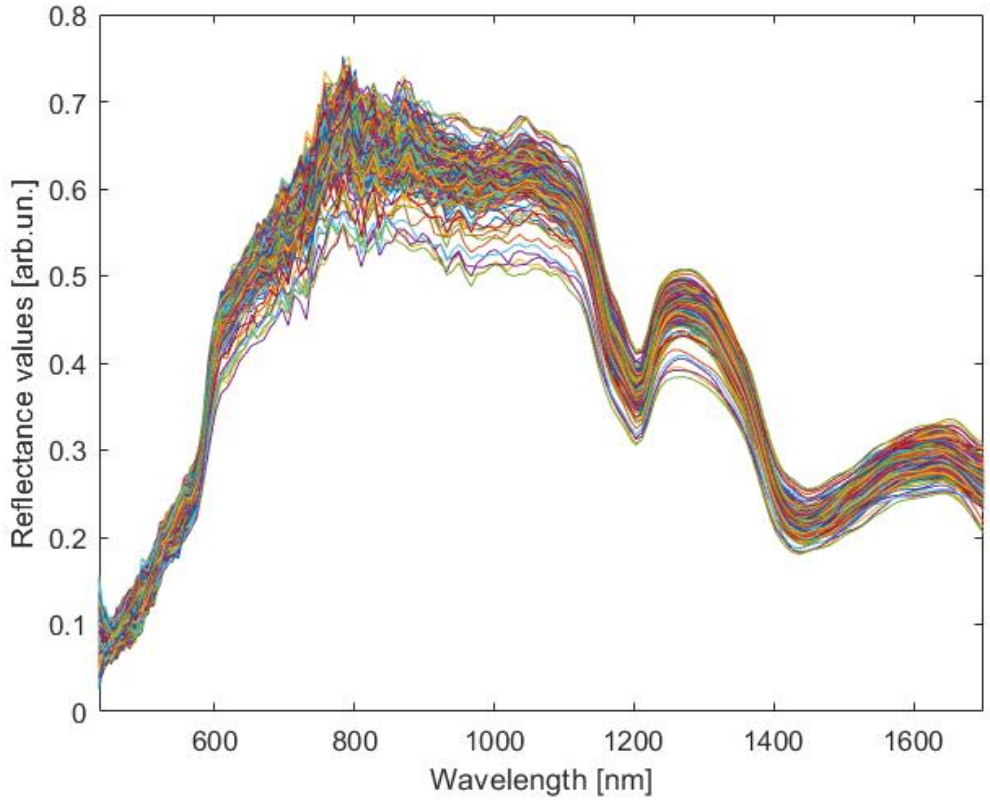
Spectral range $= 400\ -1700\ nm$

Collection of light reflected from the surface of ex vivo breast cancerous tissue with VIS-SWIR hyperspectral camera equipped with macro lens

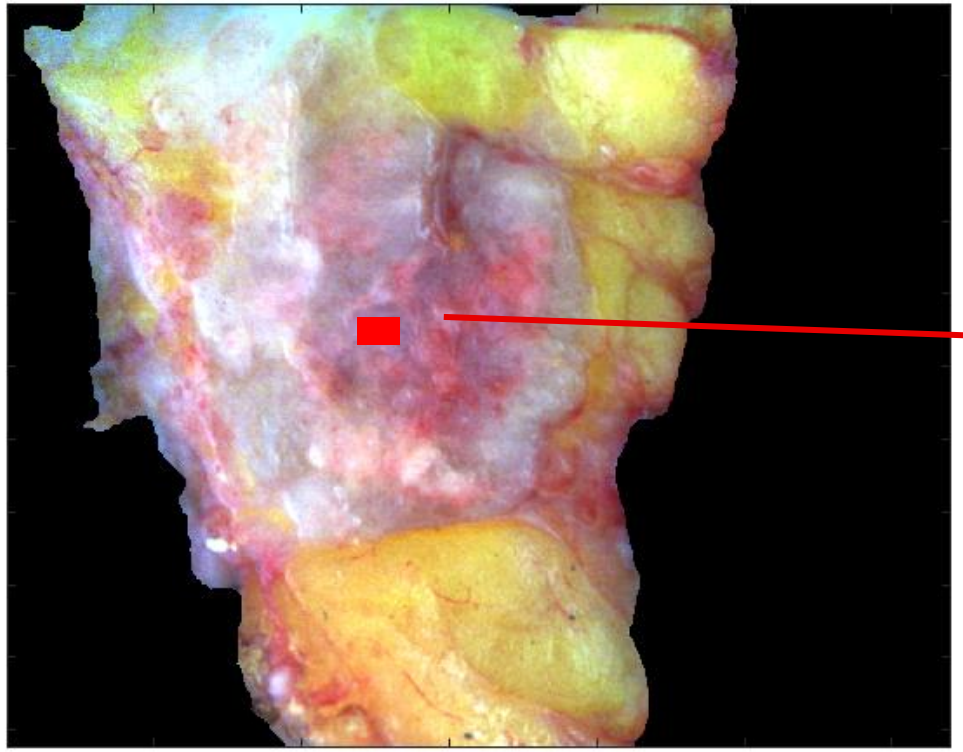
Samples are inserted inside a black box with uniform illumination and imaged in about 1 min



