STROBE
BUILDING THE MICROSCOPES OF TOMORROW

UCLA College | Physical Sciences
Physics & Astronomy

Electron Microscopy
Introduction, Applications and Opportunities

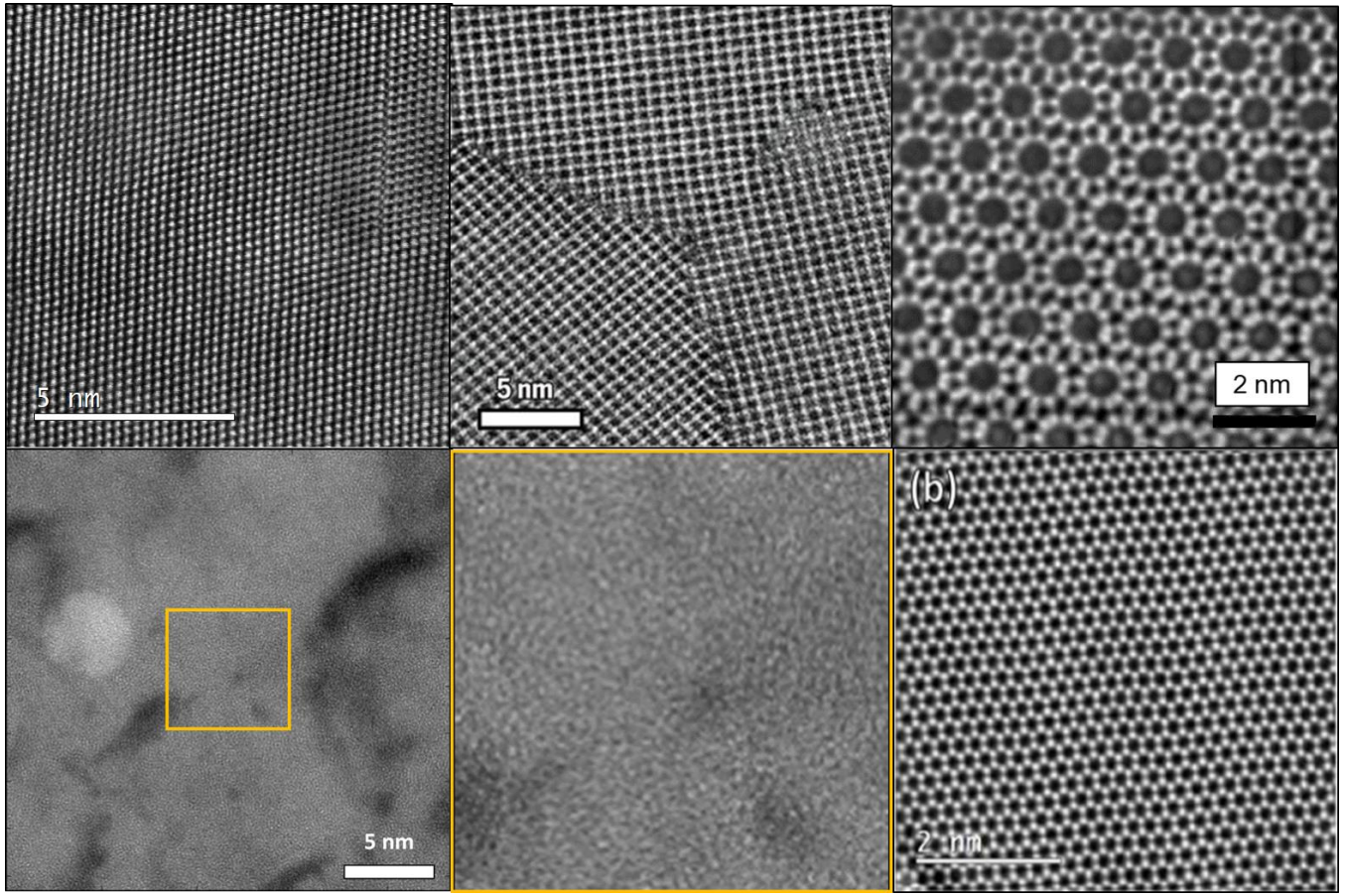
Colum O'Leary

STROBE Postdoctoral Fellow
Department of Physics & Astronomy
University of California, Los Angeles

BERKELEY LAB

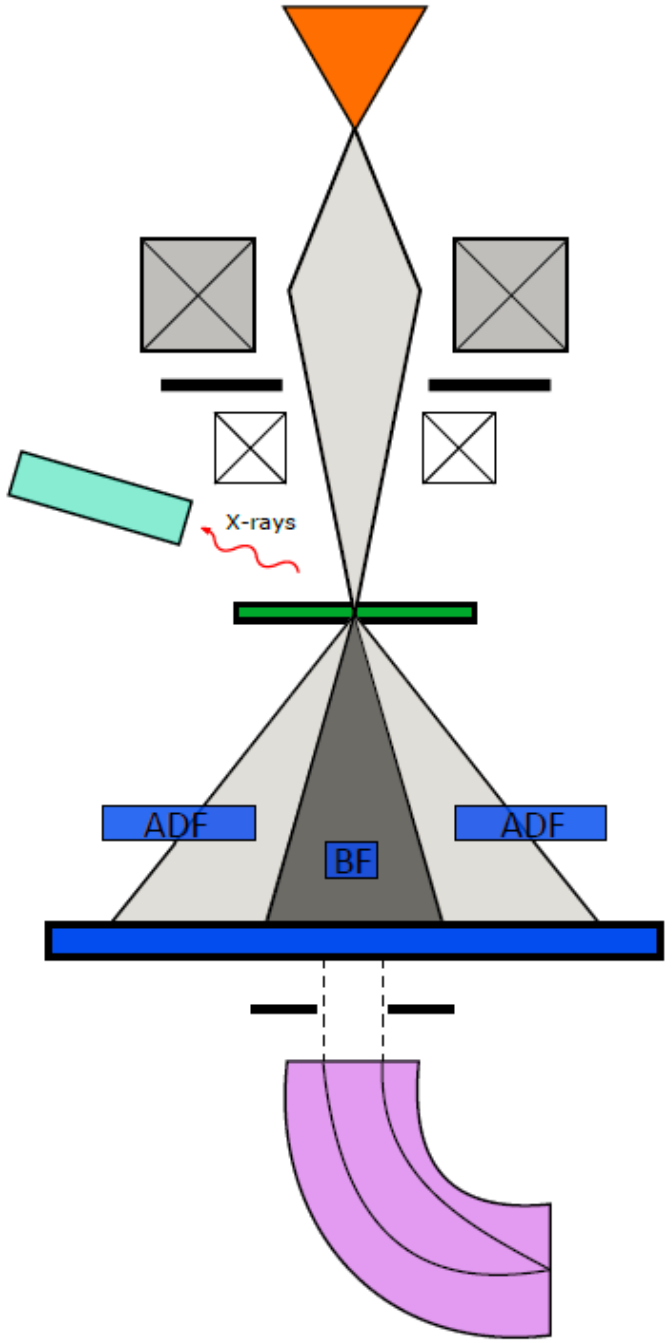
Outline

- **Why use electron microscopy?**
- Modes and applications.
- Challenges and latest developments.



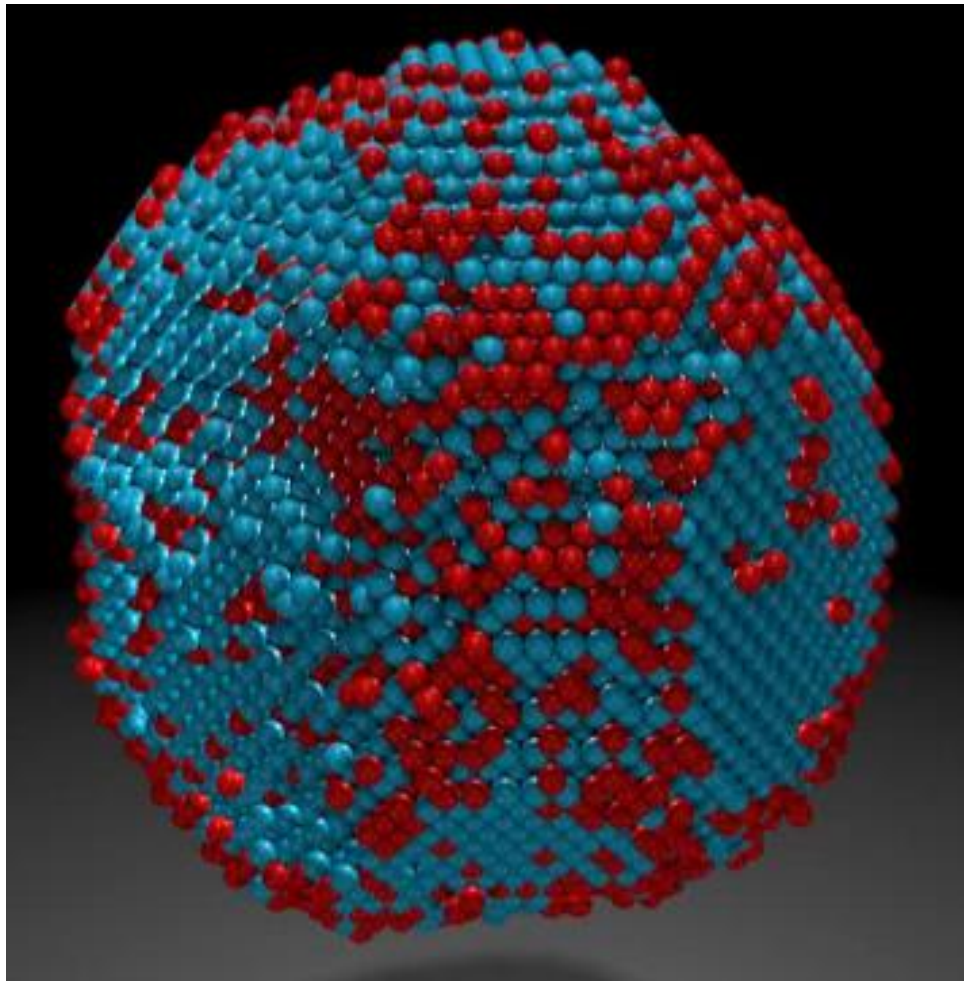
Outline

- Why use electron microscopy?
- **Modes and applications.**
- Challenges and latest developments.



Outline

- Why use electron microscopy?
- Modes and applications.
- **Challenges and latest developments.**



Why electron microscopy?

