# A novel MeV-ion beam microscope and lithography technology for biomedical research imaging (MIMMI)

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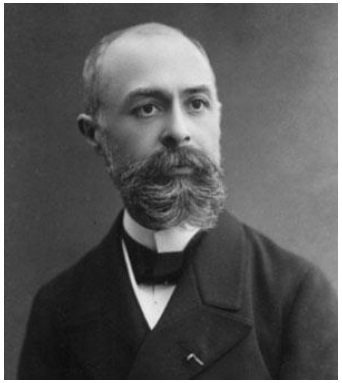
# The idea

Take three Nobel prize winning discoveries and mix with some inovations from Jyväslylä.....

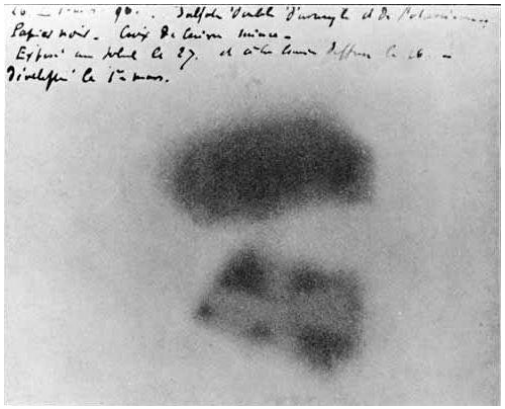
# The basis of MeV ion beam lithography

## Two Nobel Prize winning discoveries



H. Becquerel

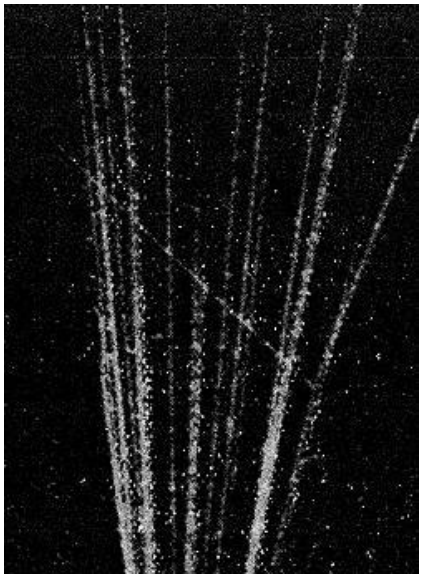
½ 1903 years Nobel Prize in Physics "*in recognition of the extraordinary services he has rendered by his discovery of spontaneous radioactivity*"



A. H. Becquerel, *Comptes Rendus* 122,(1896)420.

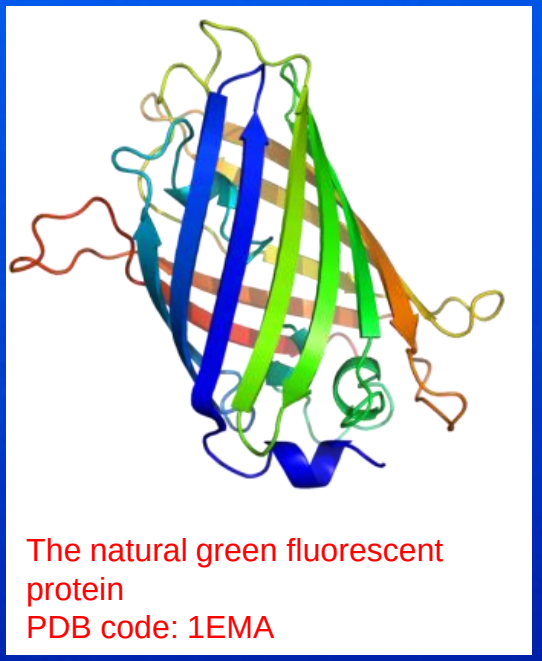
C.T.R Wilson

½ 1927 years Nobel Prize in Physics "*for his method of making the paths of electrically charged particles visible by condensation of vapour*"



P. Blackett (1925) Proc. Roy. Soc., A, vol. 107, Pl.6.

# 2008 Nobel Prize in Chemistry



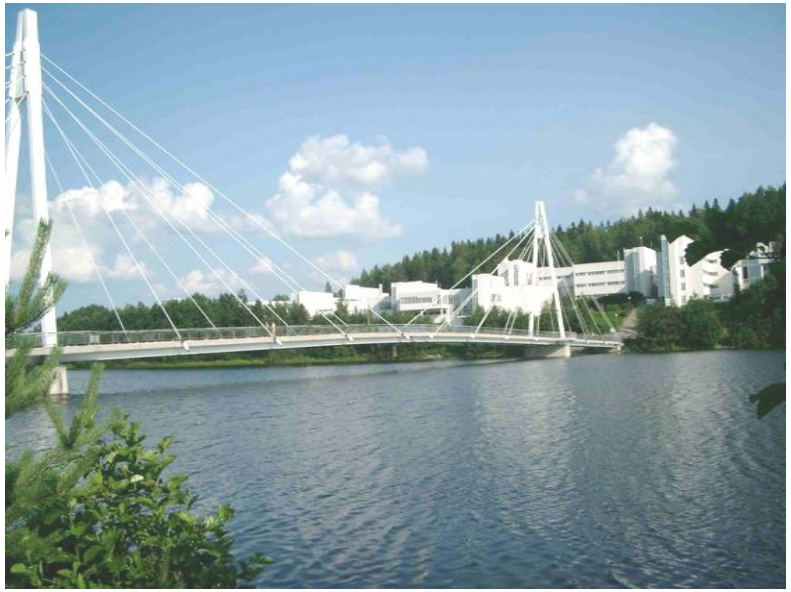
The natural green fluorescent protein
PDB code: 1EMA

***Aequorea victoria***
**From Wikpedia**

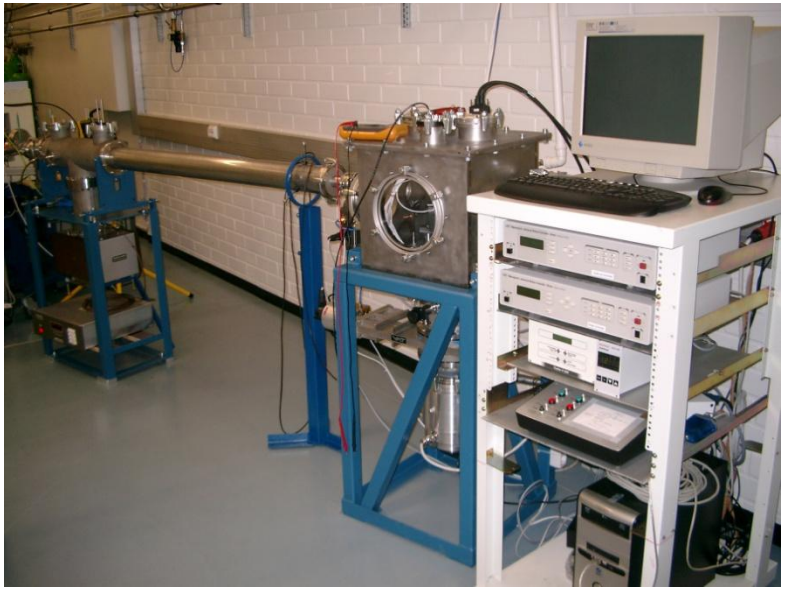
Osamu Shimomura , Martin Chalfie , Roger Y. Tsien "f*or the discovery and development of the green fluorescent protein, GFP*"