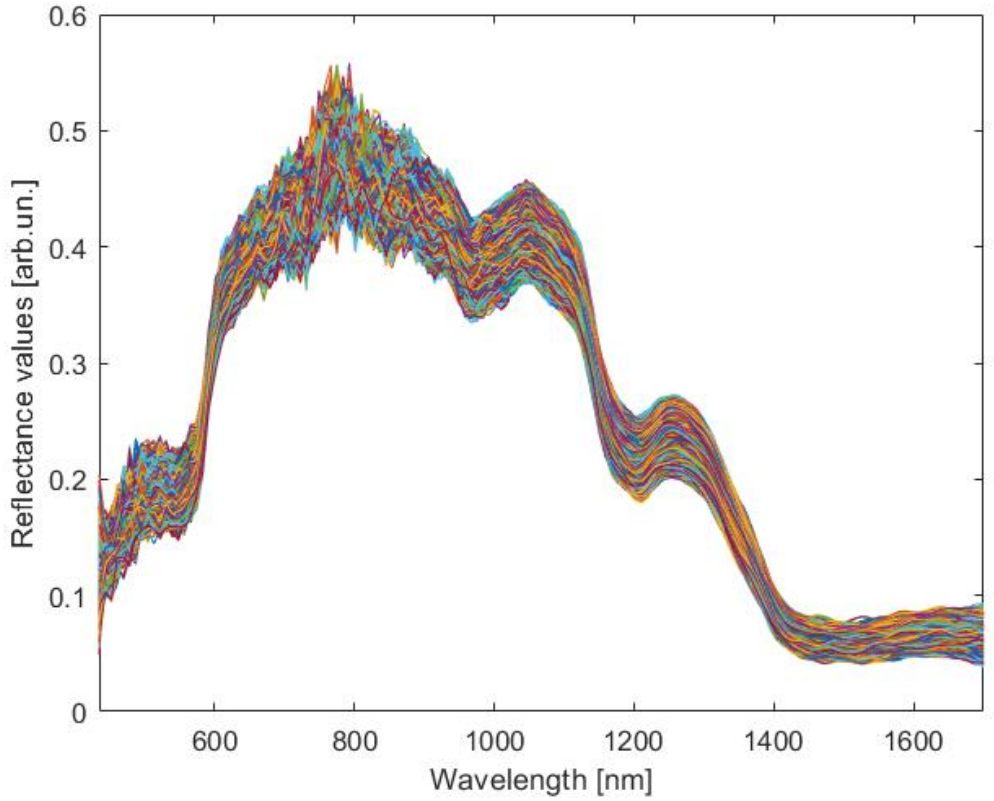
Reflectance spectra sampled from fatty region



By overlapping three grayscale images in the red, green and blue bands, a realistic RGB reconstructed picture is obtained (background was masked away)