Electron microscopy

- Incident radiation is electrons.
- At high velocities, electrons possess very short wavelengths.
- Lenses are electromagnets.
- nm/atomic-resolution imaging and diffraction studies.
- A lab in an instrument.

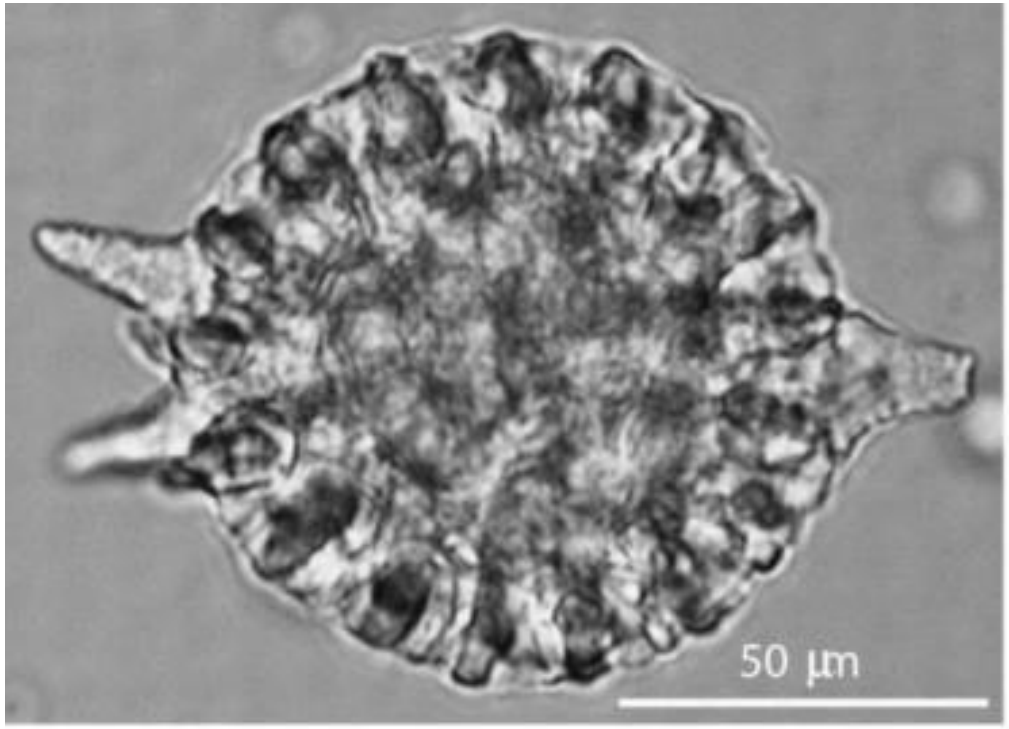
Electron microscopy

Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments



(a) Radiolarian under light microscope

From: General Chemistry: Principles, Patterns, and Applications, B. Averill & P. Elderege

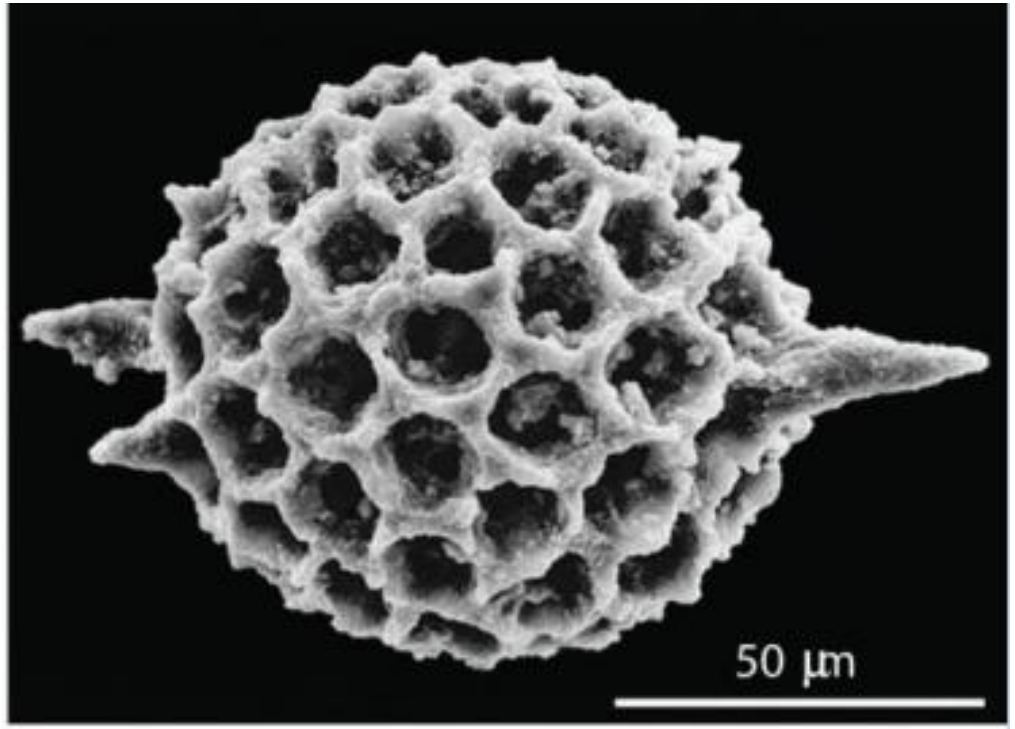
Electron microscopy

Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments



(b) Radiolarian under electron microscope

General Chemistry: Principles, Patterns, and Applications, B. Averill & P. Elderege

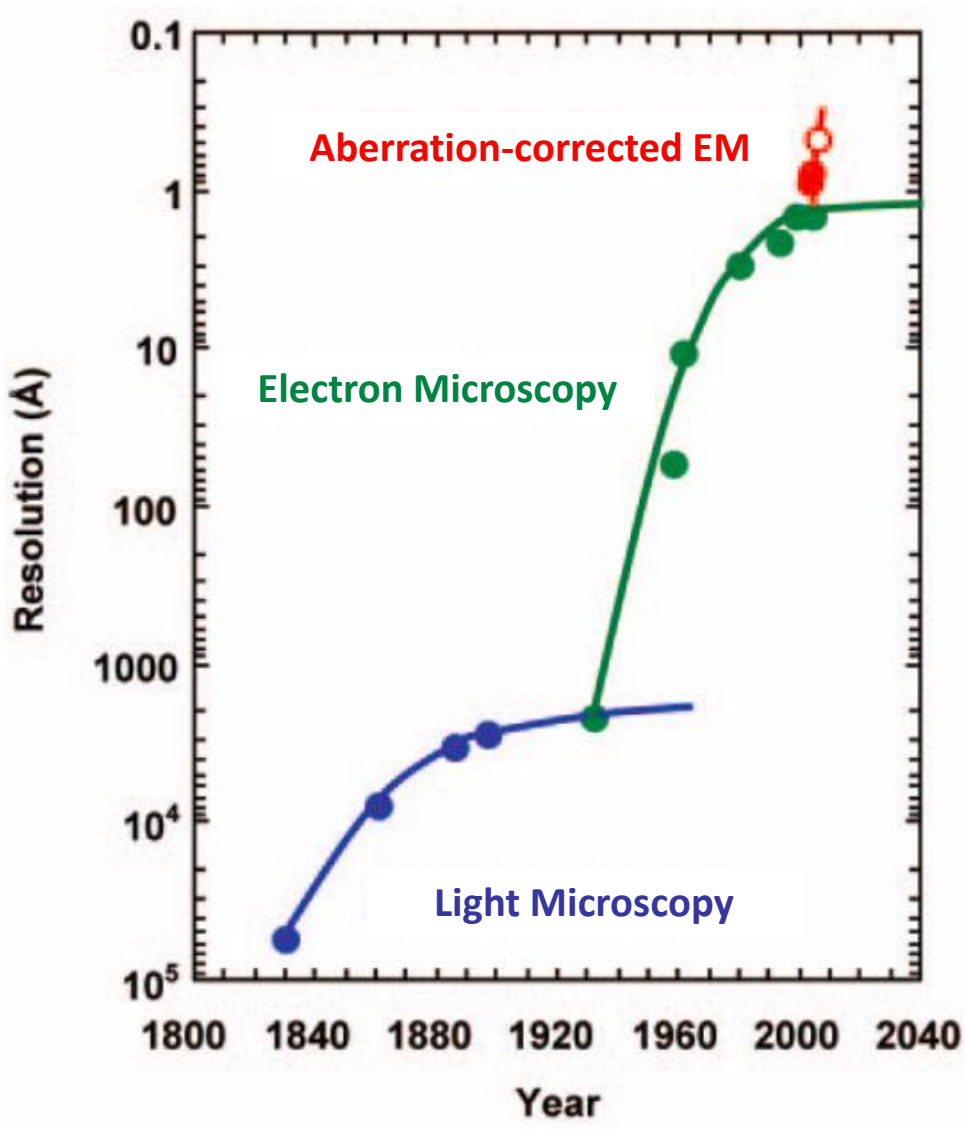
Electron microscopy

Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments



*Pennycook et al, MRS Bulletin **31**(01), 2006*

Electron microscopy

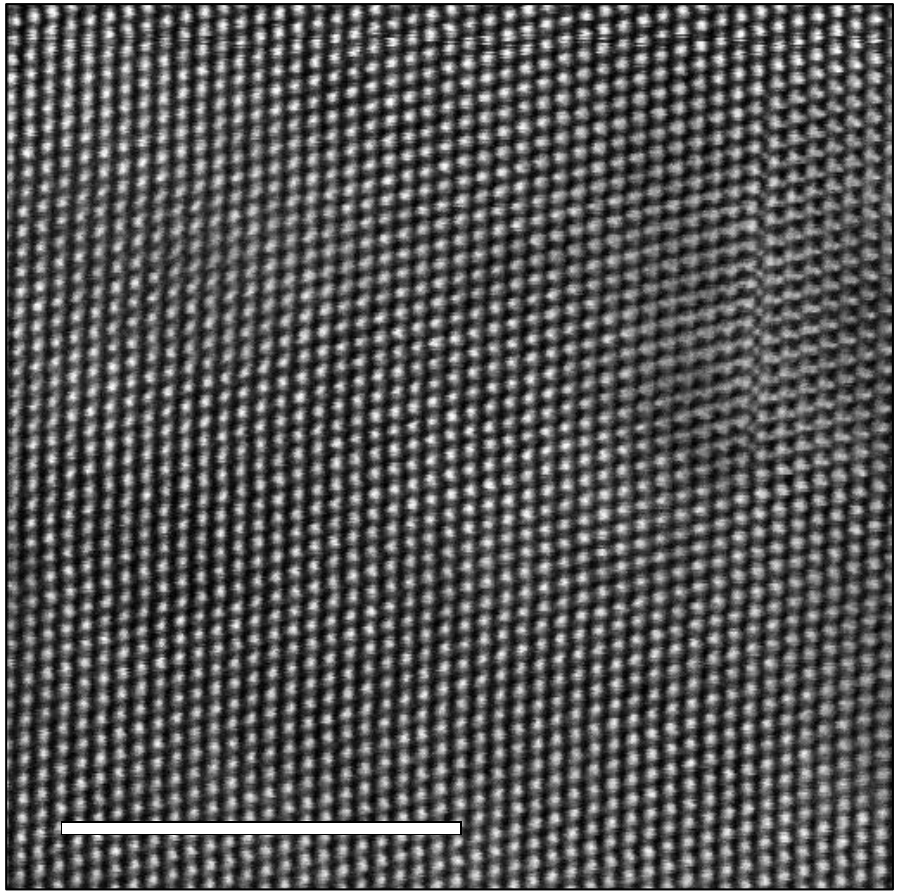
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Pt crystal