# Talk layout

- MeV ion beam lithography for biomedical applications
- MIMMA MeV ion microscope
- Commercalisation

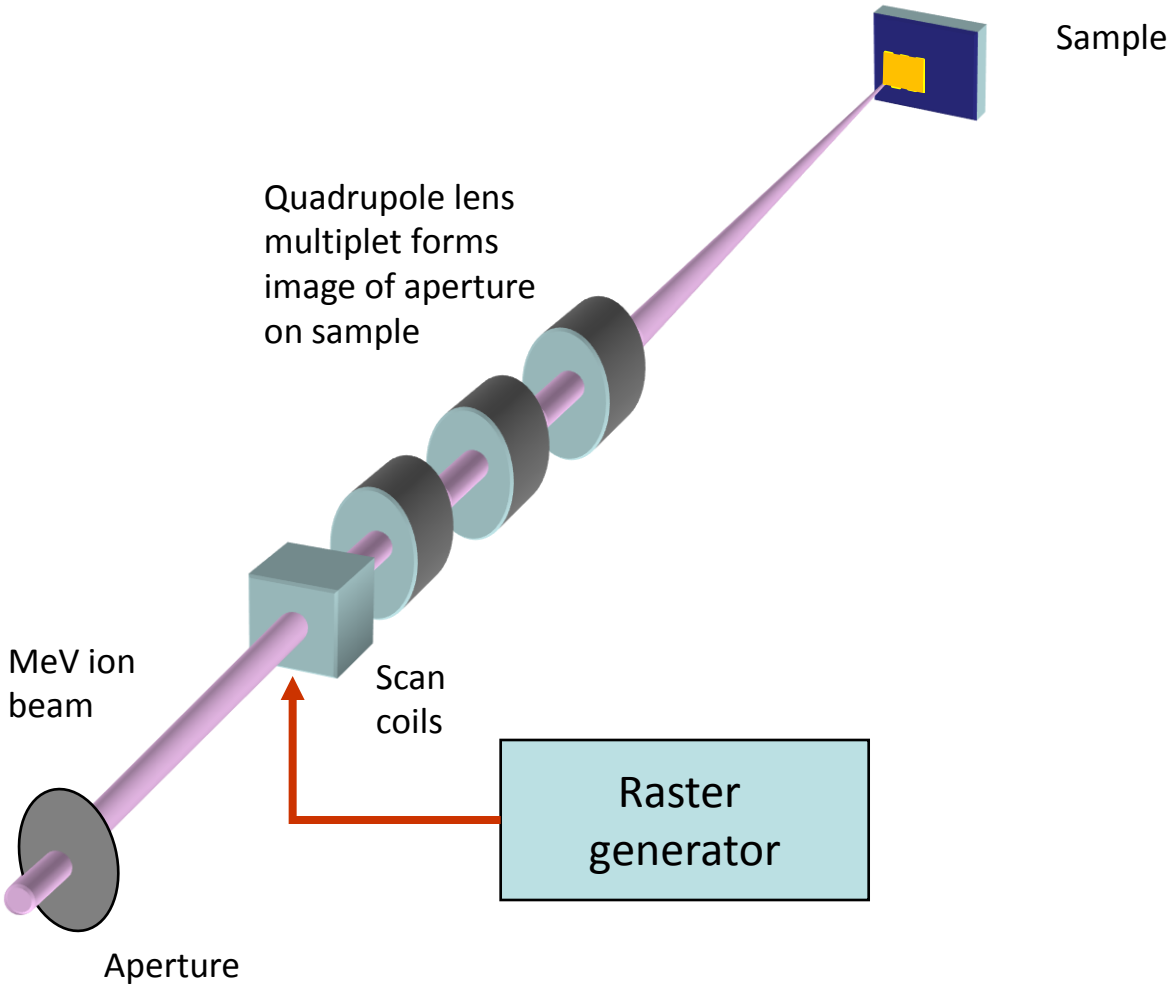


Faculty of Mathematics and Science,
University of Jyväskylä,  Finland

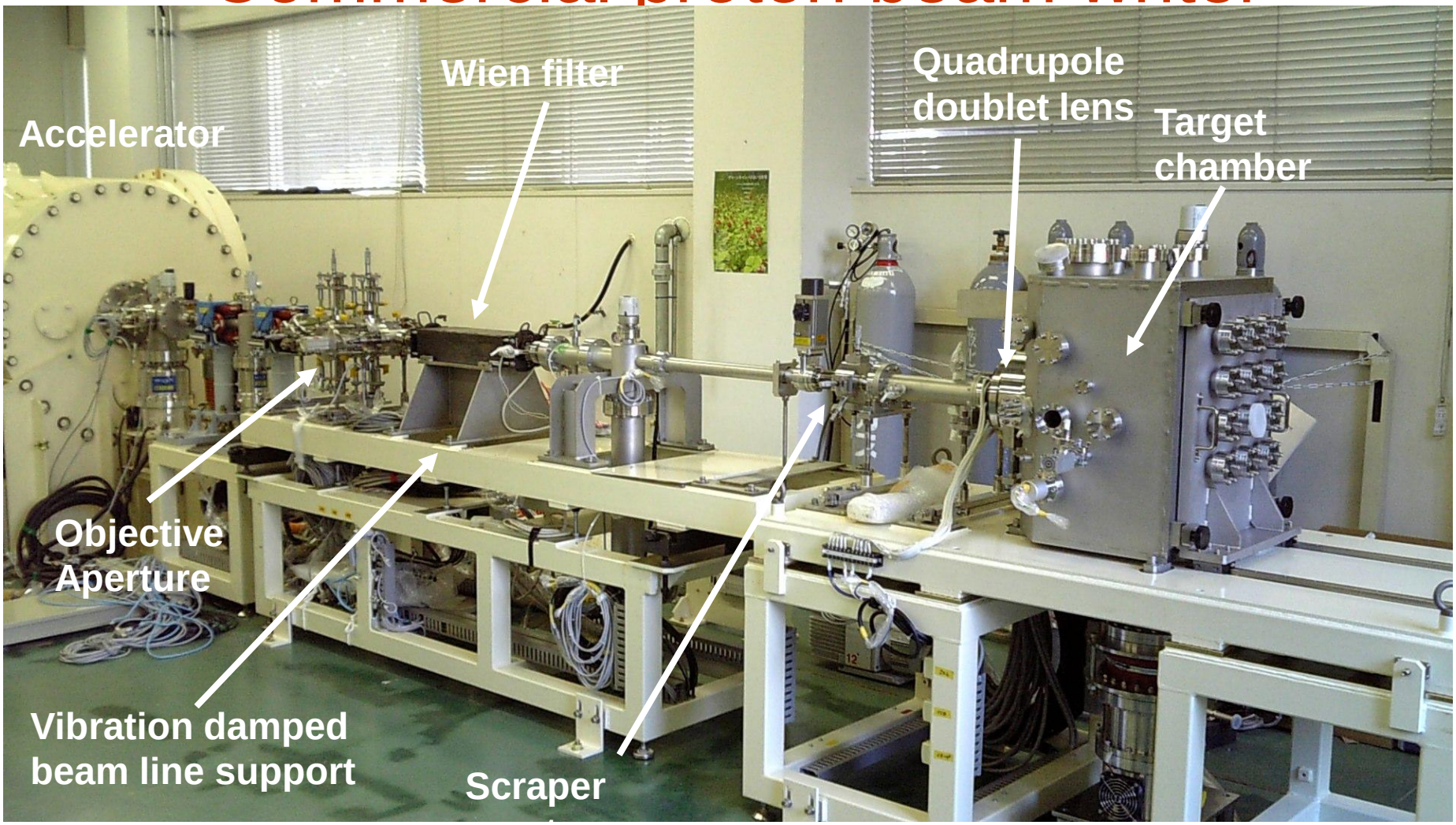