Reflectance spectra sampled from cancerous region



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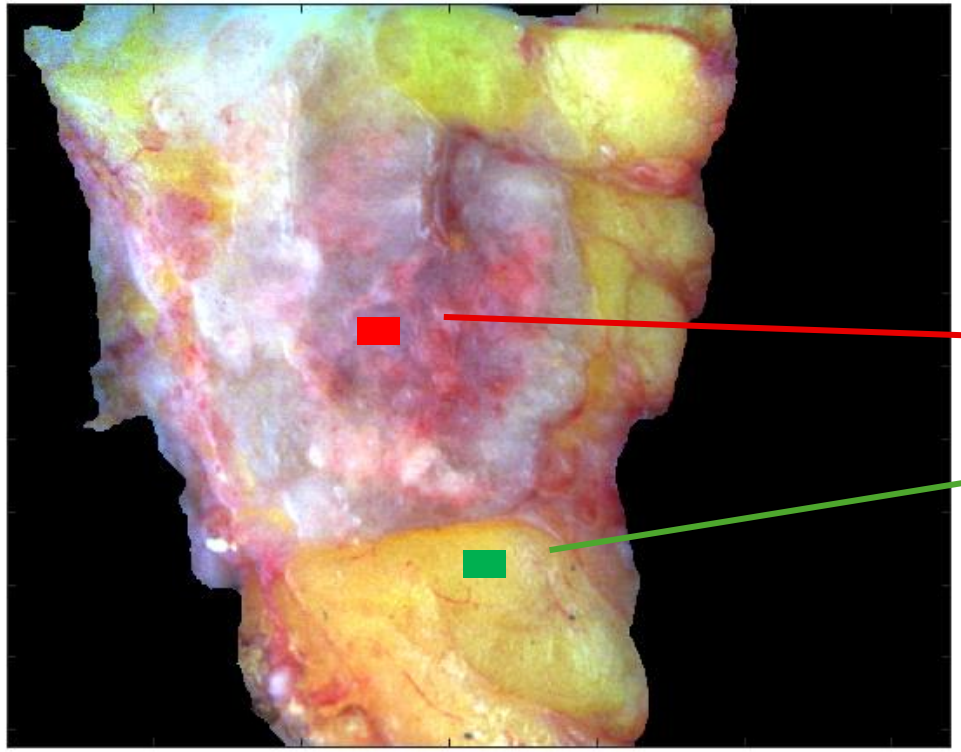
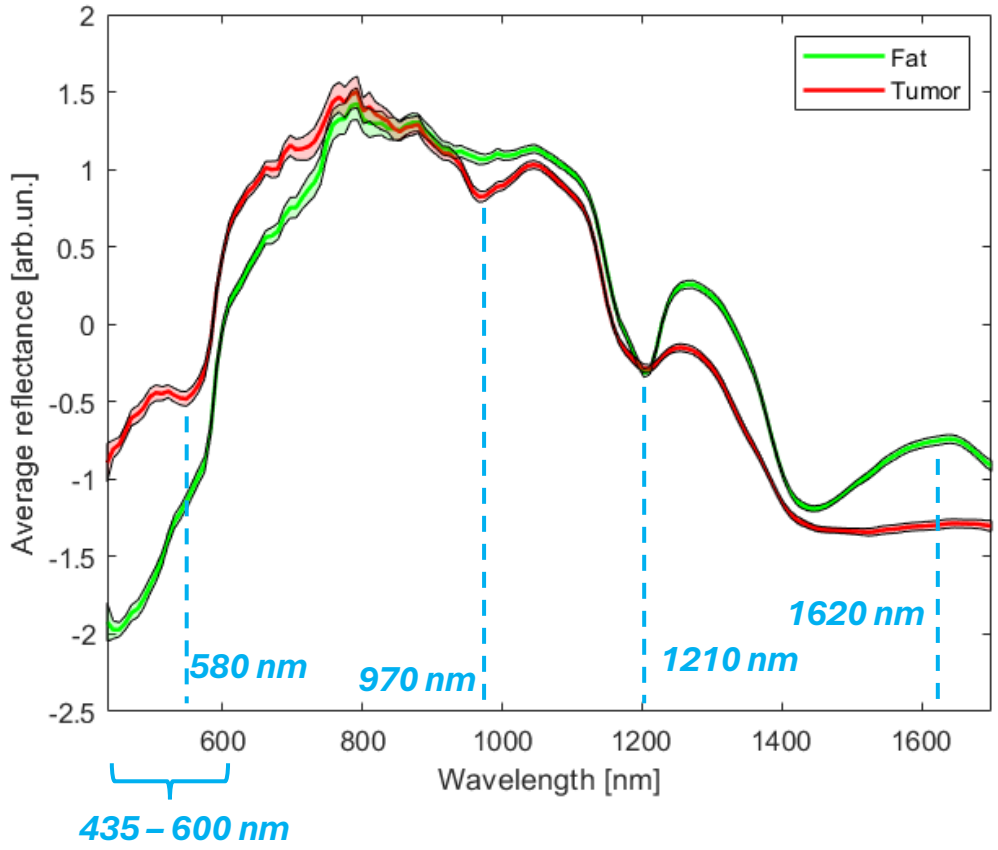