Crystal structure and imperfections

Electron microscopy

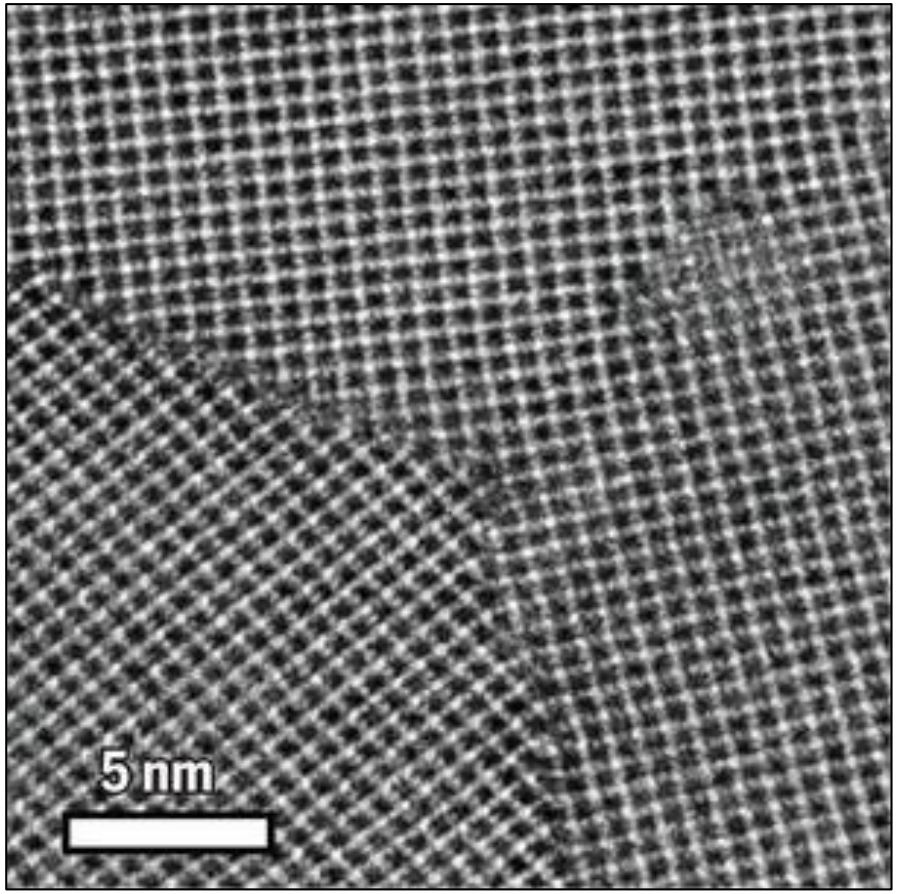
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Lead-halide perovskite



Crystal structure and imperfections

*Rothmann et al, Science **370**(6516), p.* eabb5940, 2020.

Electron microscopy

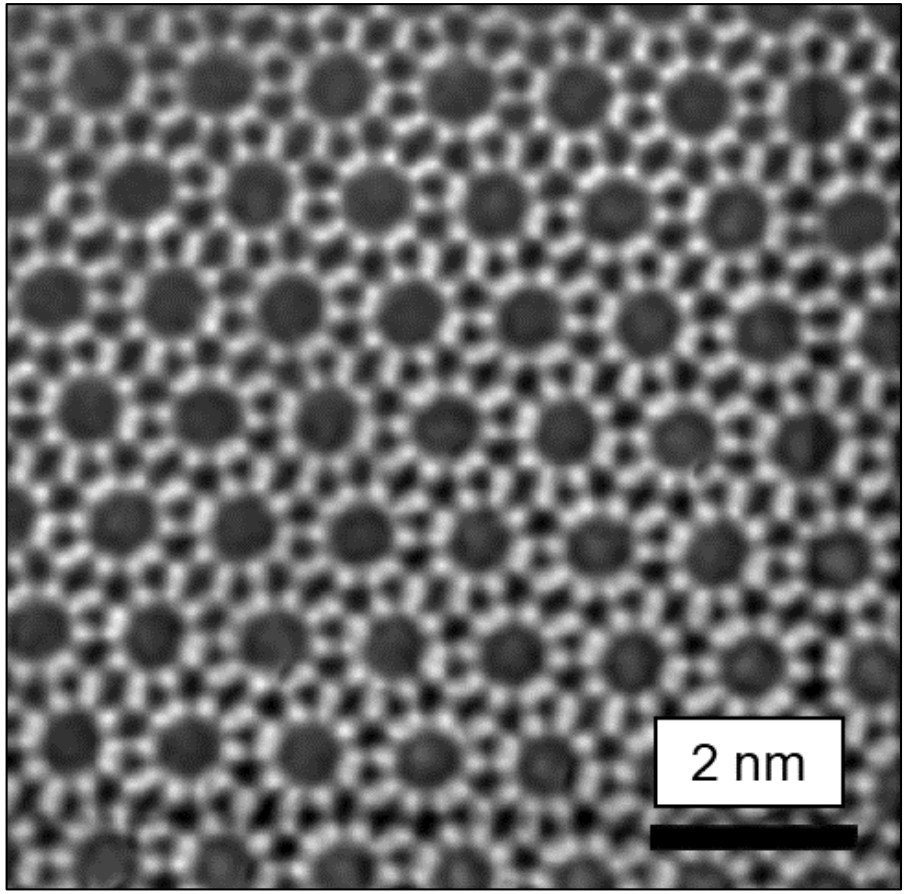
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Zeolites



Porous nanostructures

O'Leary et al, Appl. Phys. Lett. ***116****, p. 124101, 2020*

Electron microscopy

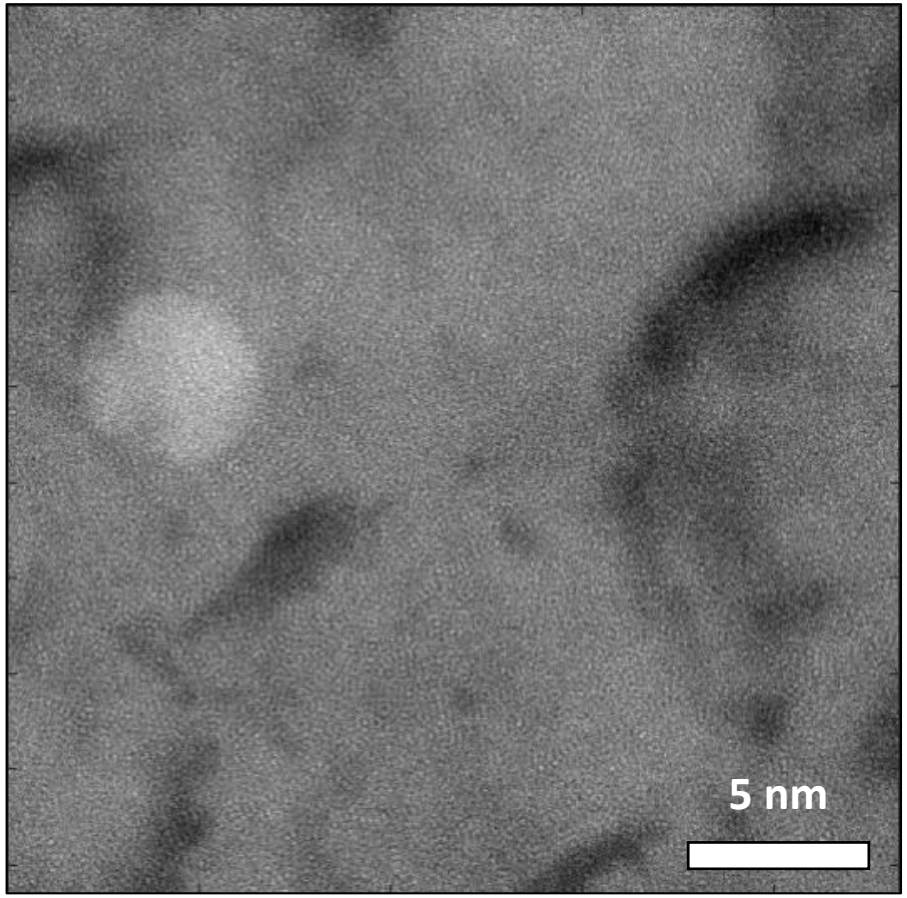
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Ta film w/ Au nanoparticle