# MeV ION BEAM LITHOGRAPHY

# Proton Beam Writing (PBW) with focused beam

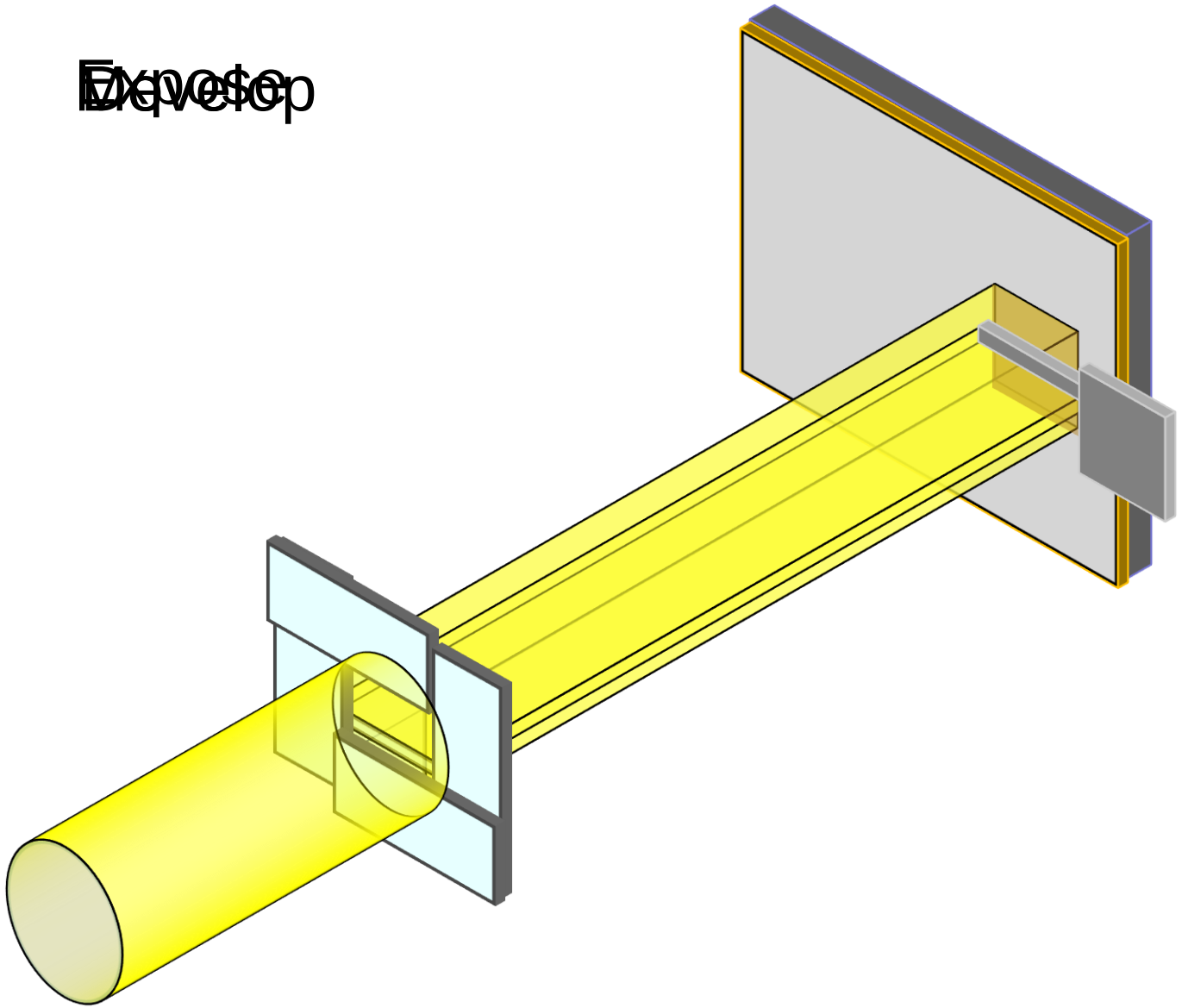


# Commercial proton beam writer

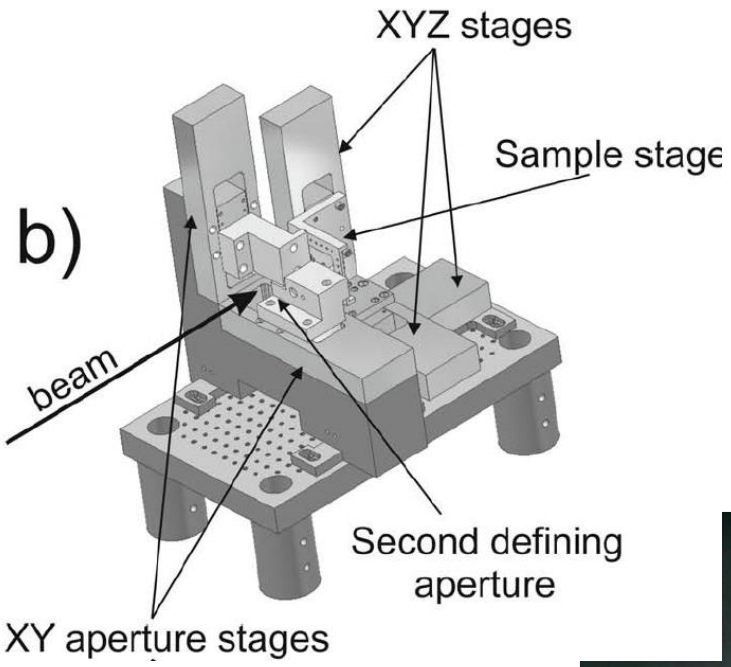


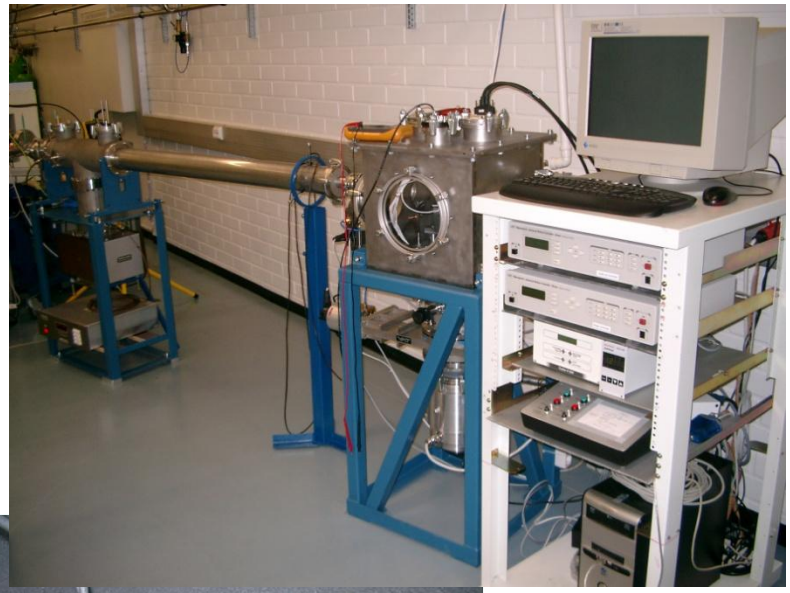

A commercial proton beam writing accelerator at the Center for Flexible Micromachining, Shibara Institute of