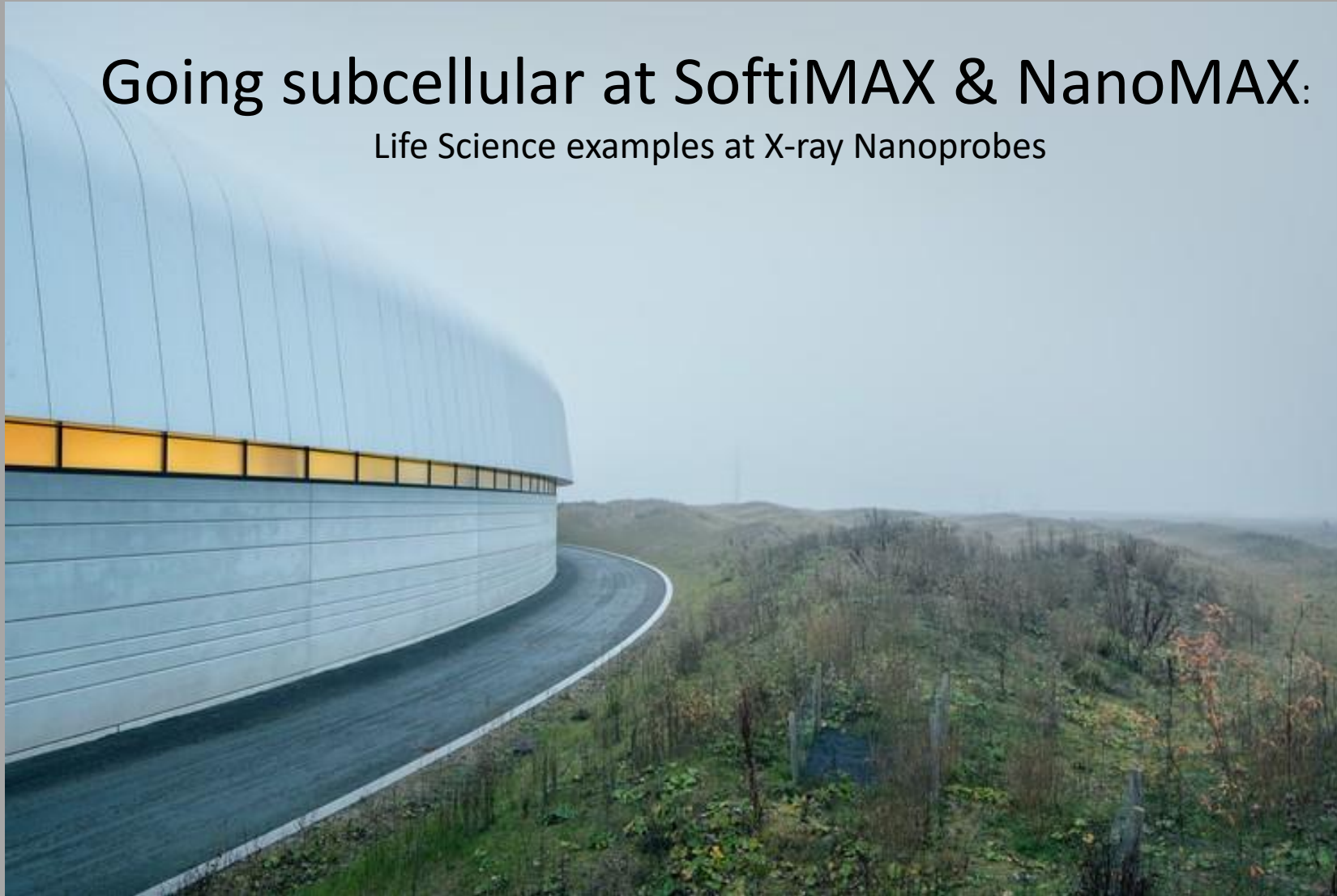


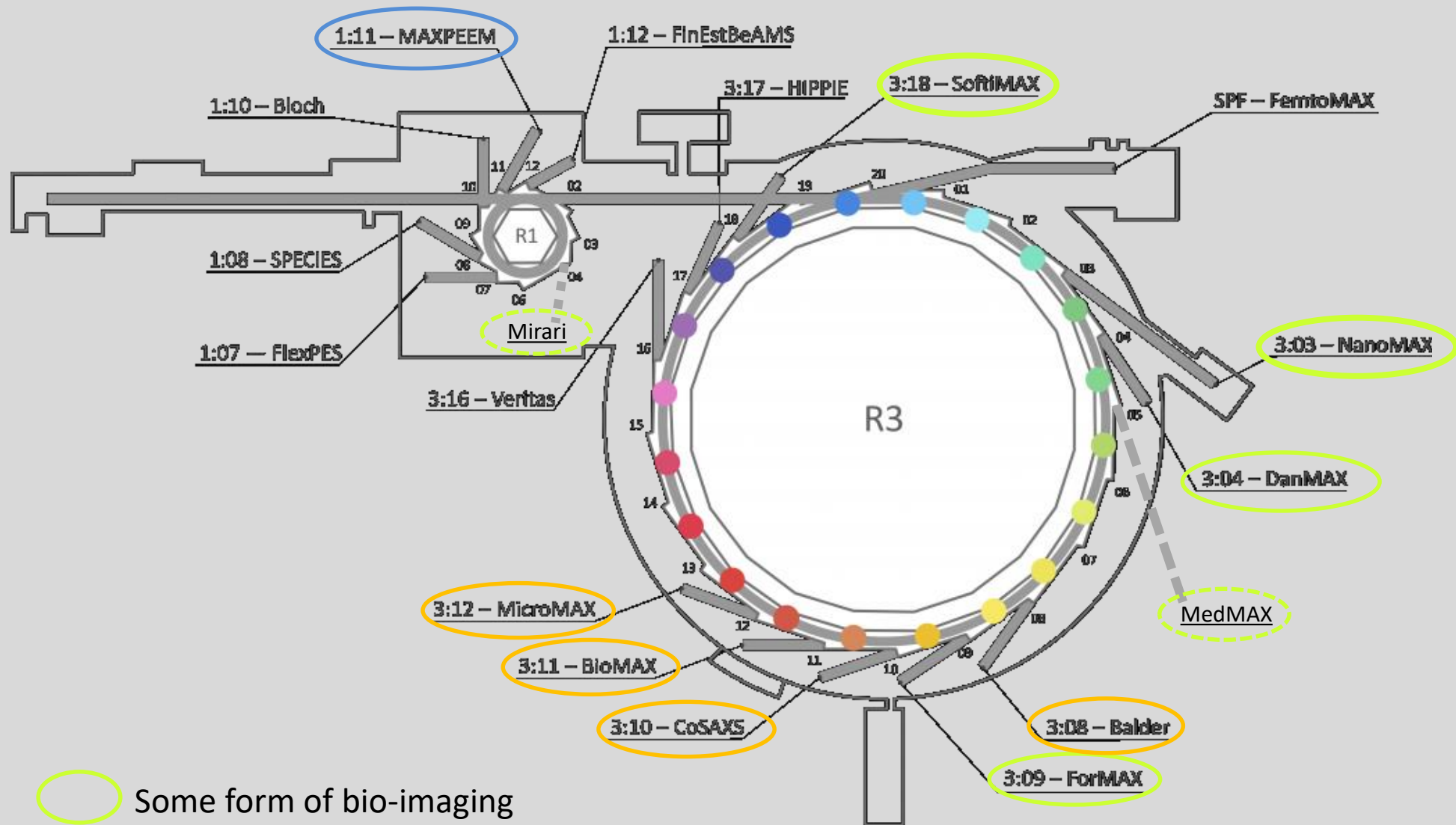
# Going subcellular at SoftiMAX & NanoMAX:




Life Science examples at X-ray Nanoprobes



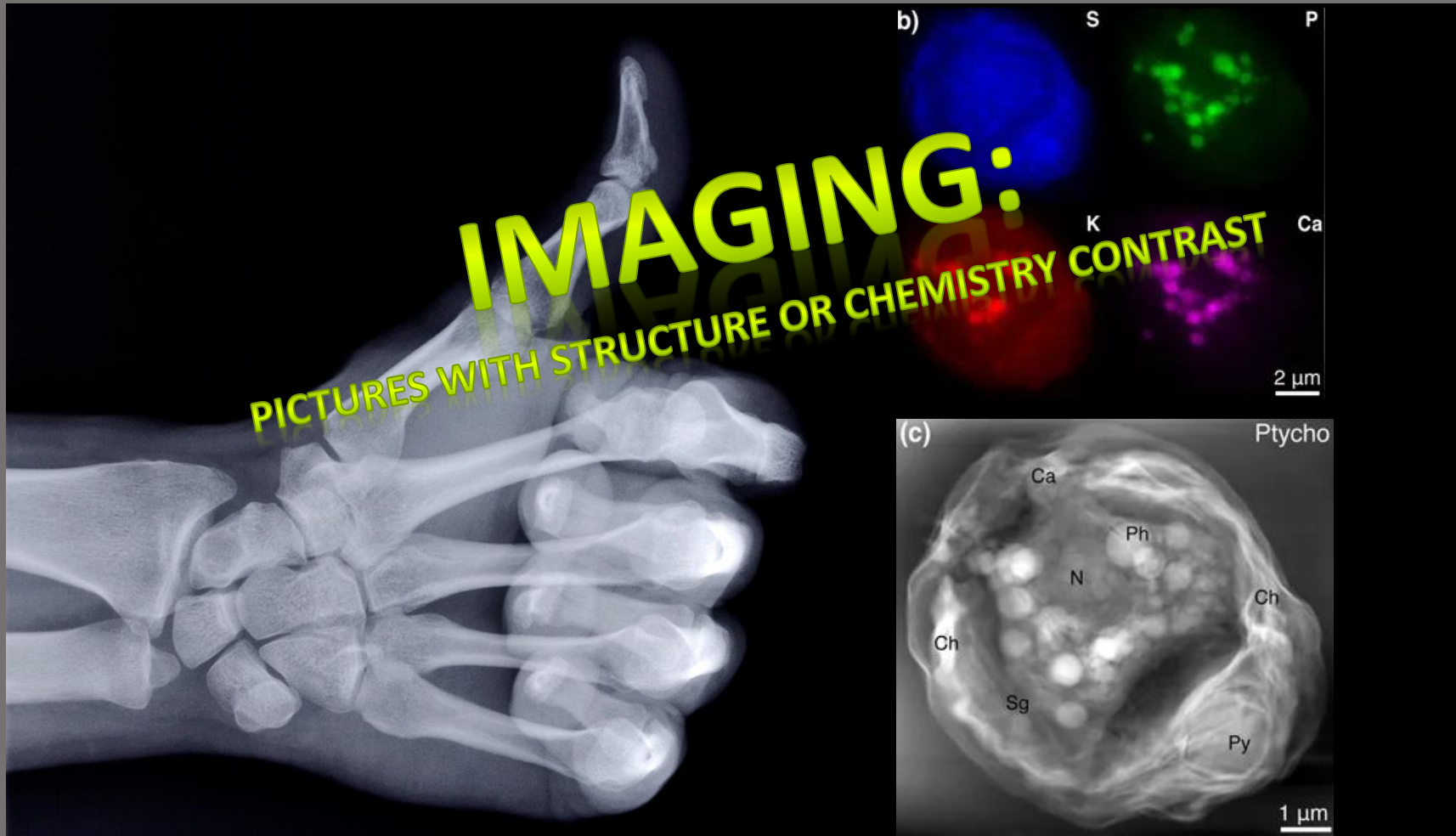
Karina Thånell

# Which beamlines?



-  Some form of bio-imaging
-  Also *bio*, but not imaging
-  Also *imaging*, but no *bio*

# IMAGING: PICTURES WITH STRUCTURE OR CHEMISTRY CONTRAST



# Flavours of imaging at MAX IV

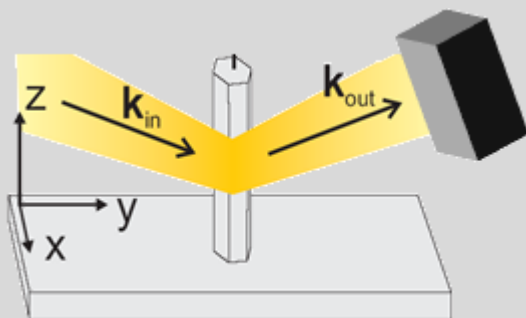
**Direct imaging:** spatial resolution is determined by the x-ray probe size:

- Nano-diffraction mapping
- X-ray Fluorescence mapping → NanoMAX
- Scanning Transmission X-ray Microscopy → SoftiMAX
- IR spectromicroscopy

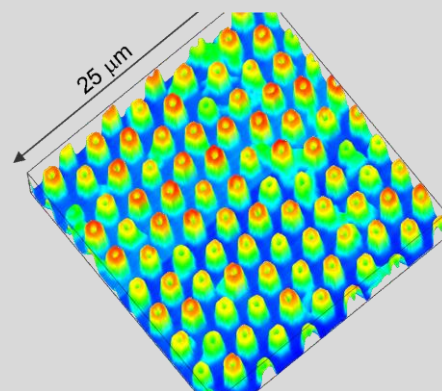
**Indirect imaging:** scattering or diffraction-based contrast is used, and 'translated' into a real space image (usually more about structure):