Amorphous materials

Electron microscopy

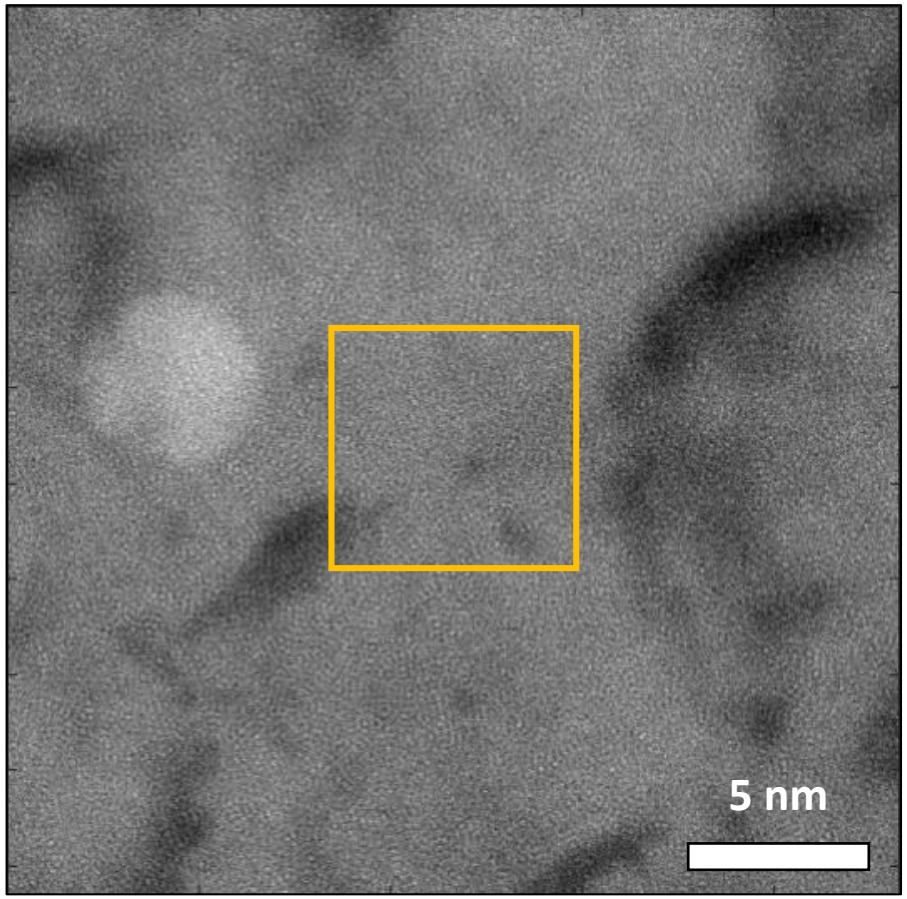
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Ta film w/ Au nanoparticle



Amorphous materials

Electron microscopy

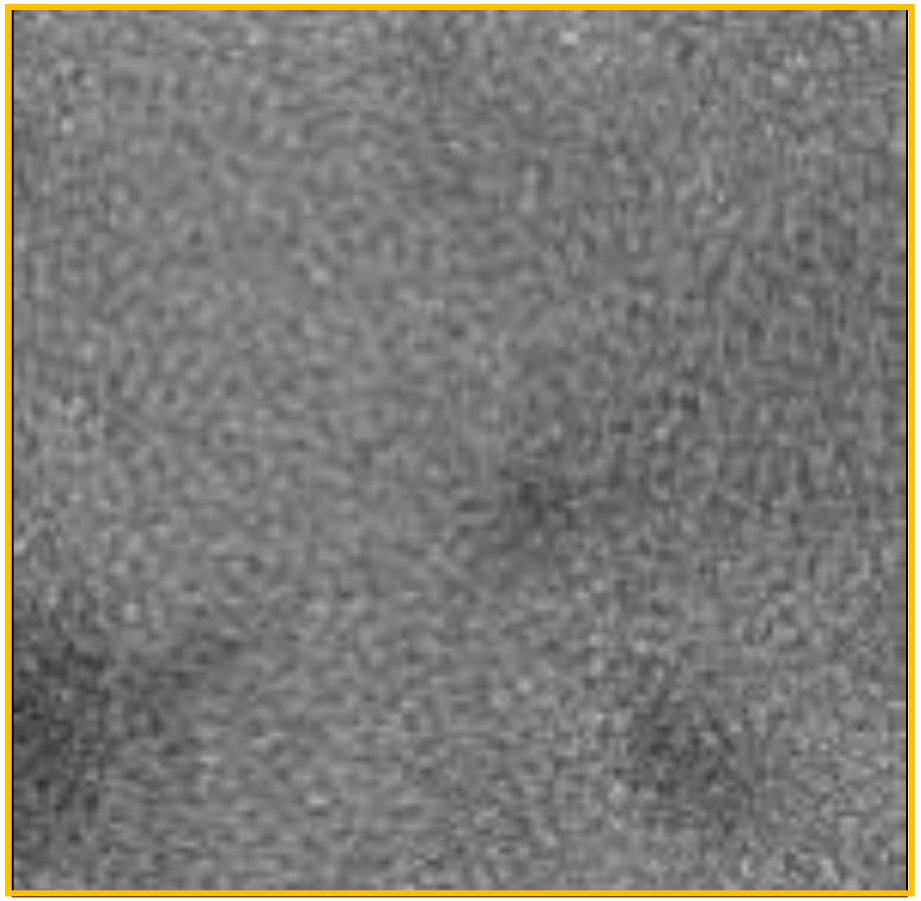
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Ta film w/ Au nanoparticle



Amorphous materials

Electron microscopy

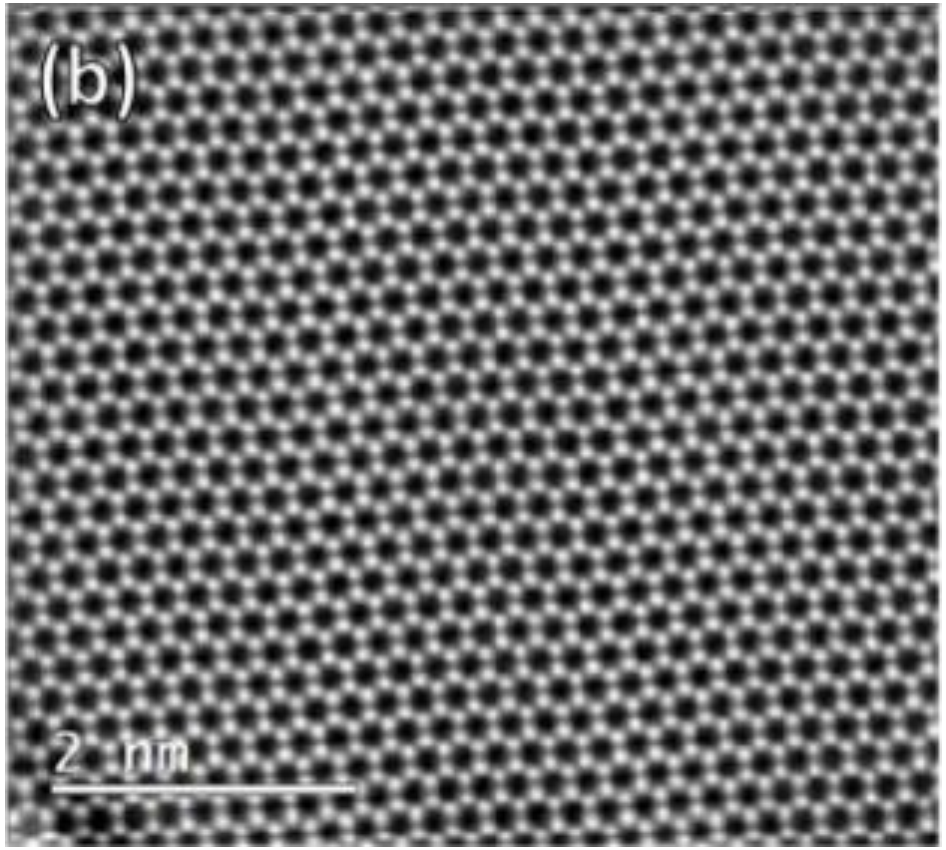
Visible light

- Wavelength: 400 - 700 nm
- Resolution: approx. 200 nm

Electrons

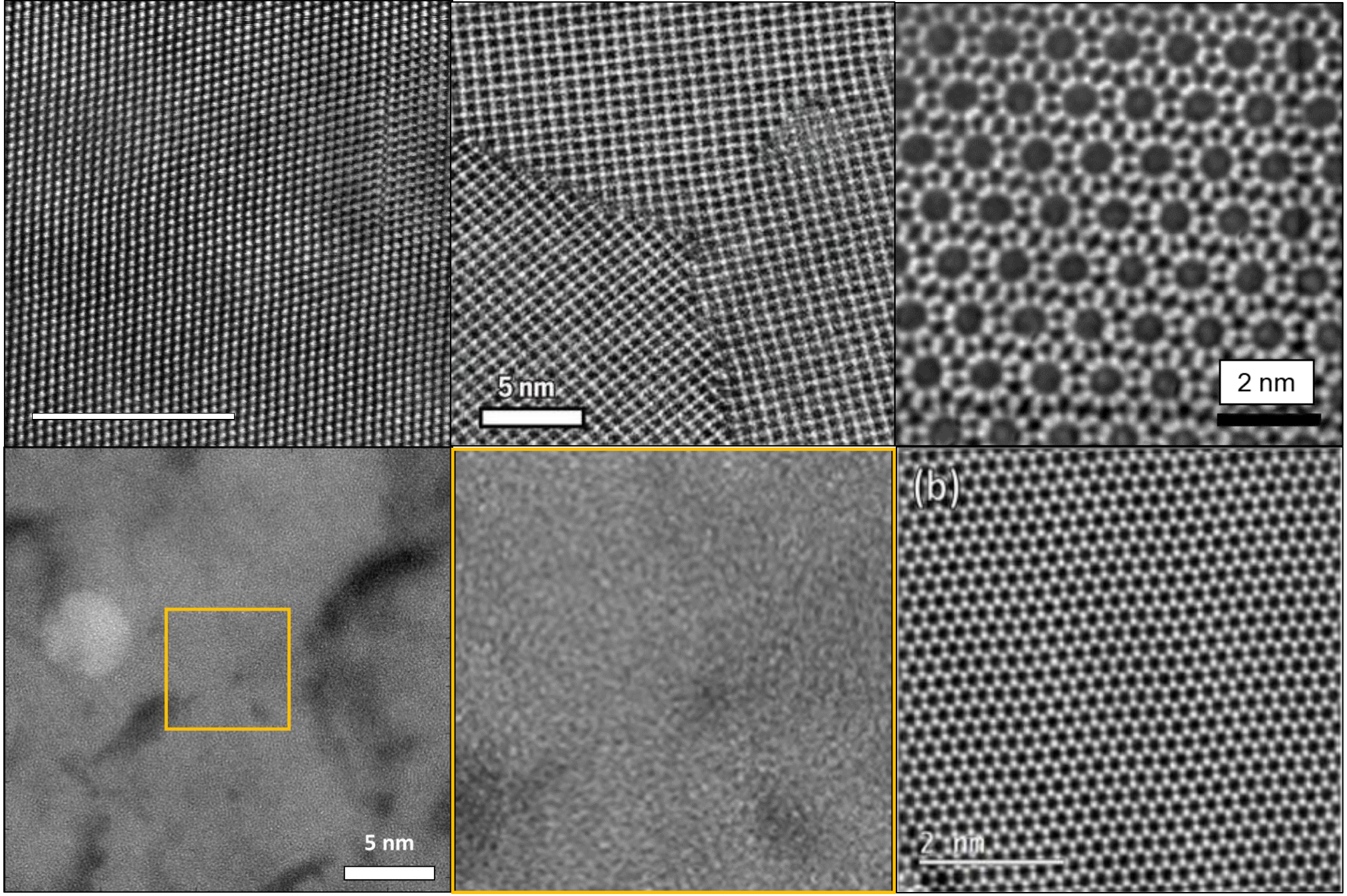
- Wavelength @ 300kV: 0.00197 nm
- Resolution: < 1 Å for modern instruments