Average SNV spectra of fatty and cancerous regions



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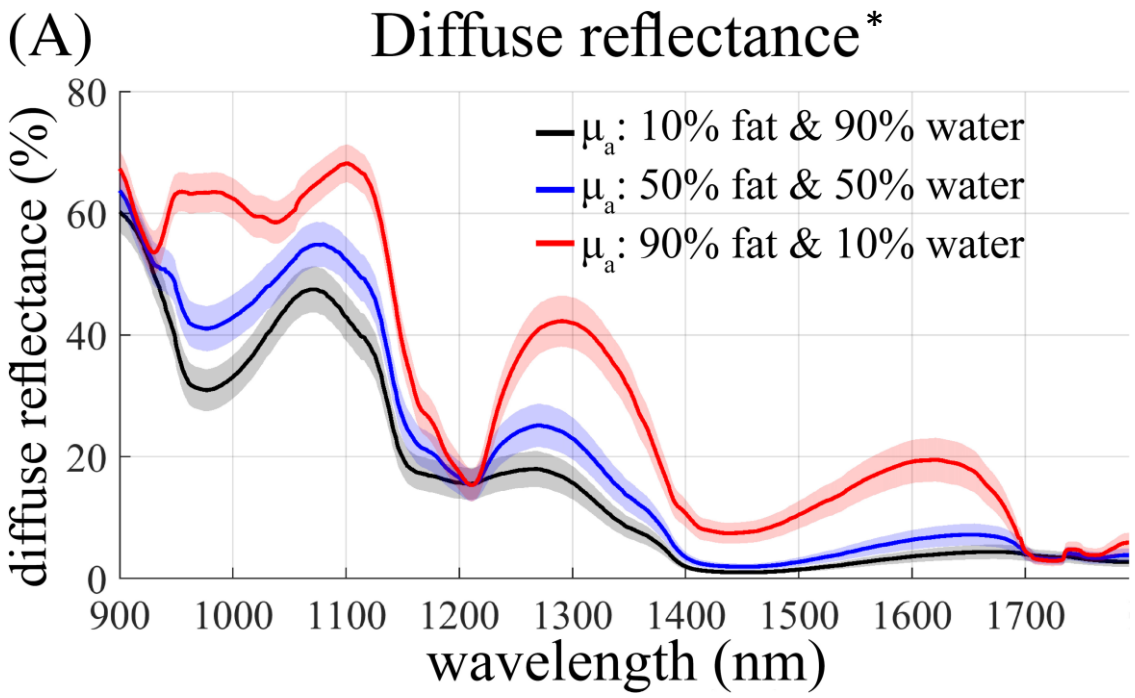
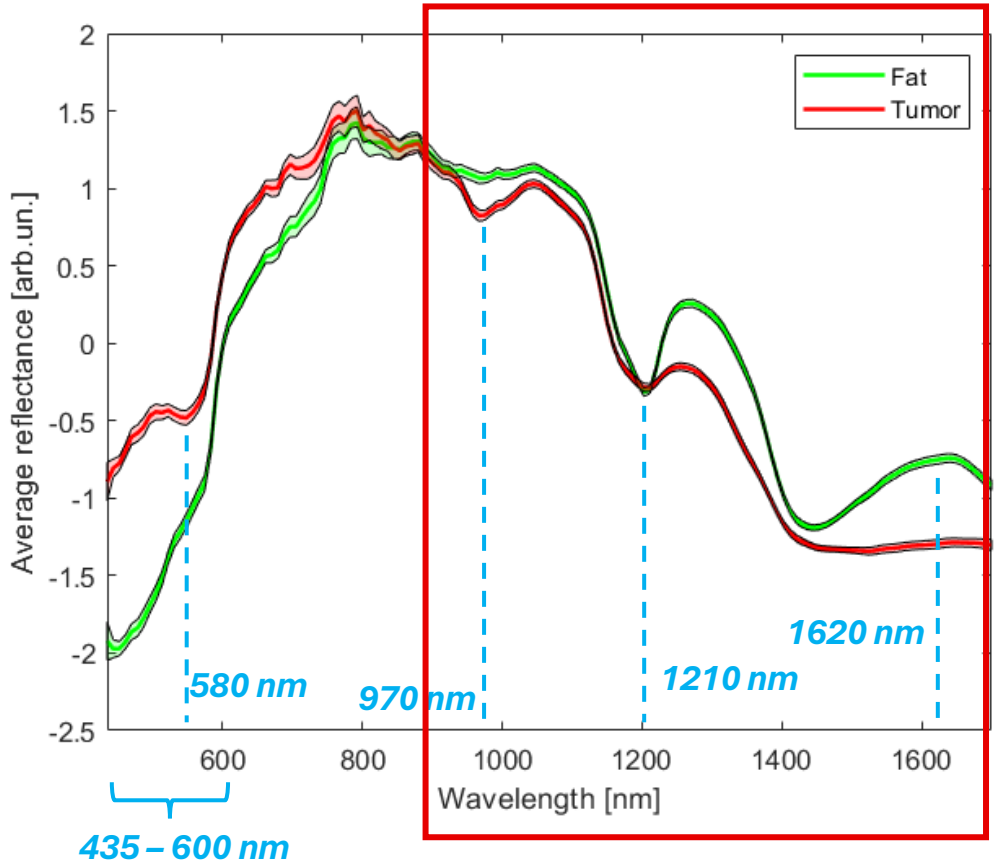