- Phase contrast imaging/tomography
- SAXS tensor tomography
- Ptychography (Bragg & Forward) & CDI

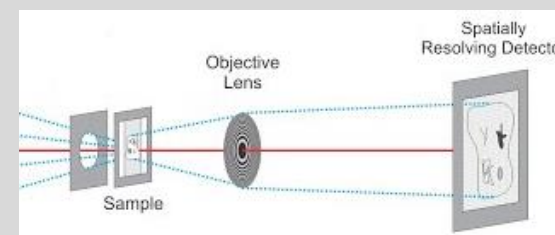
**Other:** - AFM-IR imaging, STM, SEM (+EDX), SPELEEM techniques



Single object [local probe]

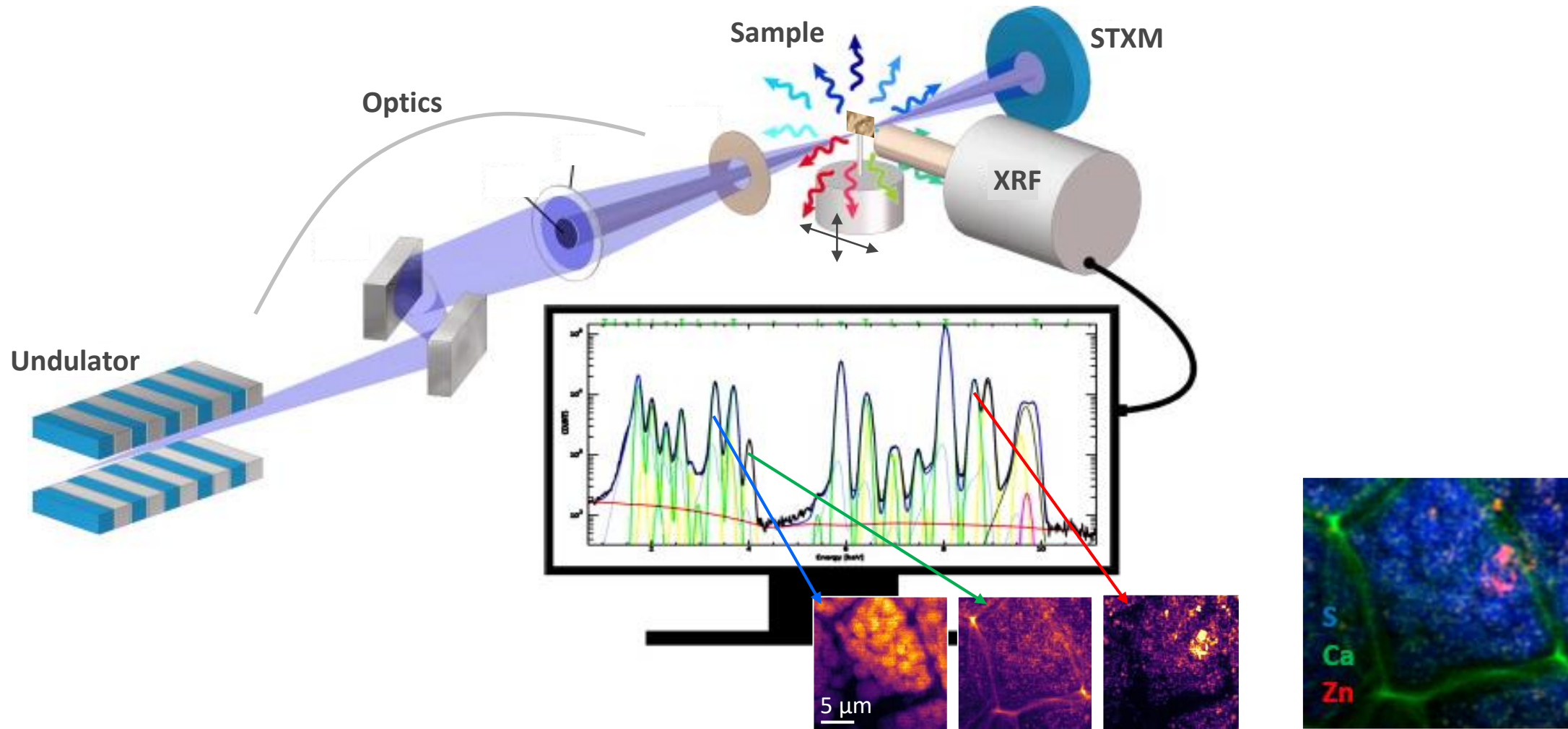


Map [scanning probe]



Full-field [resolving detector]

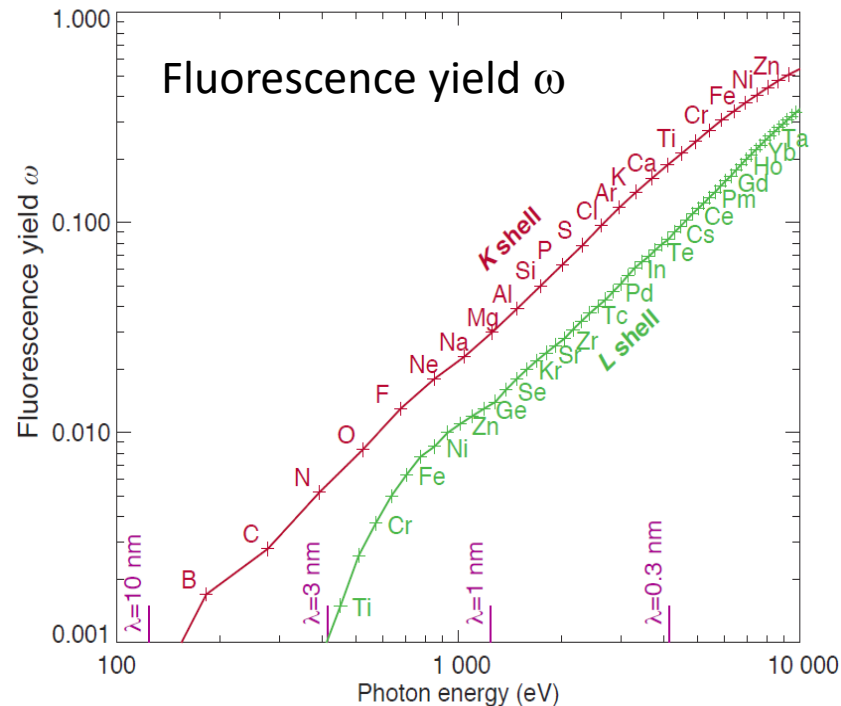
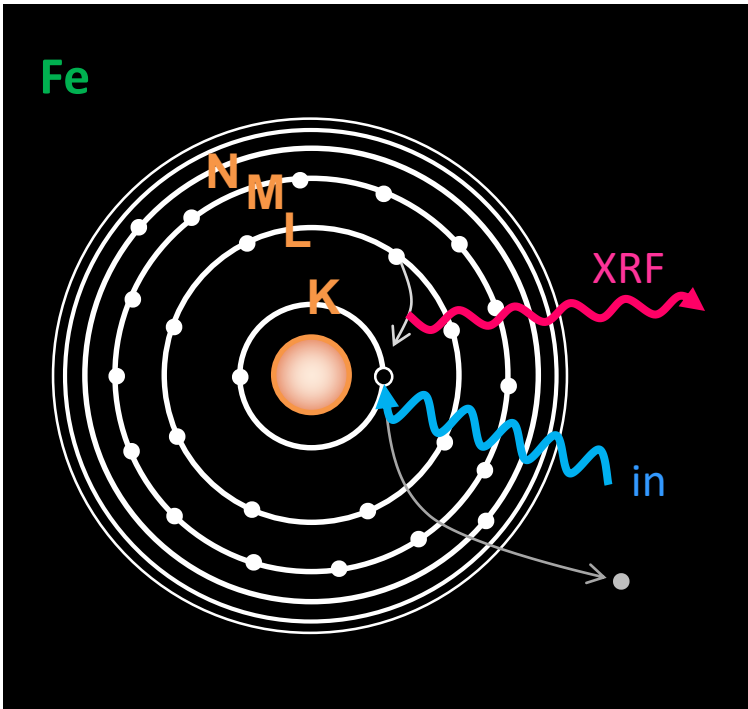
# Scanning X-ray nanoprobe principle