Graphene



Nanomaterials

O'Leary et al, Microscopy and Microanalysis (in press), 2021


5 nm
2 nm
5 nm
(b)
2 nm

Practicalities

Light microscope

- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

Practicalities

Light microscope

- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

Practicalities

Light microscope

- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

Practicalities

Light microscope

- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

Practicalities

Light microscope

- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

Practicalities

Light microscope

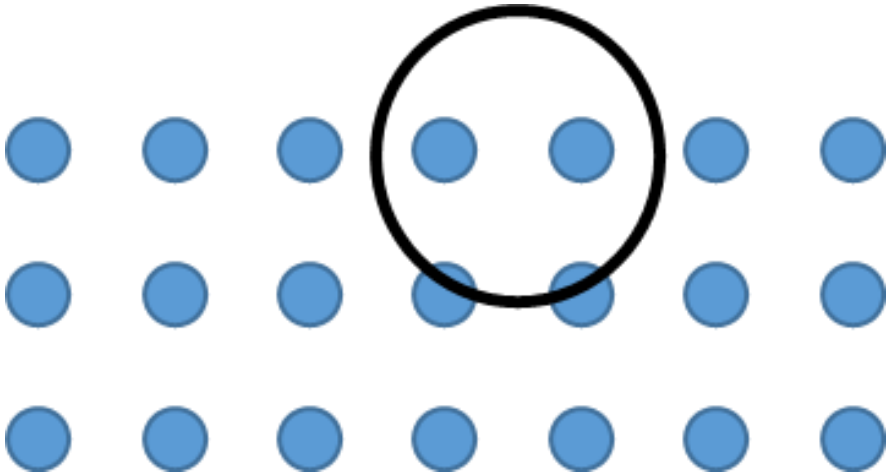
- Uses visible light
- Minimum sample prep
- Inexpensive equipment
- Live or dead samples
- No screen/camera needed
- Resolution ~ 10^{-7}m

Electron microscope

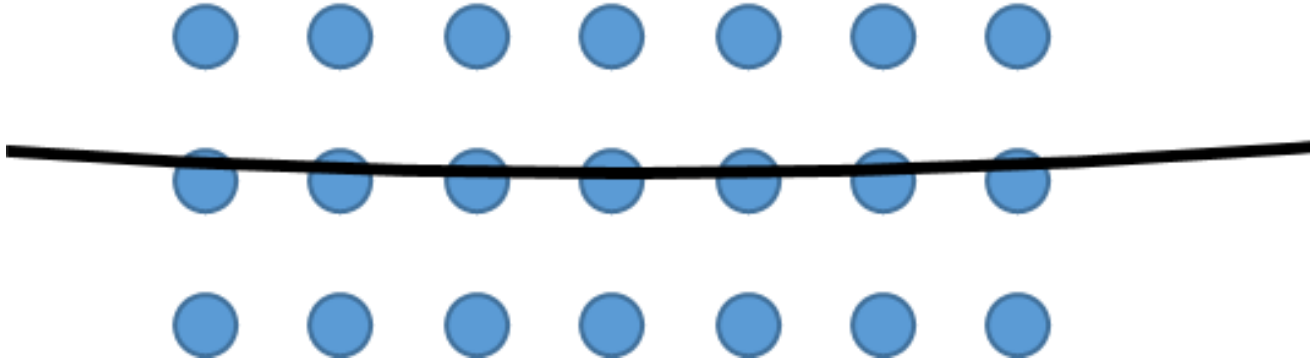
- Uses electrons
- Thin, clean sample
- Expensive, high maintenance
- Dead samples
- Screen/camera is required
- Resolution < 10^{-10} m

X-rays or electrons: Ewald Sphere

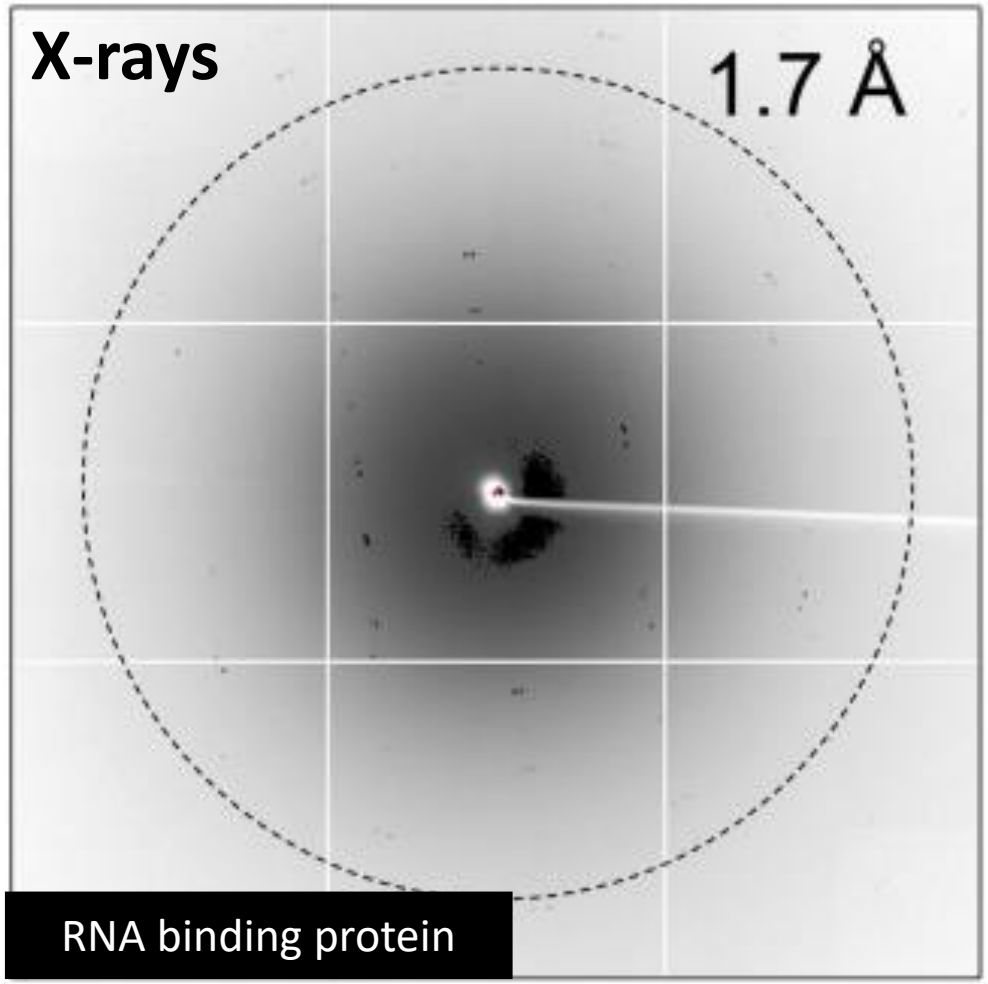
$Cu_K\alpha$ X-rays (λ = 1.54 Å)

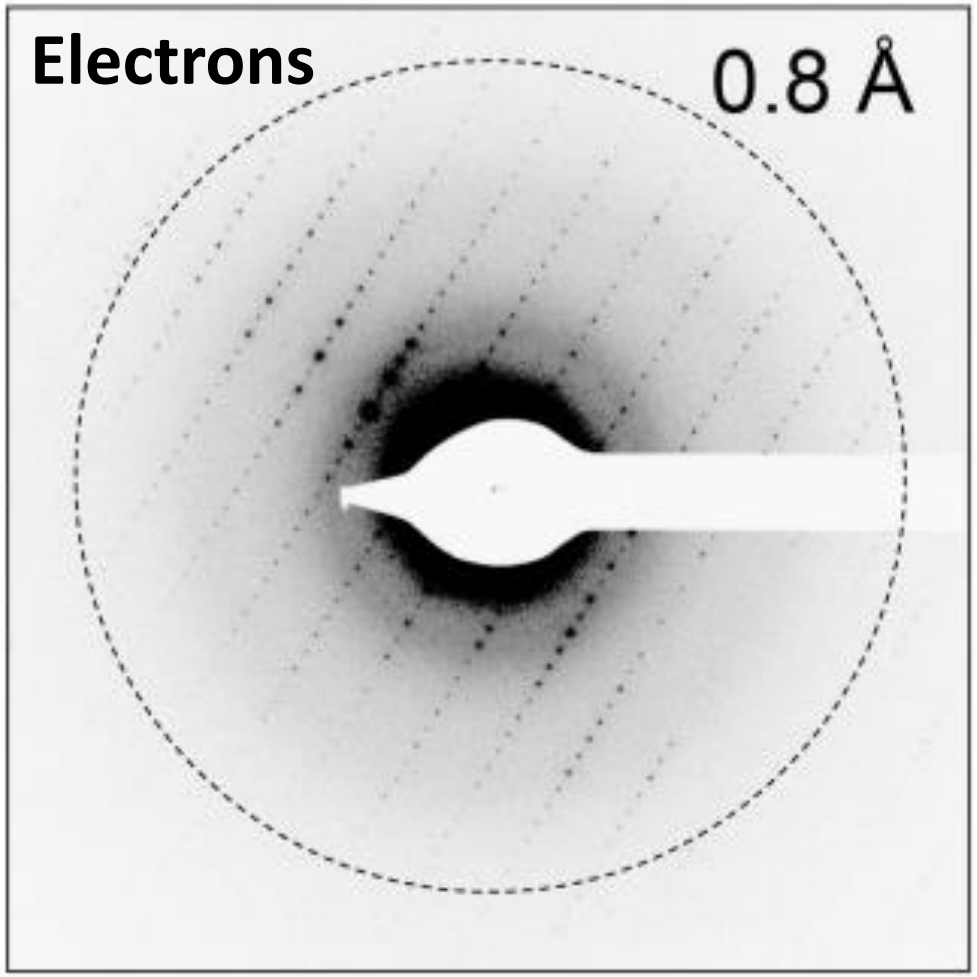


200keV electrons (λ = 0.02 Å)



X-rays or electrons: Diffraction patterns





*Luo et al, Nature Structural & Molecular Biology **25**(4), 2018*

X-rays or electrons?