Technology, Japan (S. Sangyuenyongpipat, H. J. Whitlow, S. T. Nakagawa and E. Yoshida, Proc CAARI 2008, American Institute of Physics, In press)

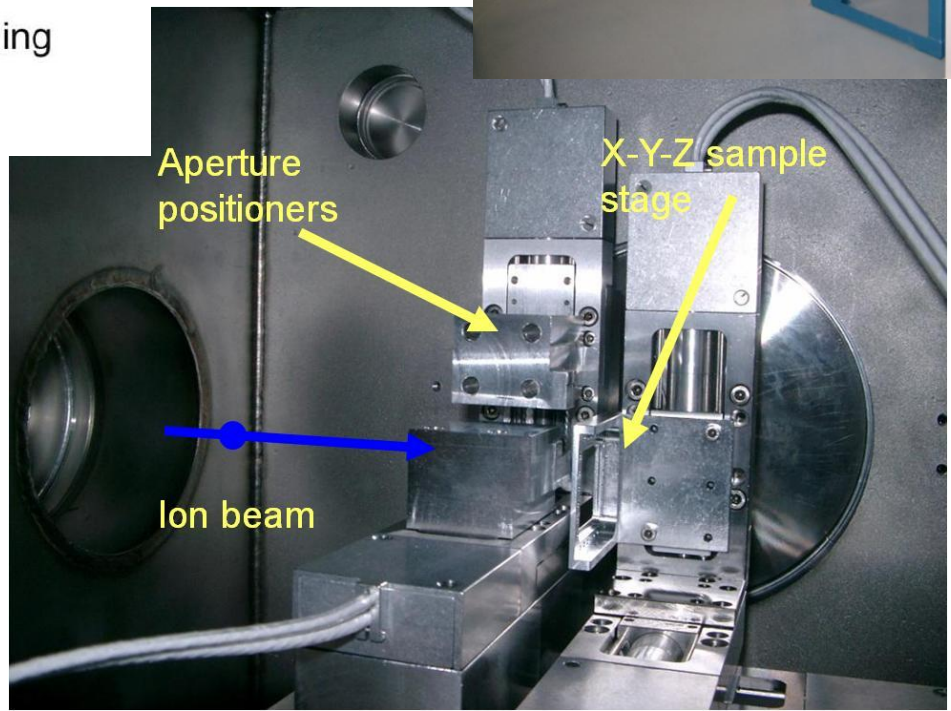
# Lithography with shaped beams



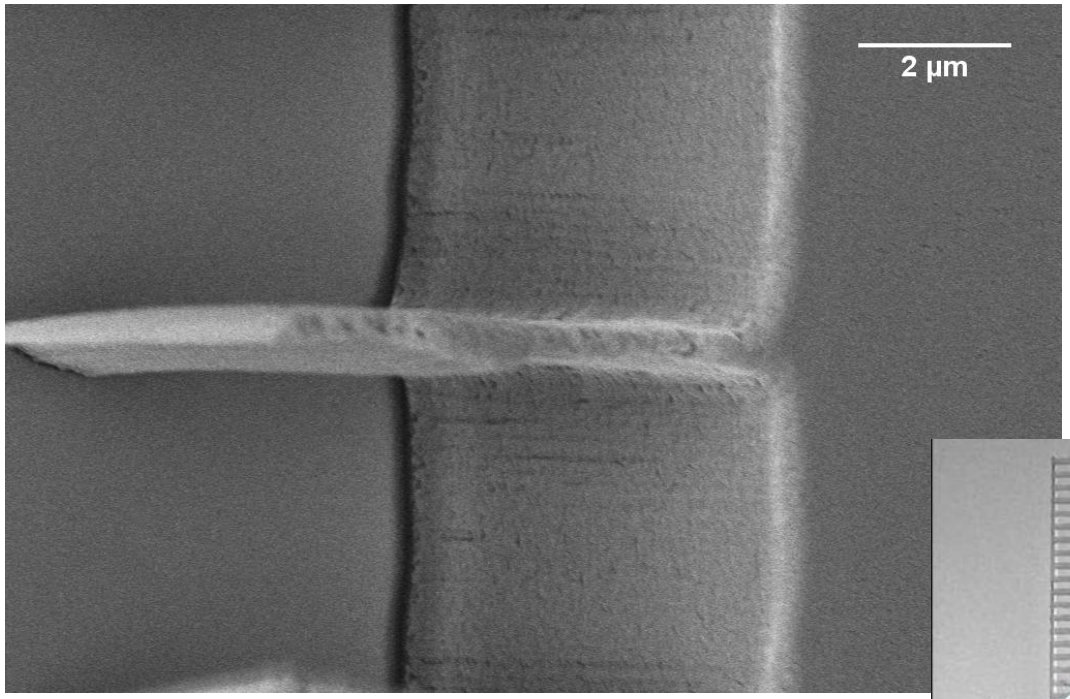
# Jyväskylä MeV ion beam lithography system