# X-ray Fluorescence

/eks-rey flʊə'res(ə)ns, flɔ:'res(ə)ns/

X-ray Fluorescence is the emission of **characteristic**, secondary X-rays by a material that has absorbed (more energetic) X-rays, or other electromagnetic radiation. The emitted light corresponds to an energy difference between two atomic levels and its wavelength is unique for each element.



## XRF

- Hard X-rays = better
- No need for labelling: it's element specific!
- Forget about the low Z elements...
- XRF sensitivity: down to  $\approx 1-100 \mu\text{g/kg}$

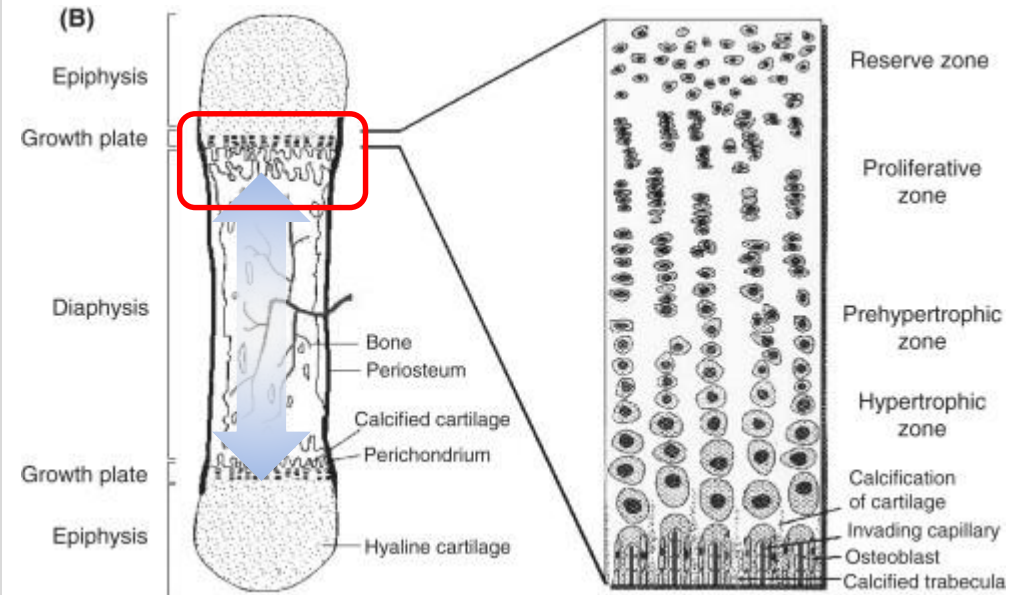
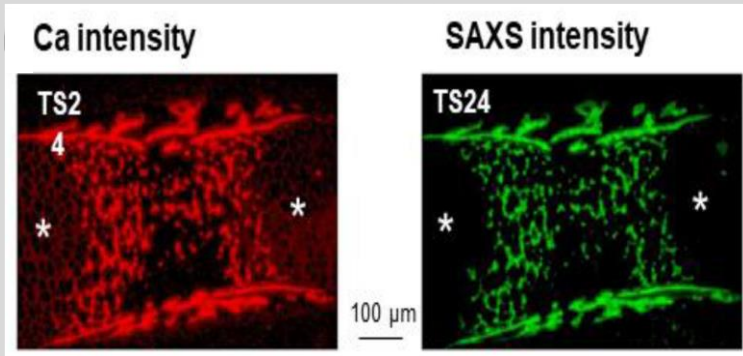
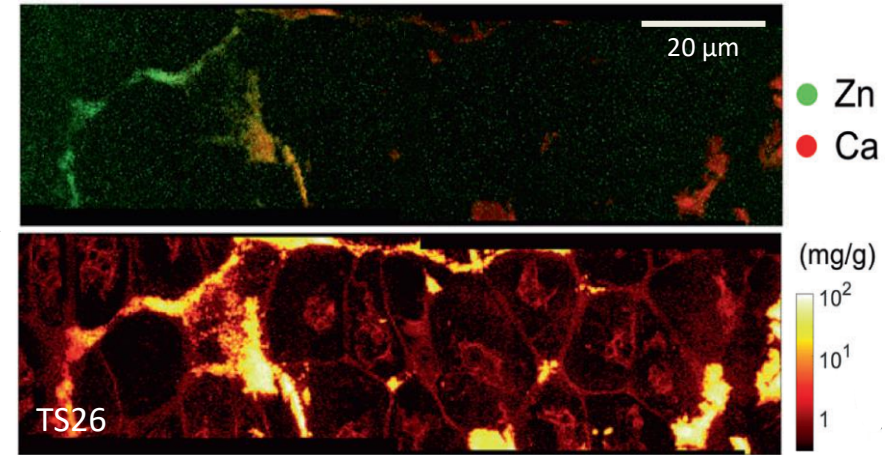
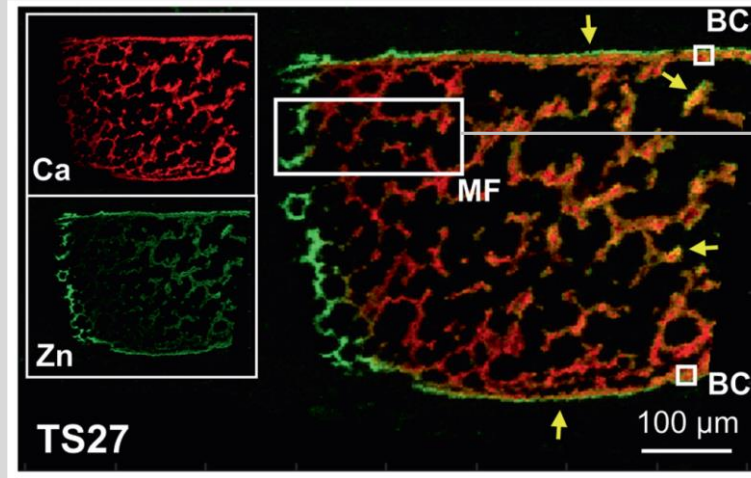
## NANO MAX