X-rays

- Minimum sample prep
- Do not interact strongly with light elements
- Large field of view
- Average structure atomic structure
- Real-space resolution ~ 10^{-9} m

Electrons

- Sample must be thin, clean
- Interact strongly with light elements
- Small field of view
- Probe local structural changes
- Resolution < 10^{-10} m

X-rays or electrons?

X-rays

- Minimum sample prep
- Do not interact strongly with light elements
- Large field of view
- Average structure atomic structure
- Real-space resolution ~ 10^{-9} m

Electrons

- Sample must be thin, clean
- Interact strongly with light elements
- Small field of view
- Probe local structural changes
- Resolution < 10^{-10} m

X-rays or electrons?

X-rays

- Minimum sample prep
- Do not interact strongly with light elements
- Large field of view
- Average structure atomic structure
- Real-space resolution ~ 10^{-9} m

Electrons

- Sample must be thin, clean
- Interact strongly with light elements
- Small field of view
- Probe local structural changes
- Resolution < 10^{-10} m

X-rays or electrons?

X-rays

- Minimum sample prep
- Do not interact strongly with light elements
- Large field of view
- Average structure atomic structure
- Real-space resolution ~ 10^{-9} m

Electrons

- Sample must be thin, clean
- Interact strongly with light elements
- Small field of view
- Probe local structural changes
- Resolution < 10^{-10} m

X-rays or electrons?

X-rays

- Minimum sample prep
- Do not interact strongly with light elements
- Large field of view
- Average structure atomic structure
- Real-space resolution ~ 10^{-9} m

Electrons

- Sample must be thin, clean
- Interact strongly with light elements
- Small field of view
- Probe local structural changes
- Resolution < 10^{-10} m

Key components of electron microscopes

- Electron source with high coherence
- High vacuum inside microscope column
- Electron lenses
- Anti-contamination device
- Highly sensitive stage (piezoelectric)
- Scanning coils

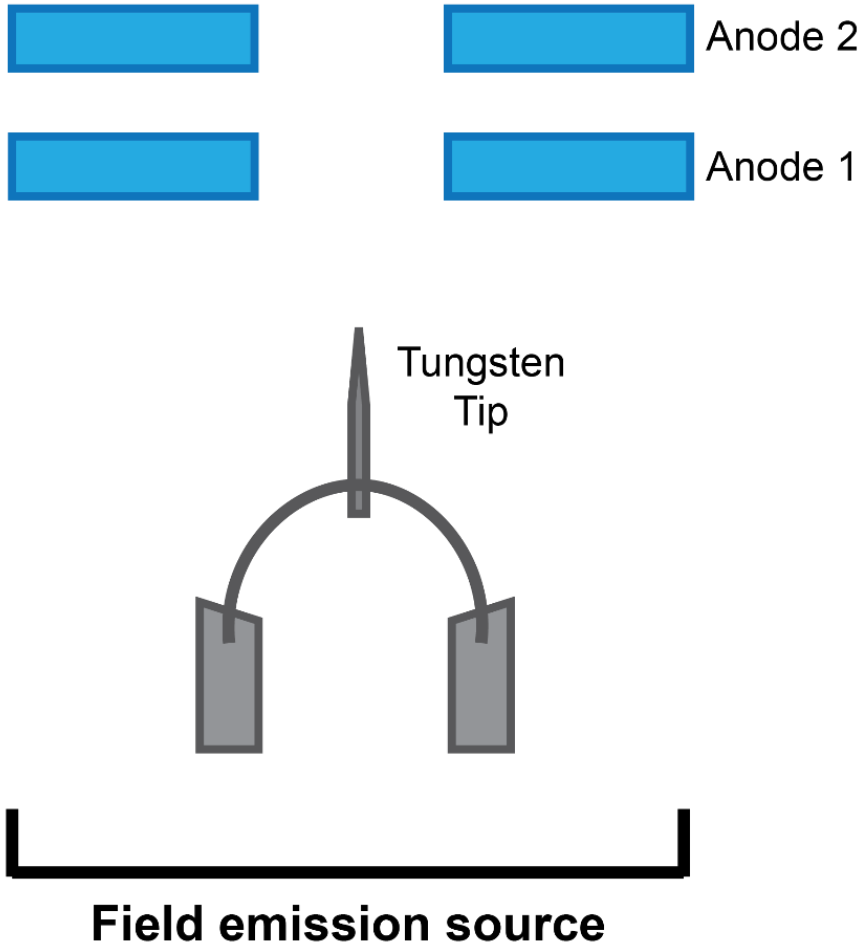
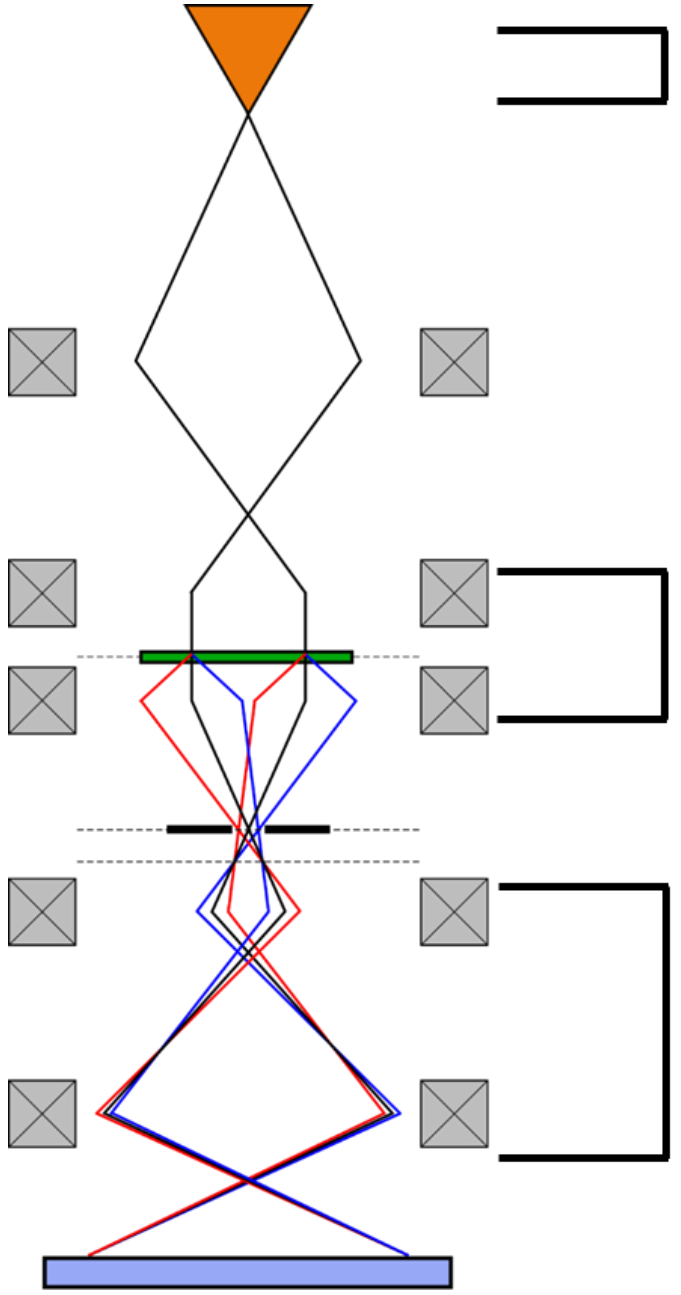


Figure from ThermoFisher Scientific

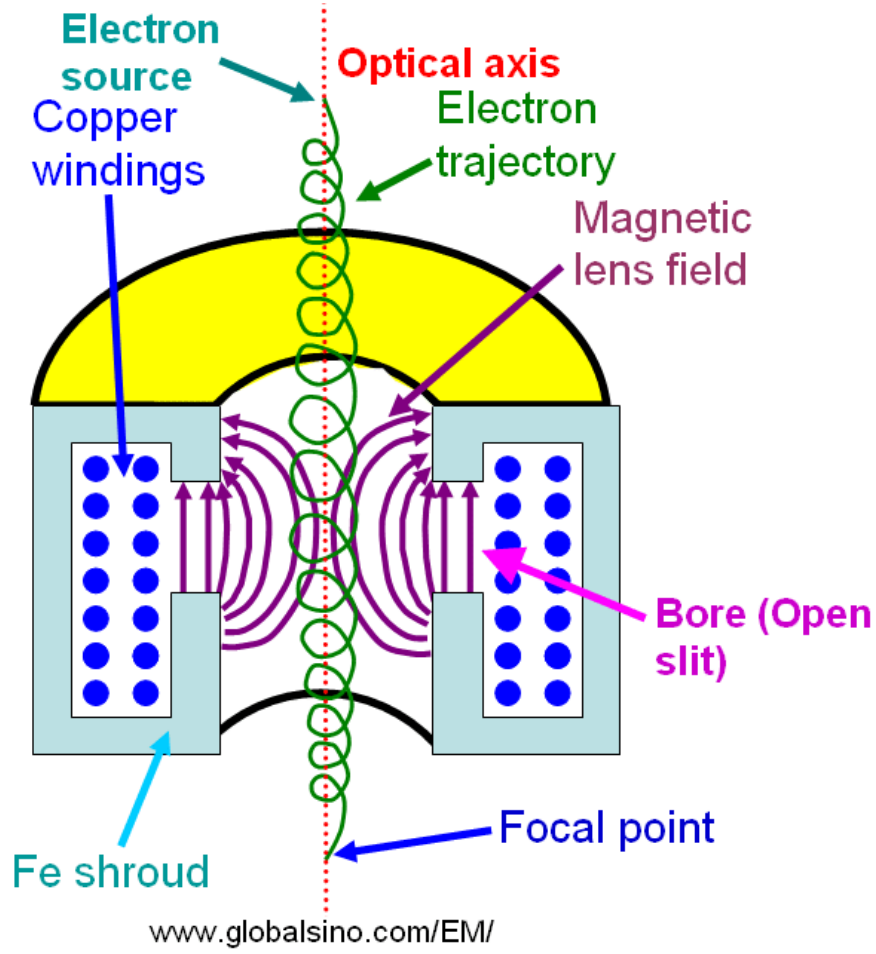
Key components of electron microscopes

- Electron source with high coherence
- High vacuum inside column
- Electron lenses
- Anti-contamination device
- Highly sensitive stage (piezoelectric)
- Scanning coils