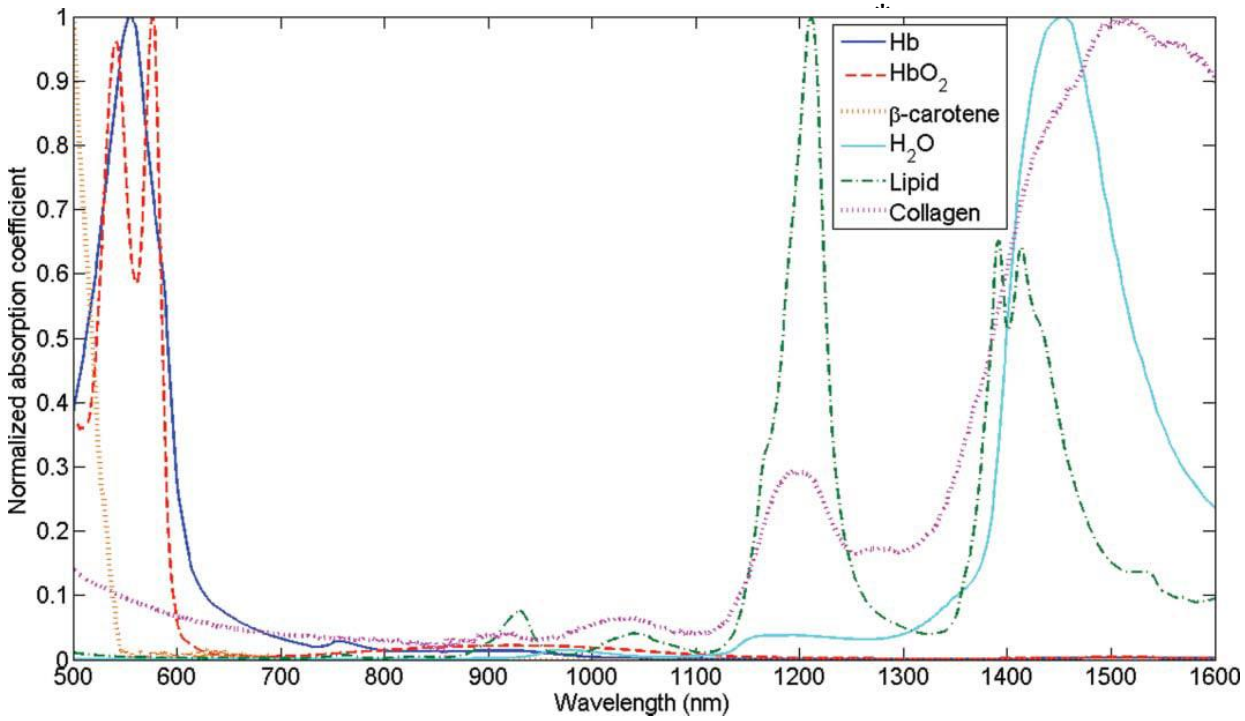
Average SNV spectra of fatty and cancerous regions