- Photon Energy 5-28 keV
- Spotsize 30-200 nm
- Sample thickness  $< 10 \mu\text{m}$
- Samples in air/He/vacuum
- Cooling? Next year...

# Embryonic bone mineralization

I. Silva Barreto, D. B. Raina, H. Dejea I Velardo, H. Isaksson - LU

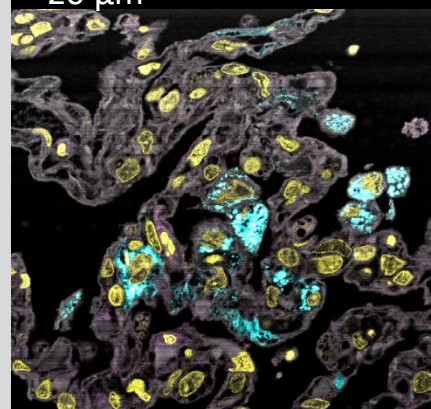
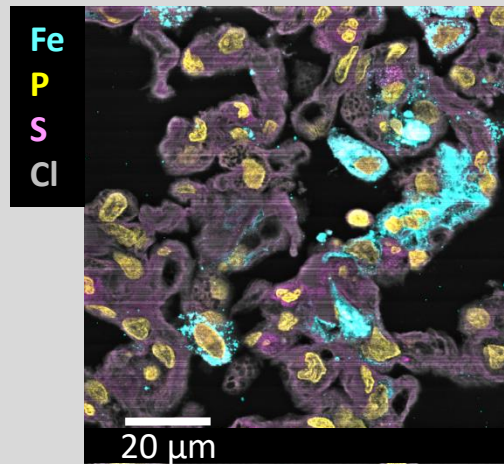
- Zn @ leading edge of Mineralization Front, Ca follows
- Ca concentration in bone center increases during bone maturation
- Non-ordered Ca deposits: not hydroxyapatite before bone formation



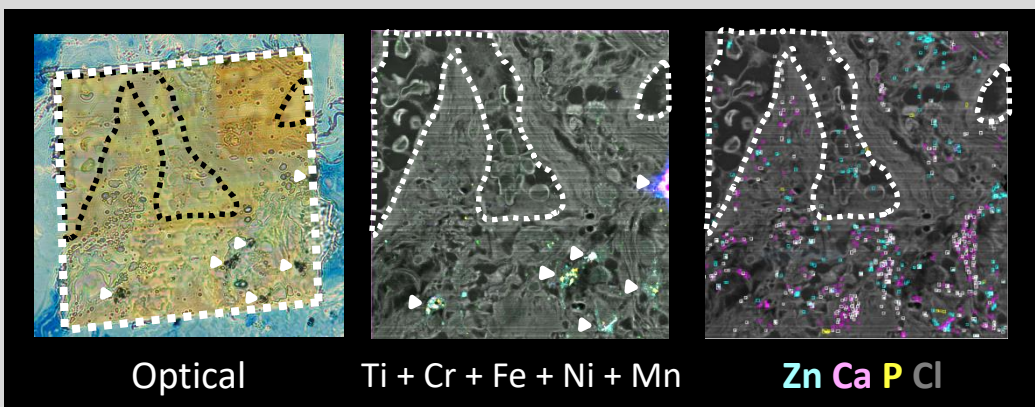
# Idiopathic pulmonary fibrosis (IPF)

*Bryan Falcones Olarte, M. Kahnt, L. Elowsson, K. Thånell, G. Westergren-Thorsson*

IPF: a fatal chronic lung disease accompanied by impaired breathing and fatigue. Some studies have found that elements such as Calcium (Ca), Zinc (Zn) and/or Iron (Fe) are important during the development and pathophysiology of IPF, and display a dysregulated metabolism.

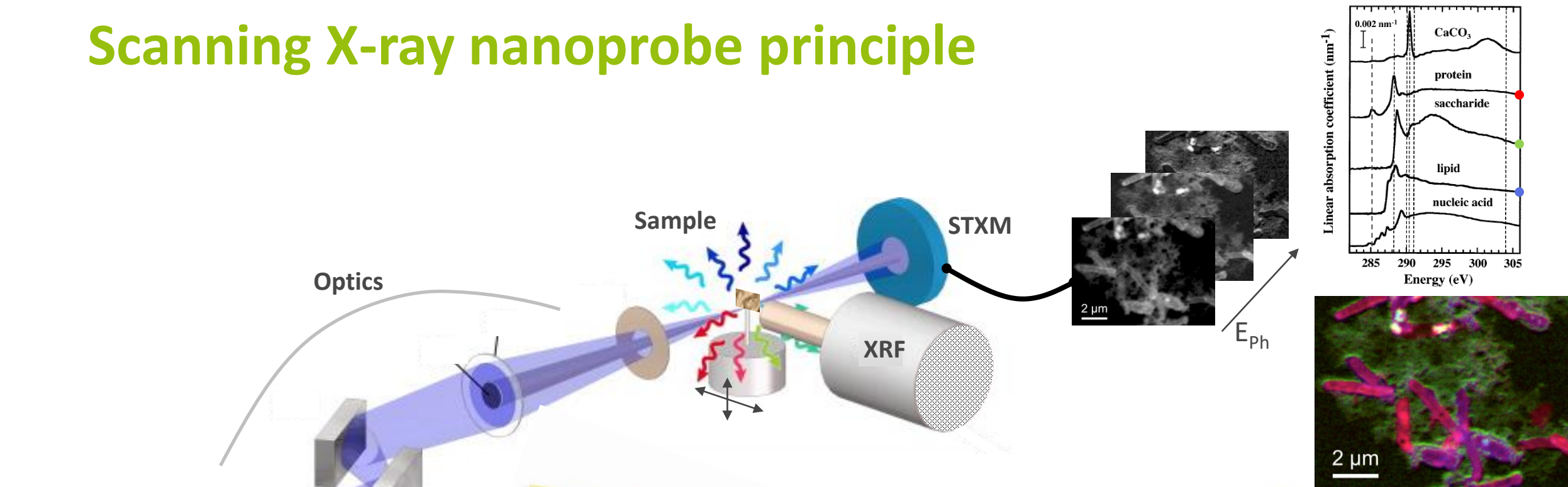


Unpublished!