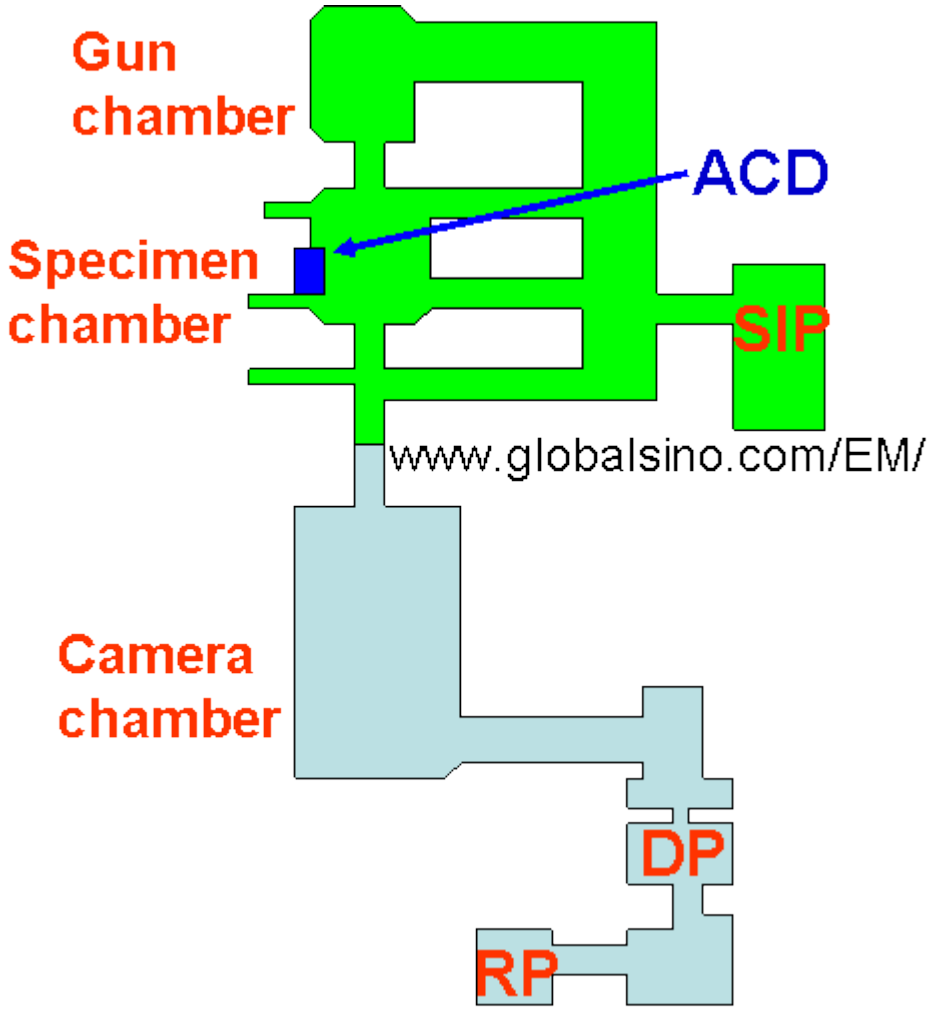
Key components of electron microscopes

- Electron source with high coherence
- High vacuum inside column
- Electron lenses
- Anti-contamination device
- Highly sensitive stage (piezoelectric)
- Scanning coils



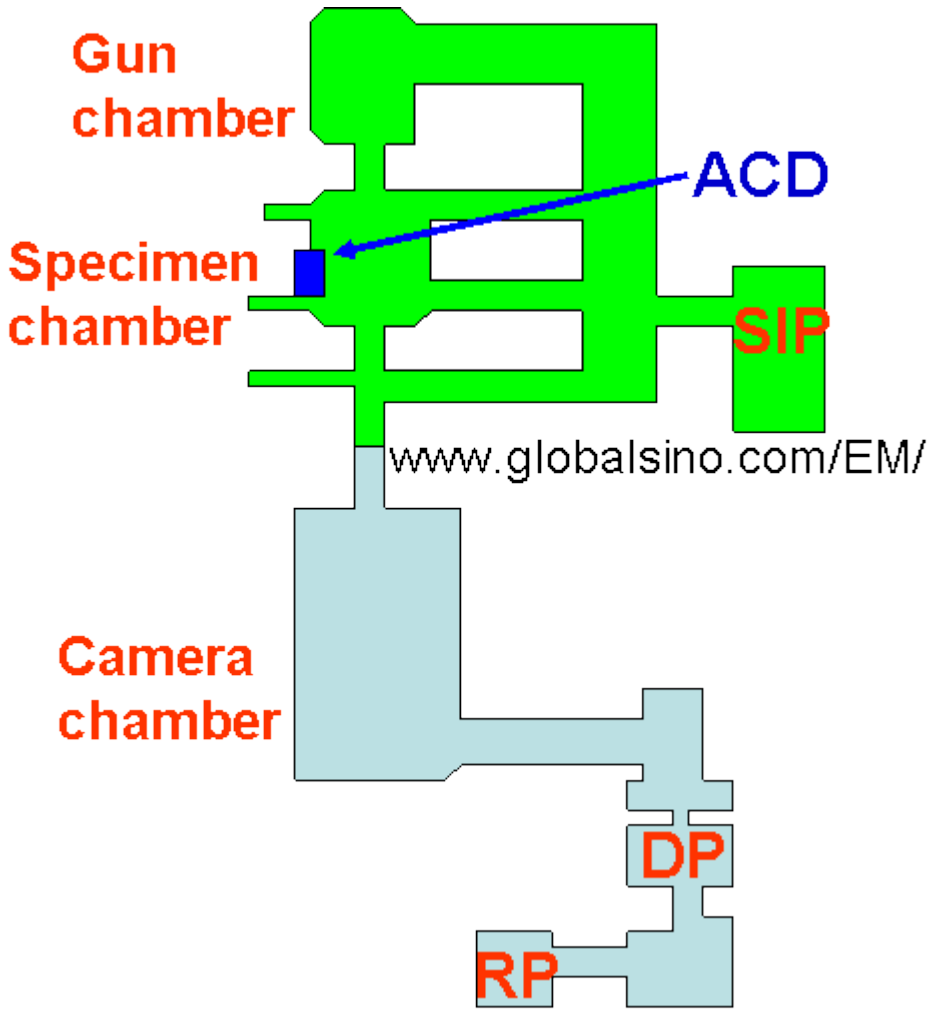
Key components of electron microscopes

- Electron source with high coherence
- High vacuum inside column
- Electron lenses
- Anti-contamination device
- Highly sensitive stage (piezoelectric)
- Scanning coils



Key components of electron microscopes

- Electron source with high coherence
- High vacuum inside column
- Electron lenses
- Anti-contamination device
- Highly sensitive stage (piezoelectric)
- Scanning coils, etc.

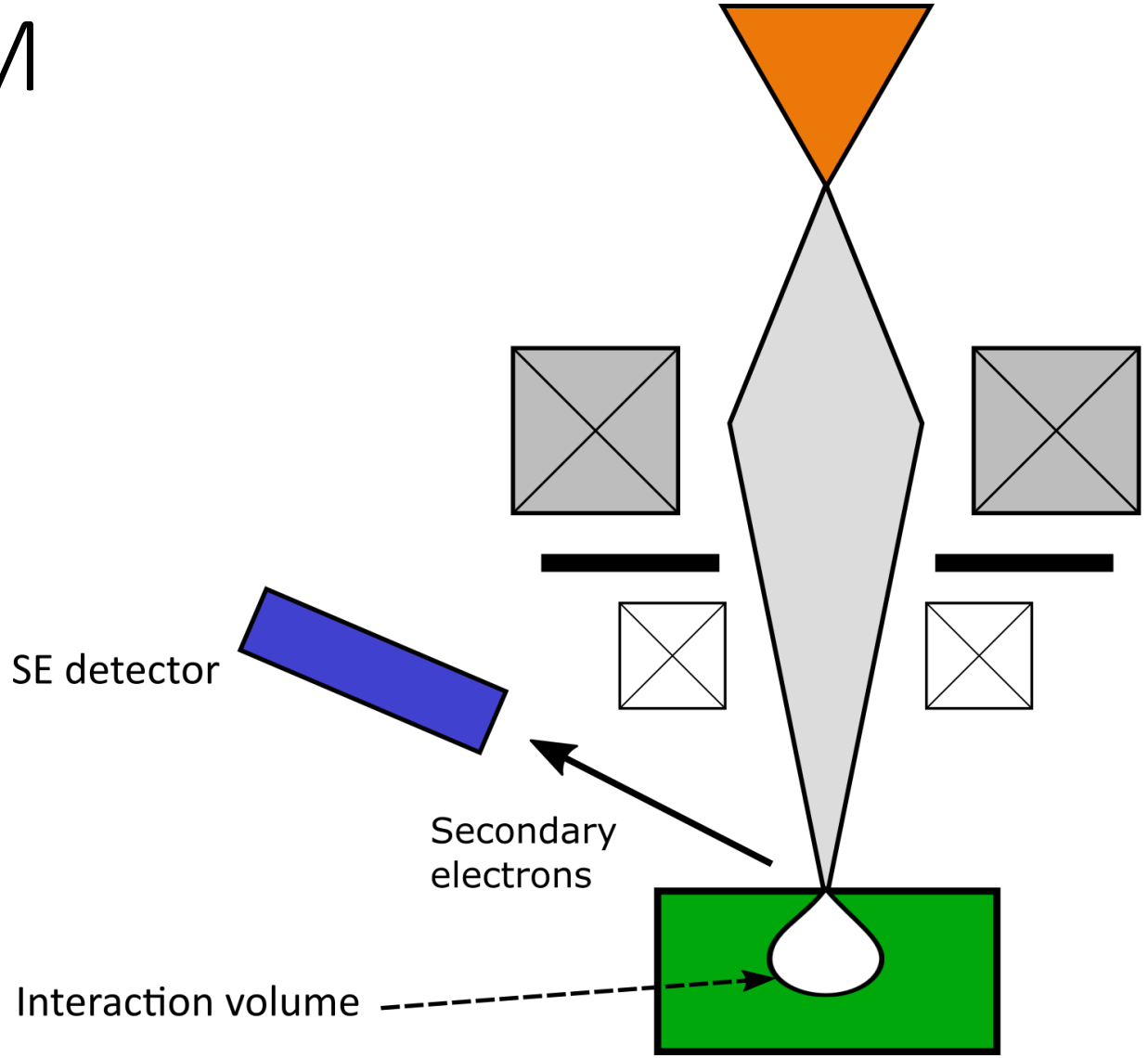


Modes of electron microscopy

Three modes of Electron Microscopy

- Scanning electron microscopy (SEM).
- Transmission electron microscopy (TEM).
- Scanning transmission electron microscopy (STEM).