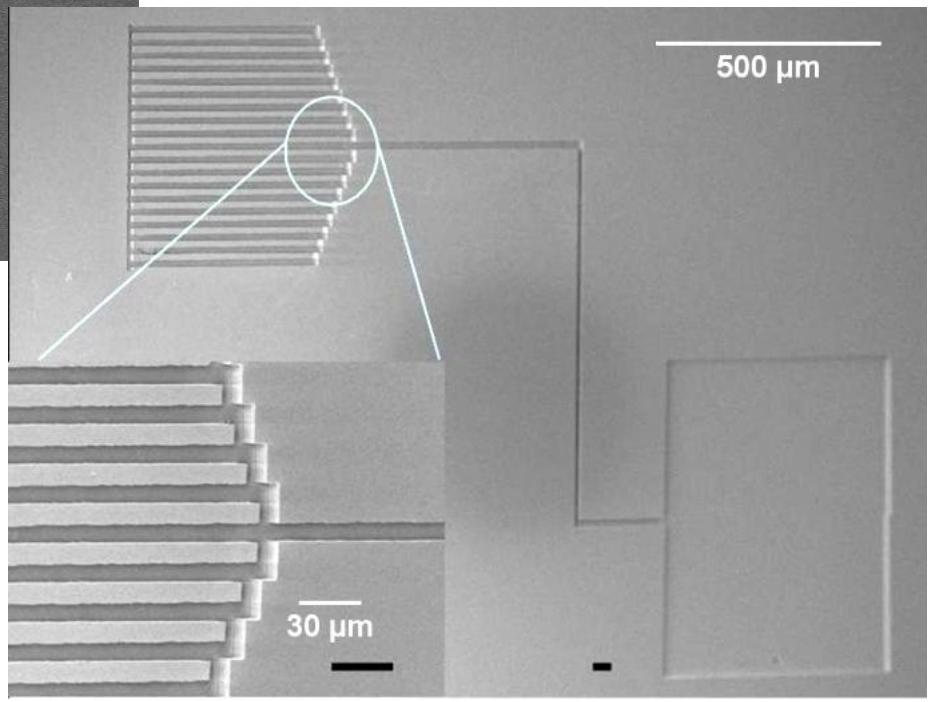
# MeV ion beam lithography in Jyväskylä




S. Gorelick, T. Sajavaara and H.J. Whitlow (unpublished result)

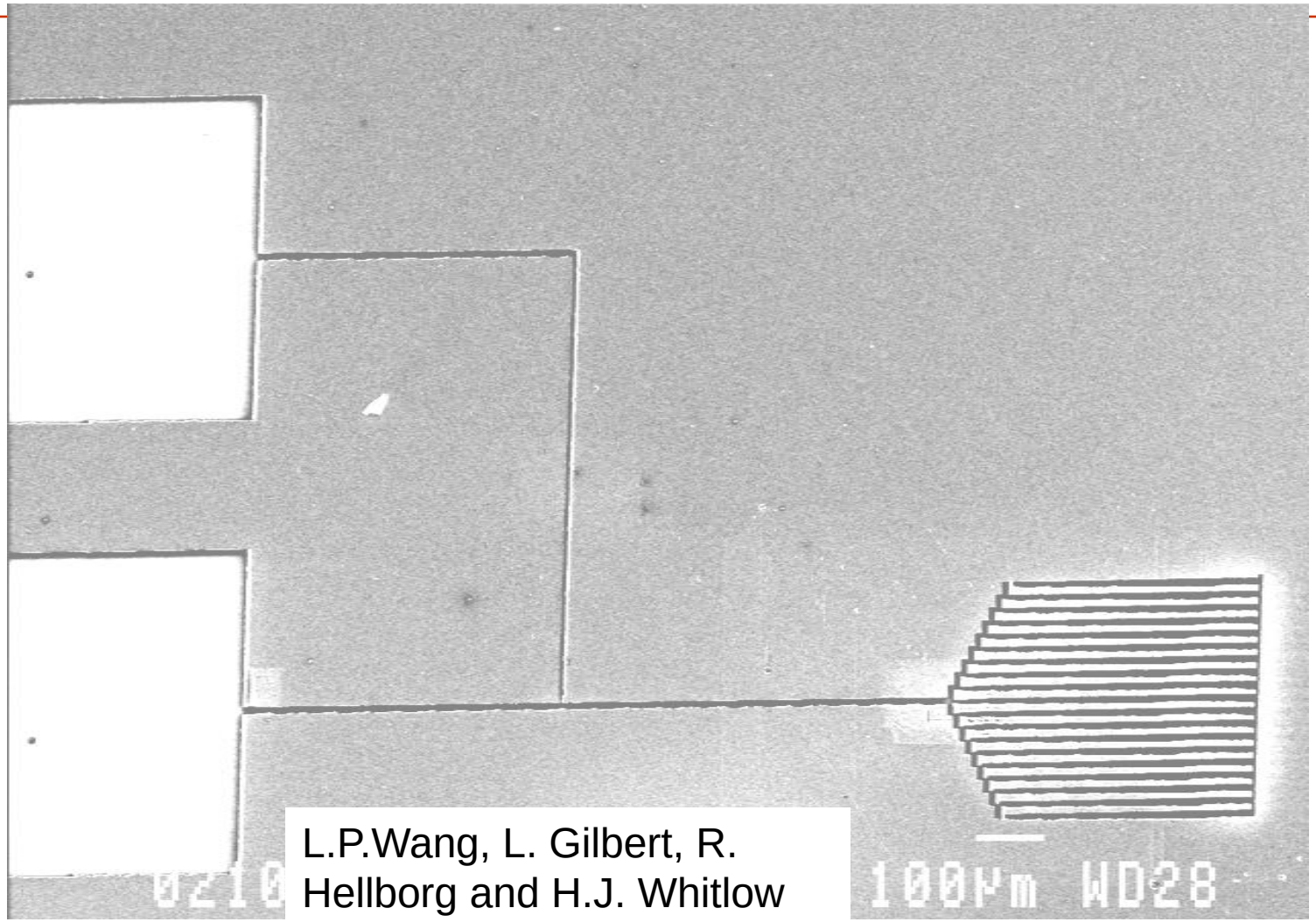
- The Jyväskylä system can produce extremely well defined ion beams. (few hundred nm here.)

- The ion beams have straight paths in 10 µm thick PMMA (about the thickness of a typical cell)




Prototype microfluidics structure comprising reservoir, capillary and micro pump. The PMMA structure is 12 µm thick and the channel width is 10 µm. (Liping Wang, Leona Gilbert and Harry J. Whitlow, University of Jyväskylä, unpublished results.)

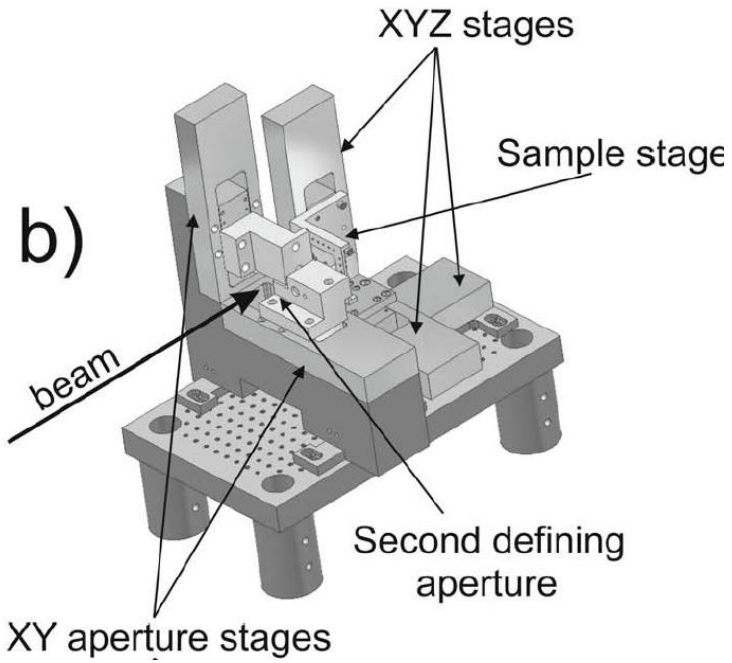
# Prototype µ-fluidics LOC device




L.P.Wang, L. Gilbert, R. Hellborg and H.J. Whitlow (unpublished results)