# Scanning X-ray nanoprobe principle



SoftiMAX

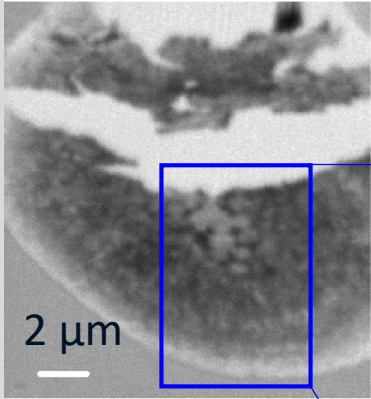
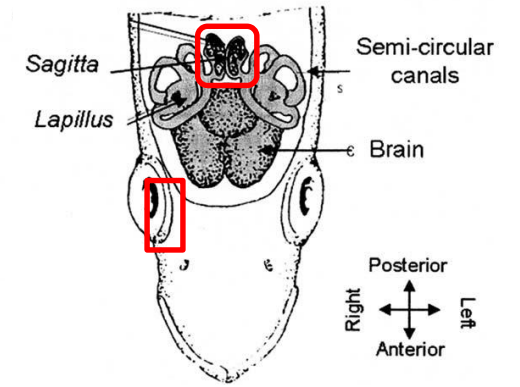
- Ph. Energy 275-2500 eV
- Spotsize 20-100 nm
- Sample thickness: 100nm to a few  $\mu\text{m}$ .
- Samples in vacuum/He/cell
- Cooling? Next year...

STXM

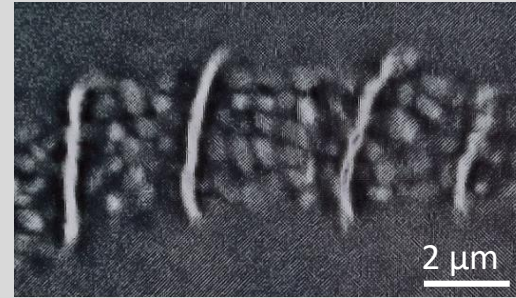
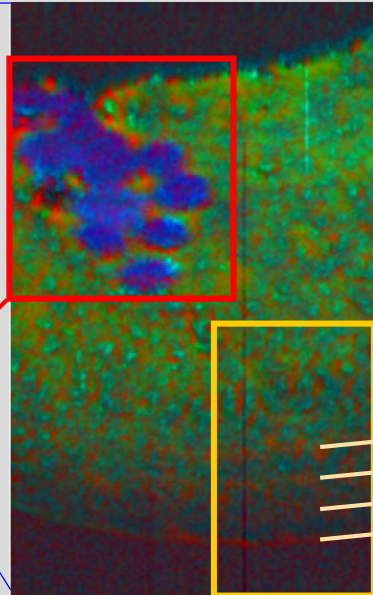
- Absorption contrast: soft x-rays = good for chemistry
- Still no labelling needed
- Yes! Low Z elements too!
- Sensitivity limit: 0.1-1%
- Quantifiable (Beer's Law)

# Otolith (CaCO<sub>3</sub> fish ear bone) in 6d old cod larva

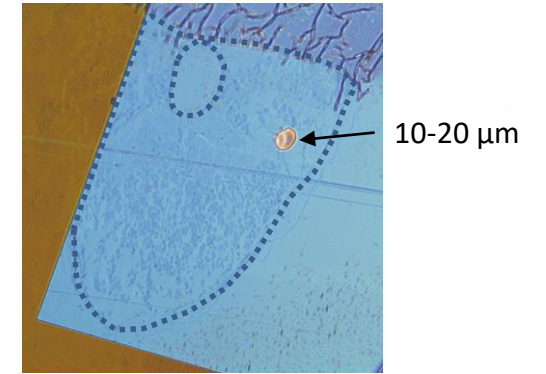
Yvette Heimbrand, K. Limburg (SLU); B. Falcones Olarte, K. Thånell (LU), AP Hitchcock



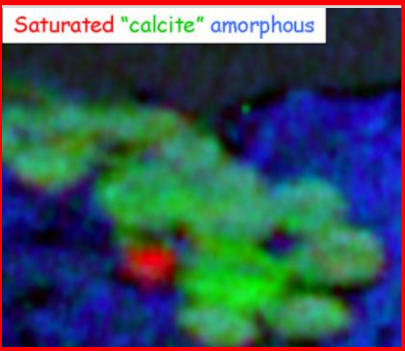
amorph calcite residu



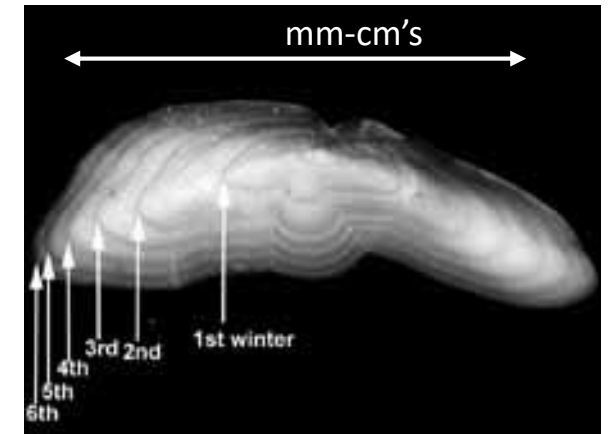
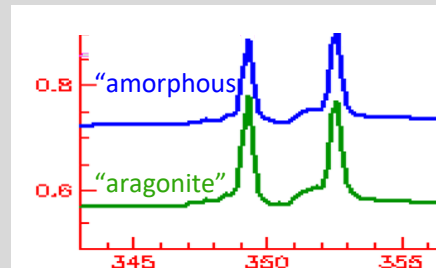
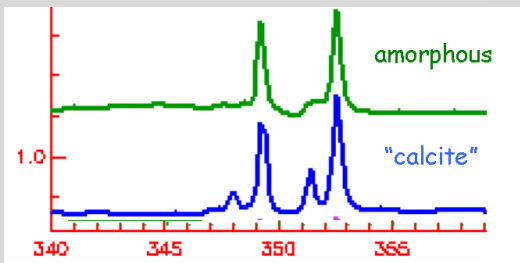
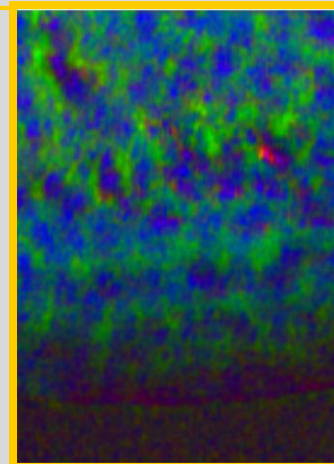
Bonus: edge of the eye