*Kho et Al., Journal of BIOphotonics, 9 july 2019

https://doi.org/10.1002/jbio.201900086

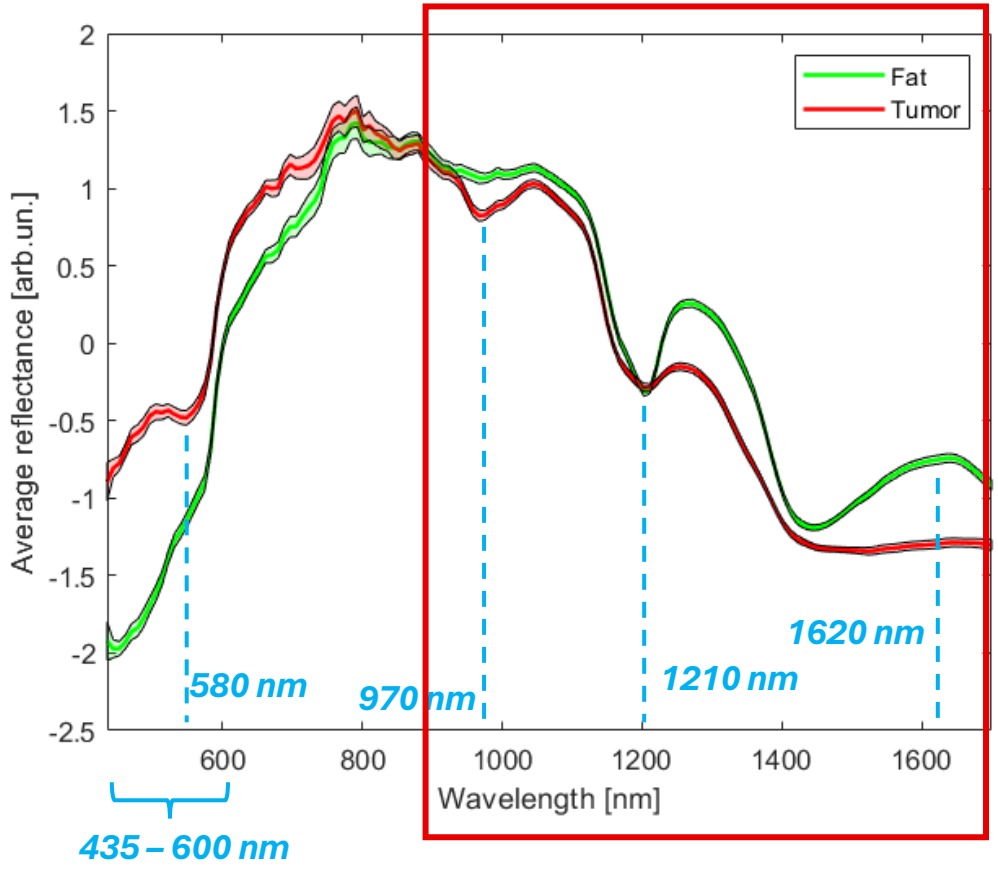
Normalized absorption of main chromophores in breast tissue*

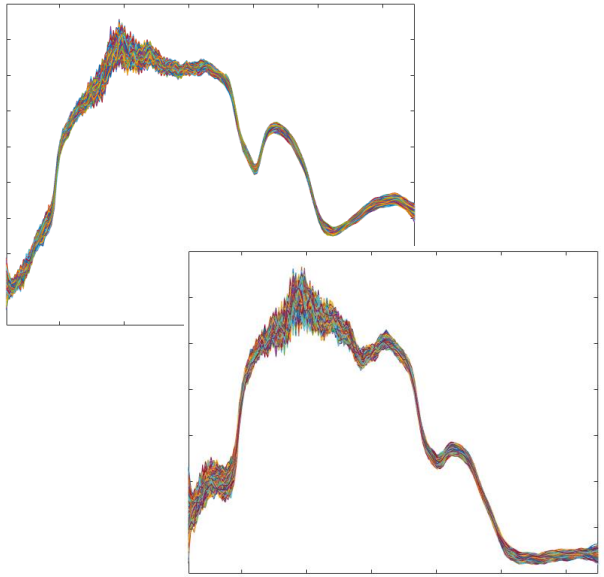


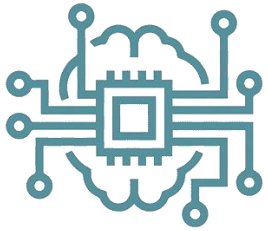
*Nachabe et Al., Journal of Biomedical Optics 16(8), 087010 (August 2011)