# MeV ion Beam Lithography Inovations

- Low cost – no expensive lenses needed (400 k€)
- Compatible with most existing accelerators for ion beam analysis
- High throughput
  - up to 70 000 times faster that focused proton/electron beam writing
  - Capable of large areas $mm^2$ / min
- Direct write
  - Easy to impliment patterns
- Vertial sidewalls
- 3D patterns possible
- **Applications**
  - Prototyping of Bio-MEMS microfluidic devices
  - Metamaterials
  - Optical waveguide circuits



# MIMMA - MeV ION MICROCOPY FOR MATERIALS ANALYSIS

# Why MIMMA?

The resolution of most light microcopes is limited by the Abbe Criteria

$$h = \frac{0.61\lambda}{n \sin\theta}$$

$$h =\sim 210\ \text{nm}$$

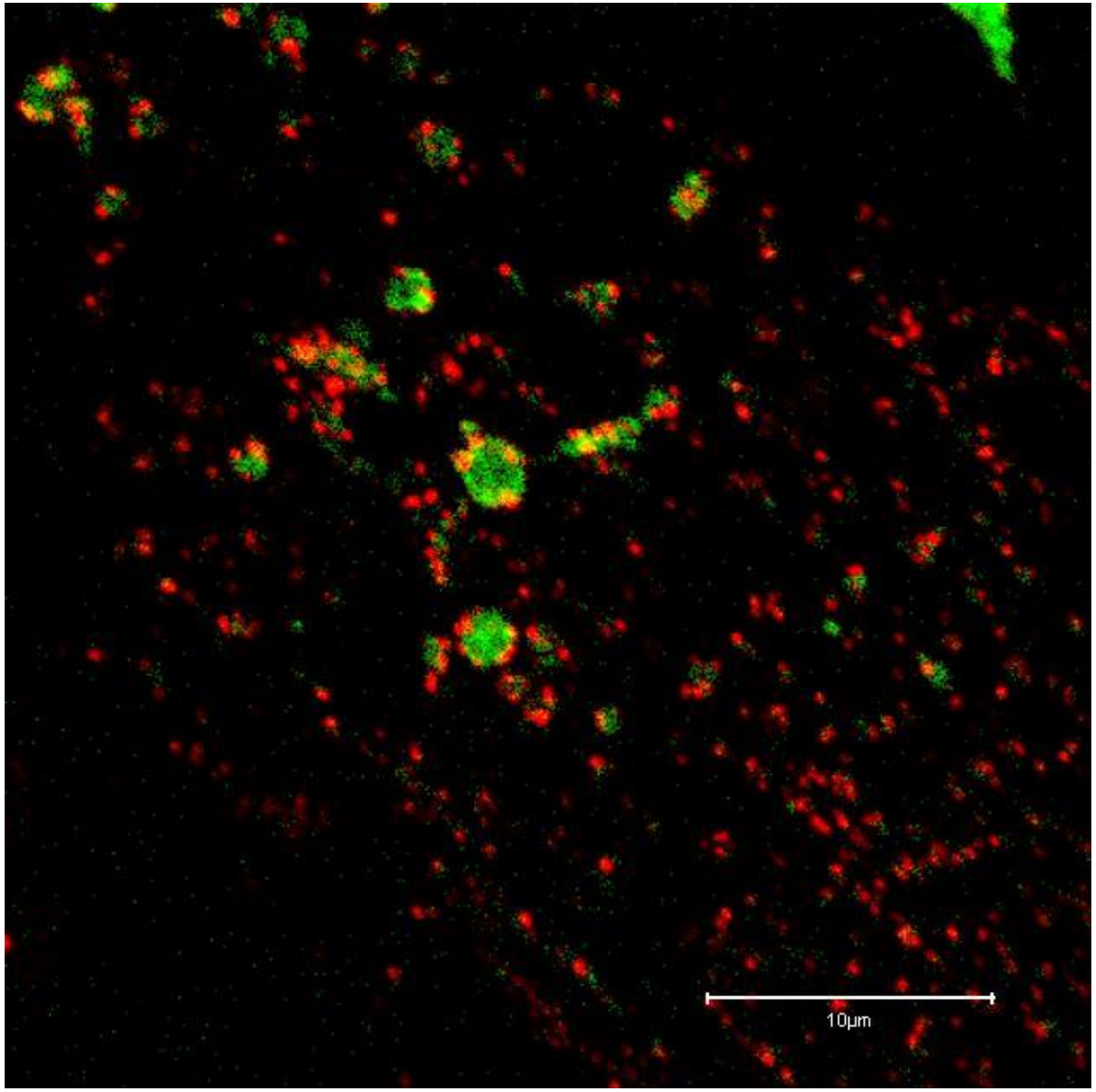

*Courtesy of the Clendening History of Medicine Library, University of Kansas Medical Center.*

In MIMMA we use MeV ions instead of UV light to breakthrough this barrier.

This will improve the available physiological information from cells and organelles.