CORE



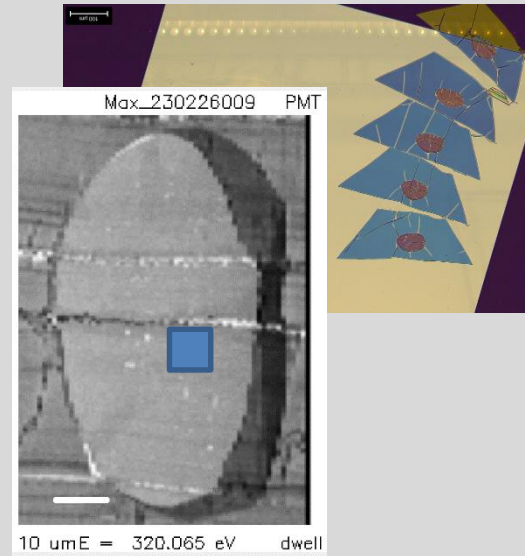
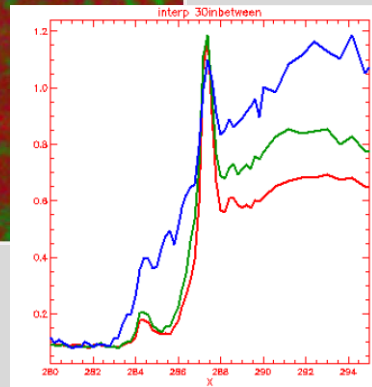
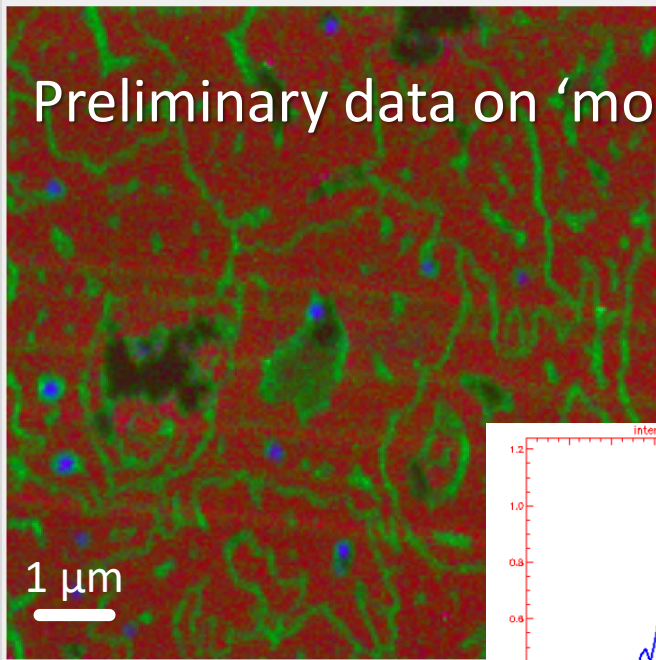
GROWTH EDGE



# Splitting hairs

Karina Thånell, B. Falcones Olarte (LU), H. Druid, B. Uusma (KI)

Preliminary data on 'modern' hair



What can be found in the hair?  
 Chemical compounds (drugs)? ≈  
 Elemental: Hg, Pb → NanoMAX,  
 Balder

- Melanin; melanosome (indole derivatives)
- Cell membrane complex: lipid bilayers
- Cortex: keratin & other polypeptides

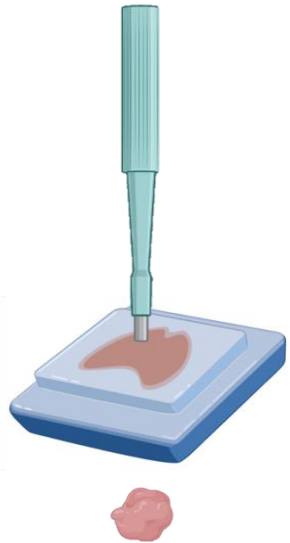
## Andrée Polar expedition 1897



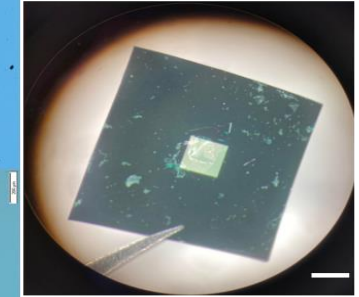
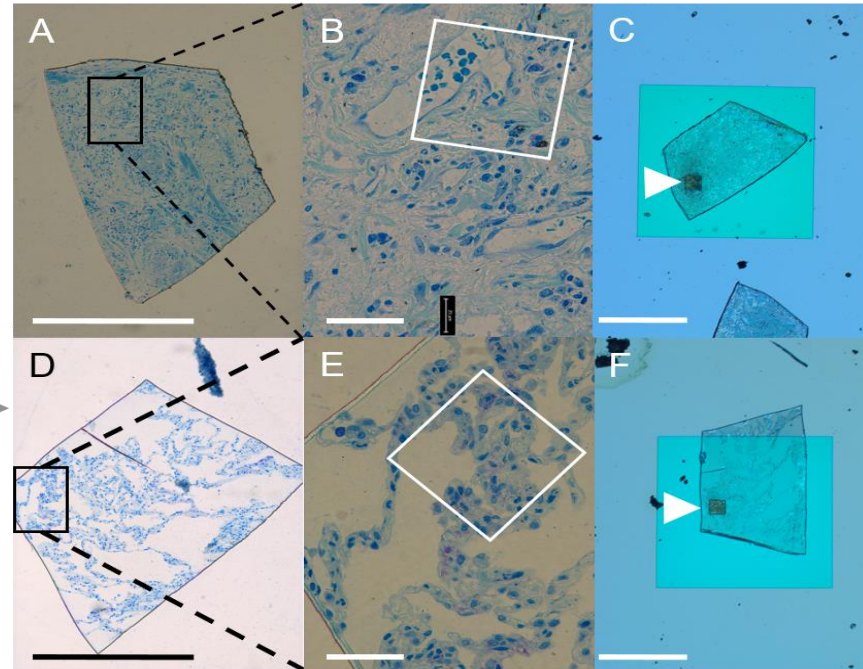
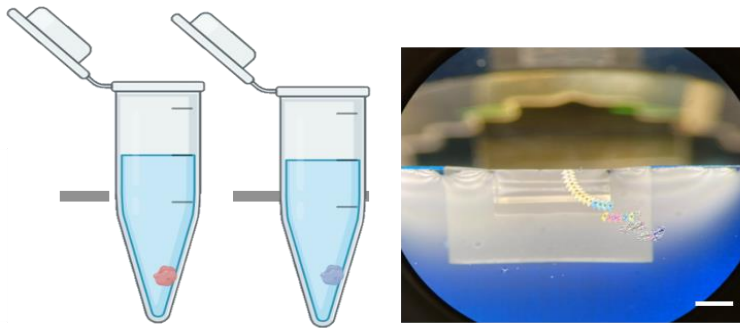
Bea's quest:  
 what killed Andree?