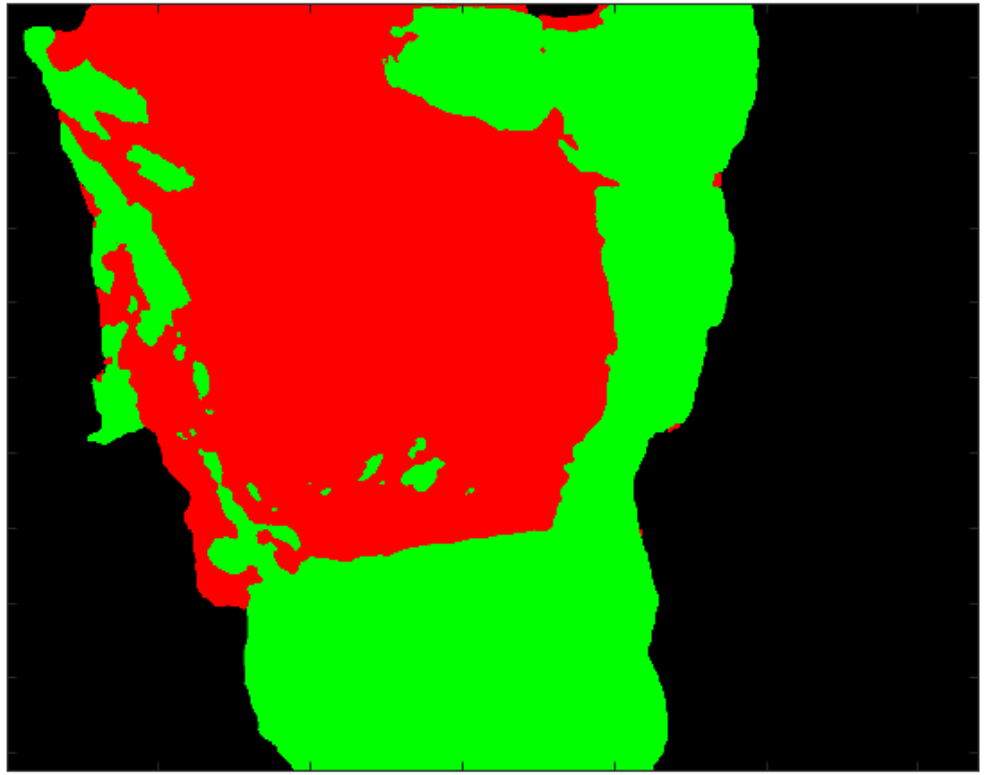
DOI: 10.1117/1.3611010

Average SNV spectra of fatty and cancerous regions





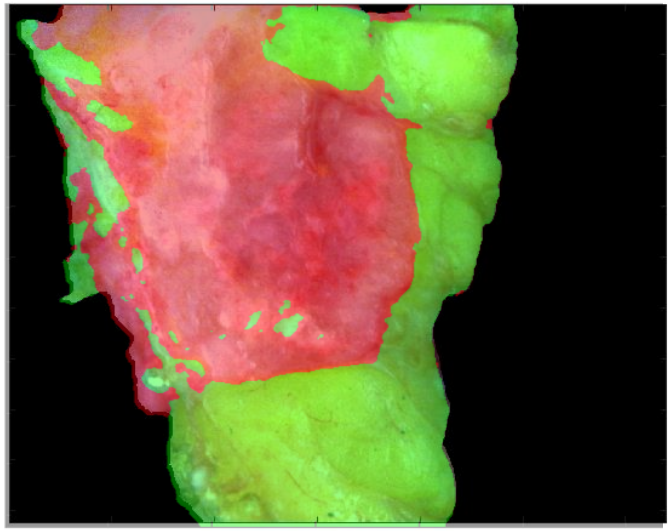




Traning of a fine tree algorithm on spectra of fatty and cancerous regions

Output **classification** (tumor in red, fat in green)

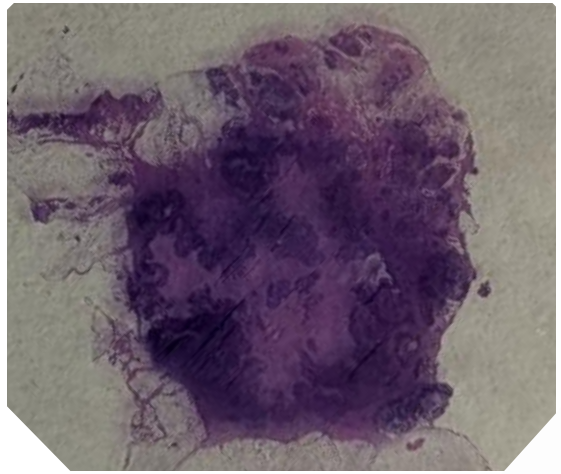
Classified image



Comparison

Evaluate sensitivity, specificity, accuracy, …

Hematoxylin and eosin (H&E)



N.B. Digital H&E-stained image is still being worked on, necessary for best image registration

How can we improve?

Upscale the protocol *to tens (or hundreds…) of patients to make classification clinically significant*

Technical improvements:

- *Improve stability of the system*
- *Improve spectral coverage in the NIR region*
- *Improve uniformity to prevent specular reflection*
- *Standardize the size of the measured region*