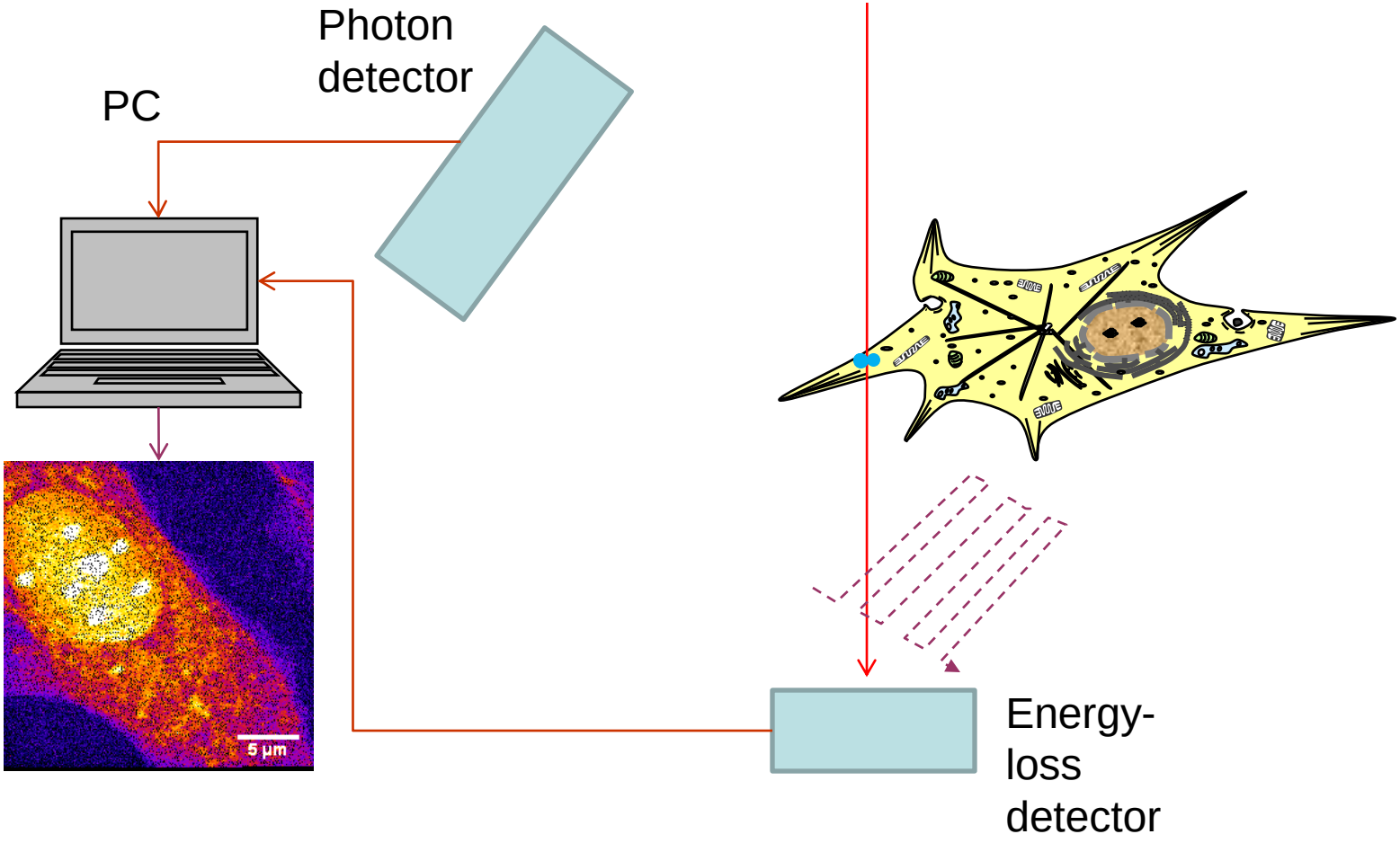
# Fluorescence confocal microscopy

Fluorescent groups called fluorophores can be attached to antibodies that combine with a specific biomolecule. The location of these proteins in the cell can then be observed from their fluorescence under uv light

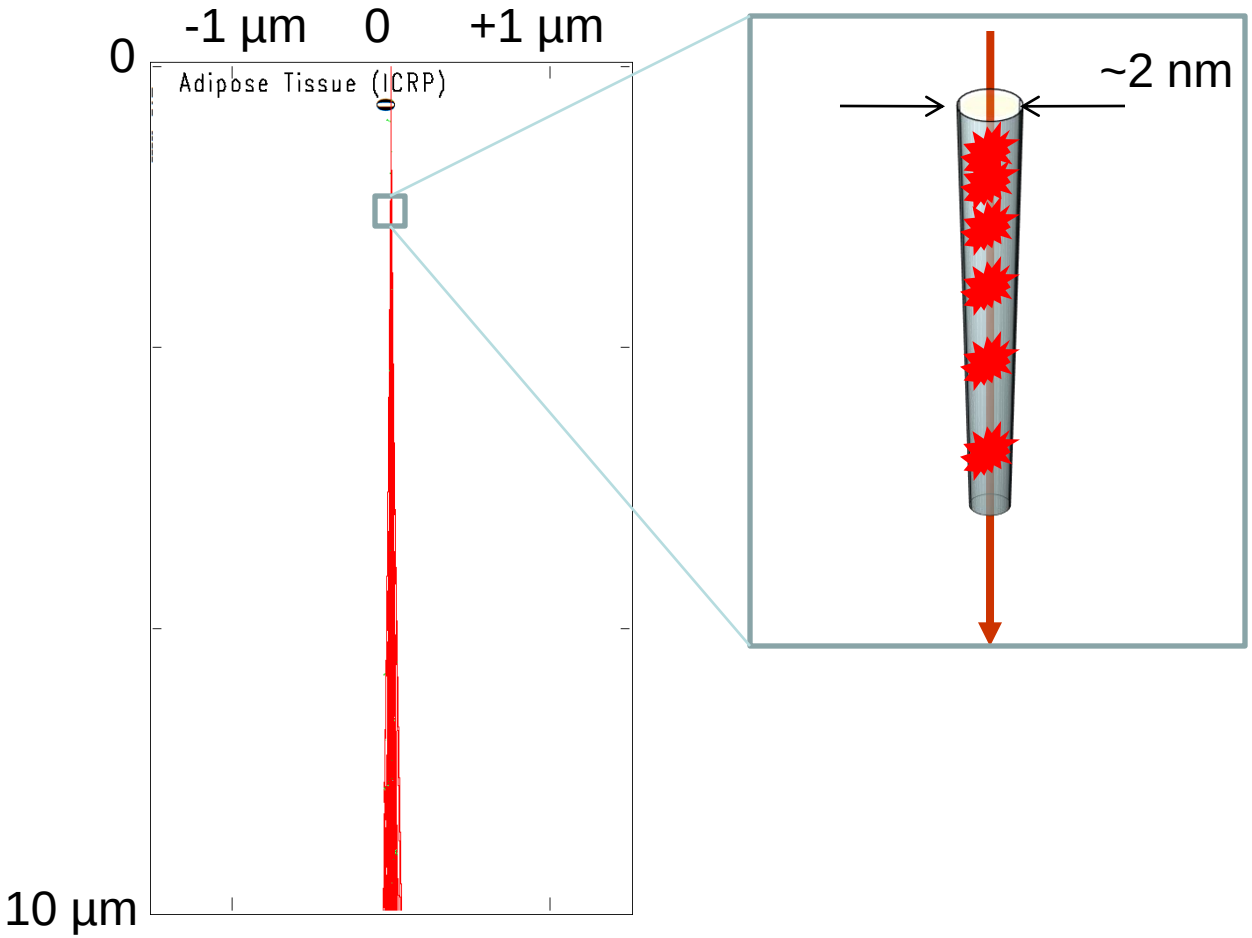



Confocal microscopy image of a human cell showing different location of two proteins in endosomes

# How MeV Ion Microscopy works



# A radically different physics concept




- Light is emitted from fluorophores only within ~2 nm of ion path
- Each ion can excite many fluorophores giving **high quantum efficiency**