# Sample preparation

Punch & Fixate

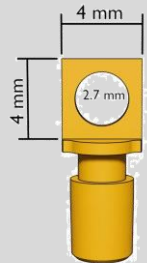


(Re)embedding and sectioning

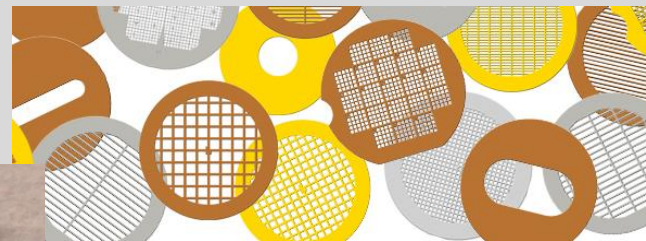
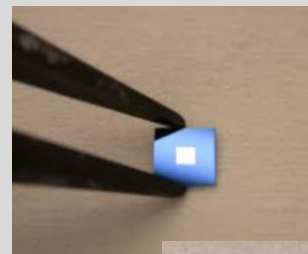


NanoMAX

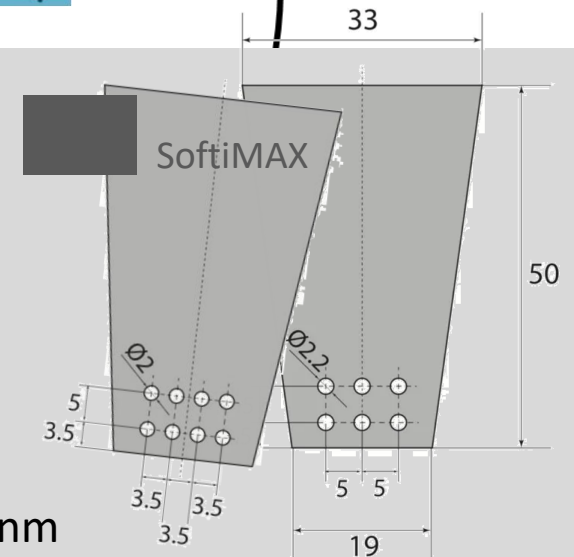
pointy



$\text{Si}_3\text{N}_4$ : 0.2-1  $\mu\text{m}$

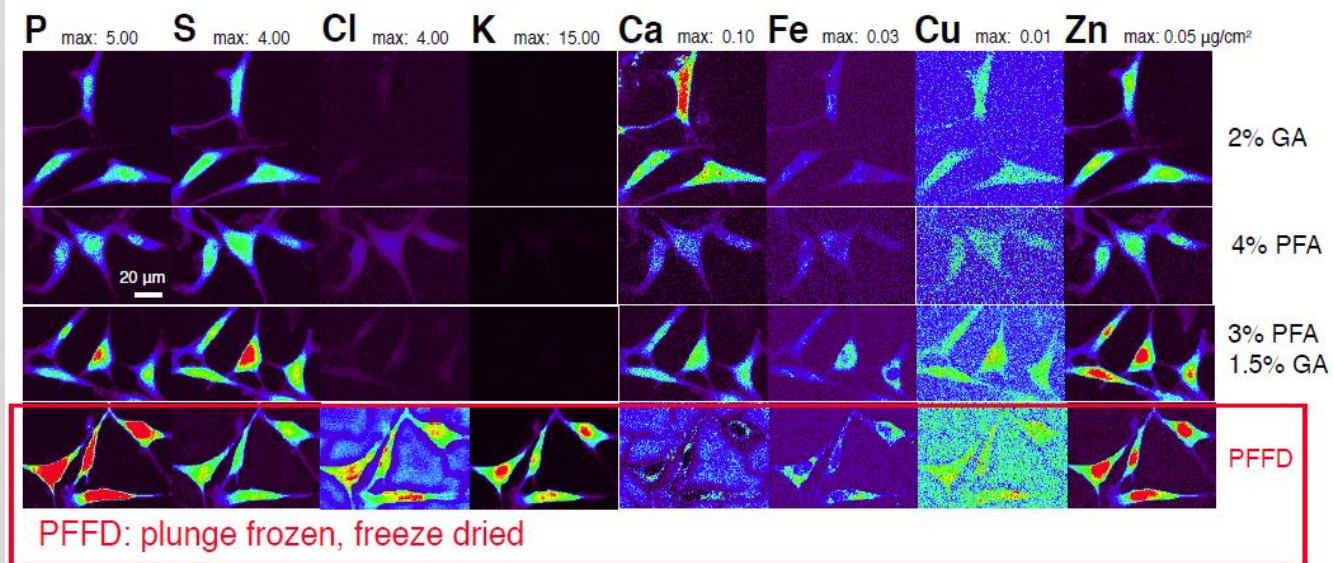


$\text{Si}_3\text{N}_4$ : 50-100 nm



# ...and finally

## Cryo preservation keeps chemistry intact



- Jin, Paunesku, Lai, Gleber, Chen, Finney, Vine, Vogt, Woloschak, and Jacobsen, *J. Microscopy* **265**, 81 (2017).
- See also Perrin, Carmona, Roudeau, and Ortega, *J. Analyt. Atom. Spectr.* **30**, 2525 (2015).

NanoMAX: maik.kahnt@maxiv.lu.se  
ulf.johansson@maxiv.lu.se  
SoftiMAX: karina.thanell@maxiv.lu.se  
jorg.schwenke@maxiv.lu.se

Data analysis software:  
XRF: PyMCA  
STXM: Axis2000, Mantis, (Quasar)  
Jupyter Notebooks

### MAX IV:

- Twice/year call for proposals
- Feb & Aug, apply per BL
- max. 3 pg. for 2-6 day beam
- judged on scientific merit
- Free of charge = publish
- Also: "Fast Access", 1 day max.
- ↳ apply throughout the season

Before an experiment, think about:

- 1) Resolution needed
- 2) Information needed: contrast
- 3) Beam damage & radiation dose
- 4) Sample preparation!!!
- 5) Combine techniques?
- 6) Contact us!