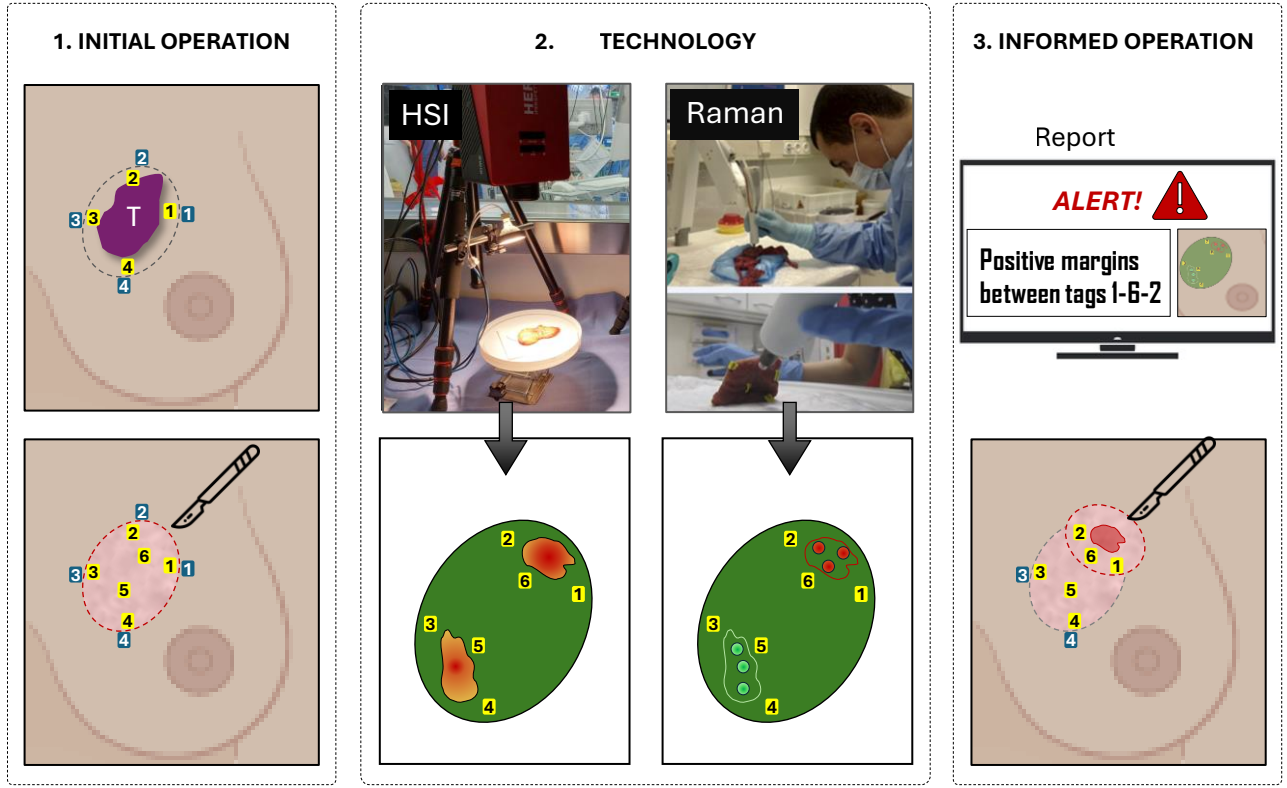
Explore ***different models*** *(KNN, Neural networks,…)*

Project: 101187508 — Spectra- BREAST — HORIZON-EIC-2024-PATHFINDEROPEN-01

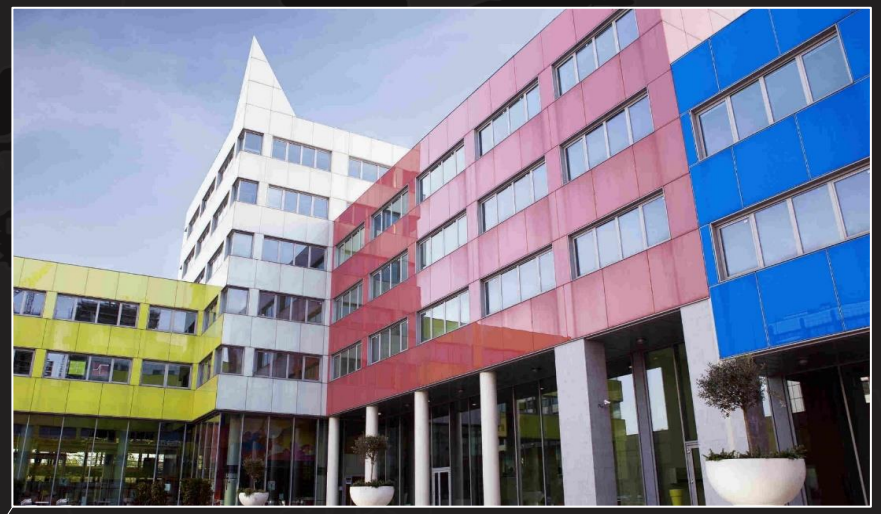
Co-funded by the European Union

Maugeri

POLITECNICO MILANO 1863



Thanks for the attention



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