# MIMMI configuration

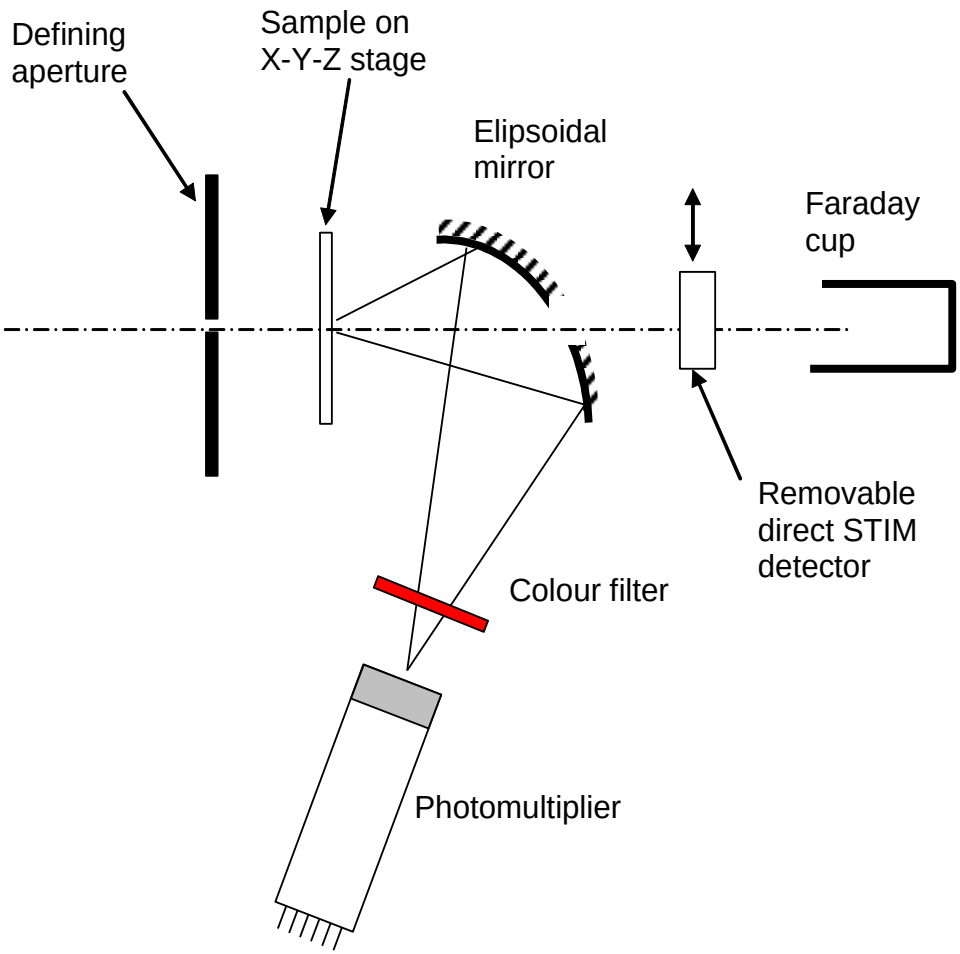
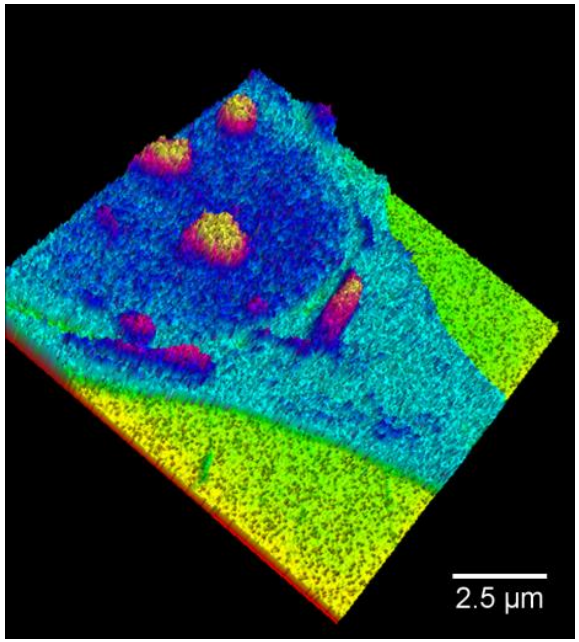



Fig. 4 Schematic illustration of set-up for MeV ion beam fluorescence imaging using the Jyväskylä PPAL system.

# Inovations of MIMMA

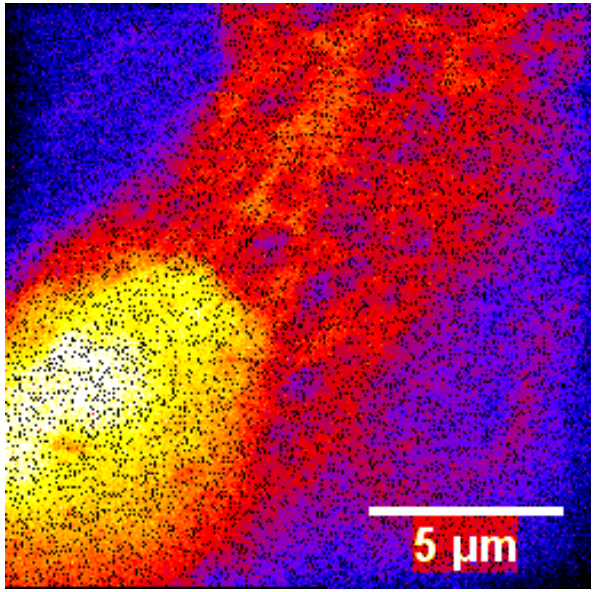
- Uses radically different physics to bust through the diffraction-limit
- Is able to image specific biomolecules by immunohistological staining. Conventional MeV ion microbeams only see different elements.
- Able to work with whole cells
- Uses conventional optical fluorescence confocal microscopy flurophores and dyes that is not posible with competing methods
- Extension to 3D nanotormography is straightforward
- Low-cost: Expensive ion or light lenses are not required.
- Advanced optical image processing gives high speed and resolution data collection.
- Use modern  photonics readout developed to reduce it to make microscope a simple Add-on Attachment for the MeV ion beam lithgraphy system




Structural image of human cell taken using 1 MeV $He^{+}$

# Ingedients and status for MIIMA

- MeV Ion beam with defintion of few hundred nm
  - (Tested: record is <50 nm)
- Photon detection system
  - (Concept tested in PNNL)
- Transmitted ion detection system
  - (Tested in Singapore)
- Image collection software
  - (LabView based)
- Image rendering tool
  - BioImage XD (Tested)




Structural image of human breast cancer cell taken using 1 MeV $He^+$

# Concept for commercal exploitation

- Design and prototype developed at JY (Current phase, proof of principle)

- Partner 1: Mechanical workshop company with CNC machine competence do pre-manufacture stage with JY.
- Partner 2: Software company with imaging know-how develop fast software data collection system and 3D nano-tormography with JY
- Partner 3: International accelerator system or microscope /biomedical instrument company markets the product manufactured in Finland (co-branding).

TEKES application

# Patent outlook

- Basic MeV ion Beam Lithography – already done
- Basic Mev ion induced fluorescence microscopy – probably not
- Improved concepts for fluorescence microscopy – yes
- Software/hardware to achieve sub-100 nm nanometre resolution / linewidth - yes
- Software for high speed image collection – yes
- Concept for 3D nanotormography - yes

# Market over 10 years

- Lithography system
  - Circa 400 ion beam analysis labs. If 10% buy – 40 units
  - Complete systems with accelerator 20 units

- Microscopy system
  - 100 labs worldwide 20% share 20 units
  - Complete systems with accelerator 10 units

# The End

(Sunset over Jyväskylä from an office window)

# Competing techniques

- A range of optical technqiues have been developed for sub-light wavelength cellular imaging.
  - Stimulated Emission Depletion Spectroscopy (STED), Standing wave fluorescence microscopy, 4Pi, multiphoton 4Pi, STORM, Scanning Near-Field Microscopy (SNOM), X-ray nanobeams and tomography

- **All have major drawbacks** and very few labs have facilities.
- **Optical**
  - High laser power density, need for thin samples (200 nm), expensive optics, extremely expensive fs laser facilties or limited number of dyes.
- **SNOM**
  - Near-field only allows within 100 nm of cell surface to be probed
- **X-ray naonobeams**
  - Needs synchrotron, low access, only weak interaction

**Based on**: J.H. Rice, Molecular Biosystems 3(2007)781

S.W. Hell, Nature Biotechnology 21(2003)1347

Bates et al . Science 317(2007) 1749