SEM



Electron source

Condenser/objective lens system

Objective aperture

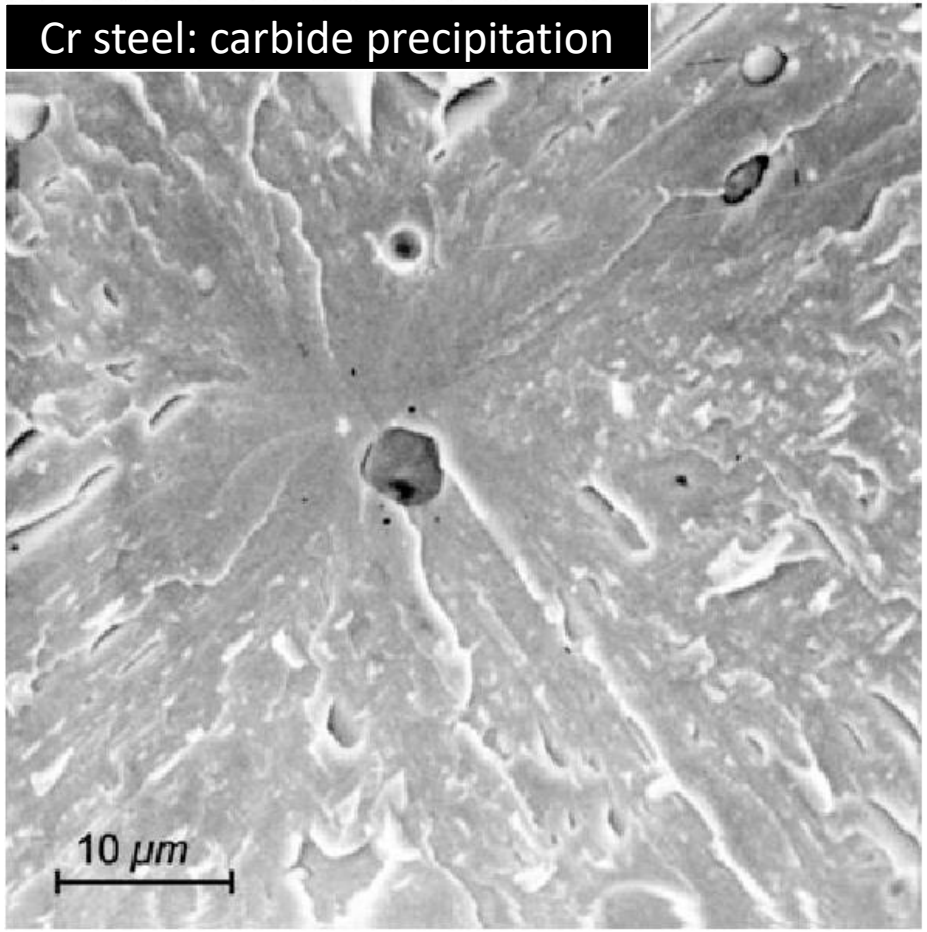
Scanning coils

Sample

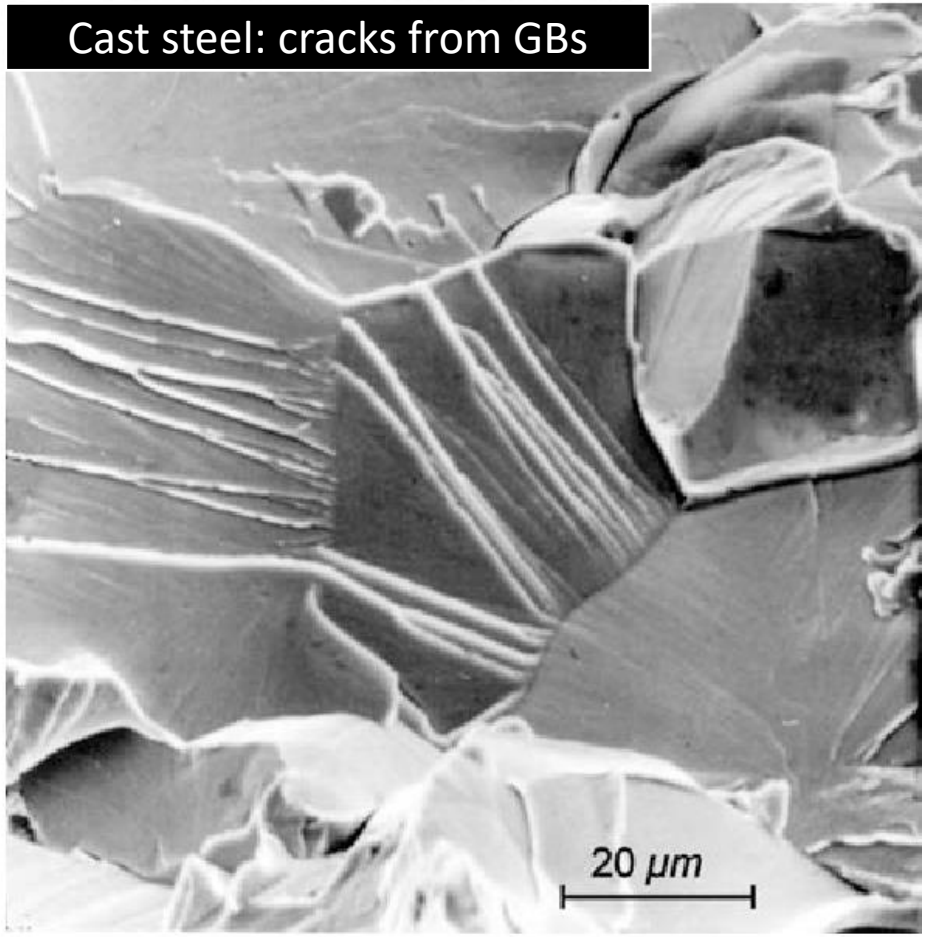
SEM - instrument

Figure from Helmholtz Energy Materials Characterization Platform
http://www.hemcp.de/zeiss-merlin-fe-sem/

SEM examples: failure analysis



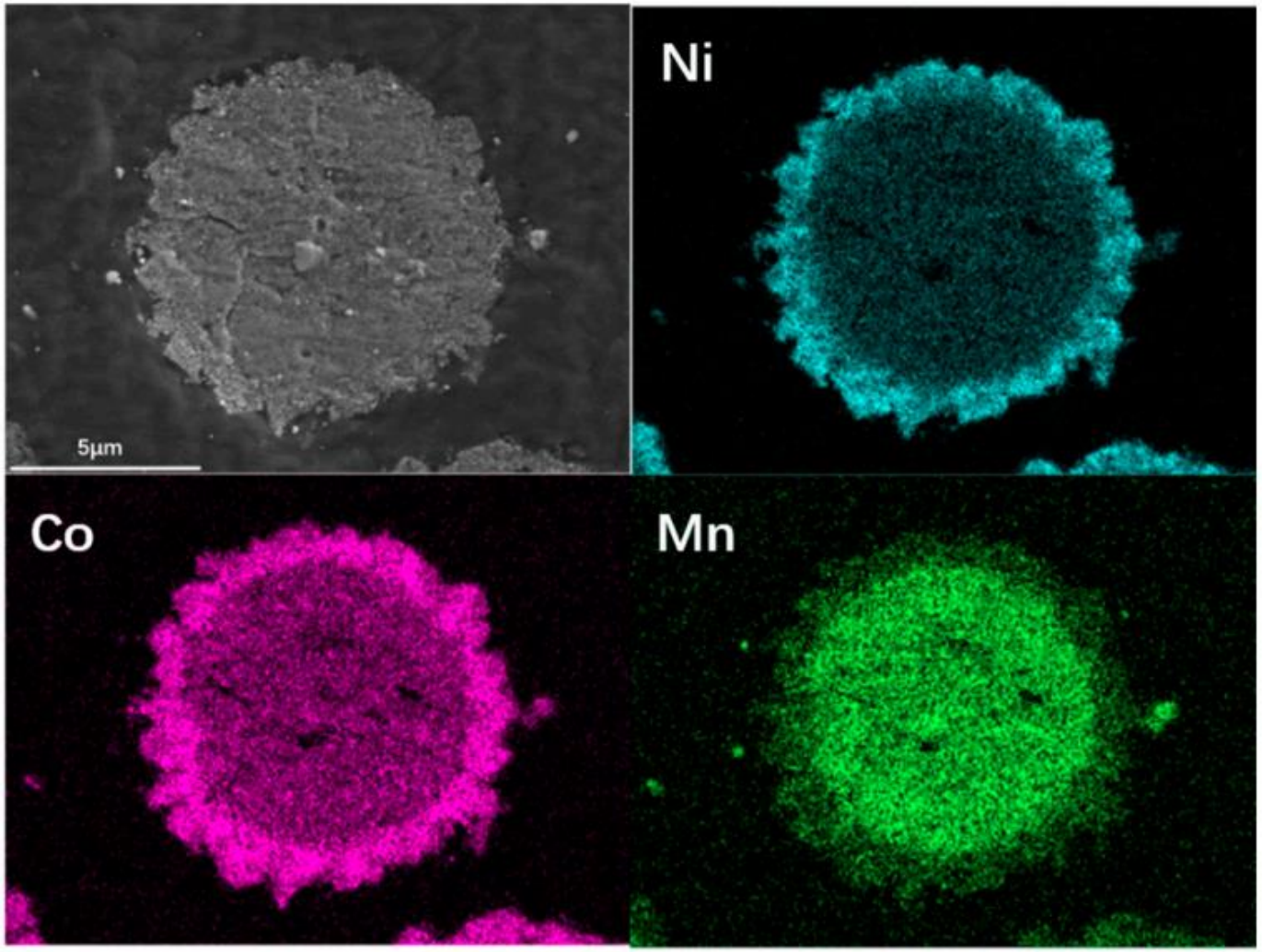
a)



b)

M Moeser, Materials Science Monographs, 2007

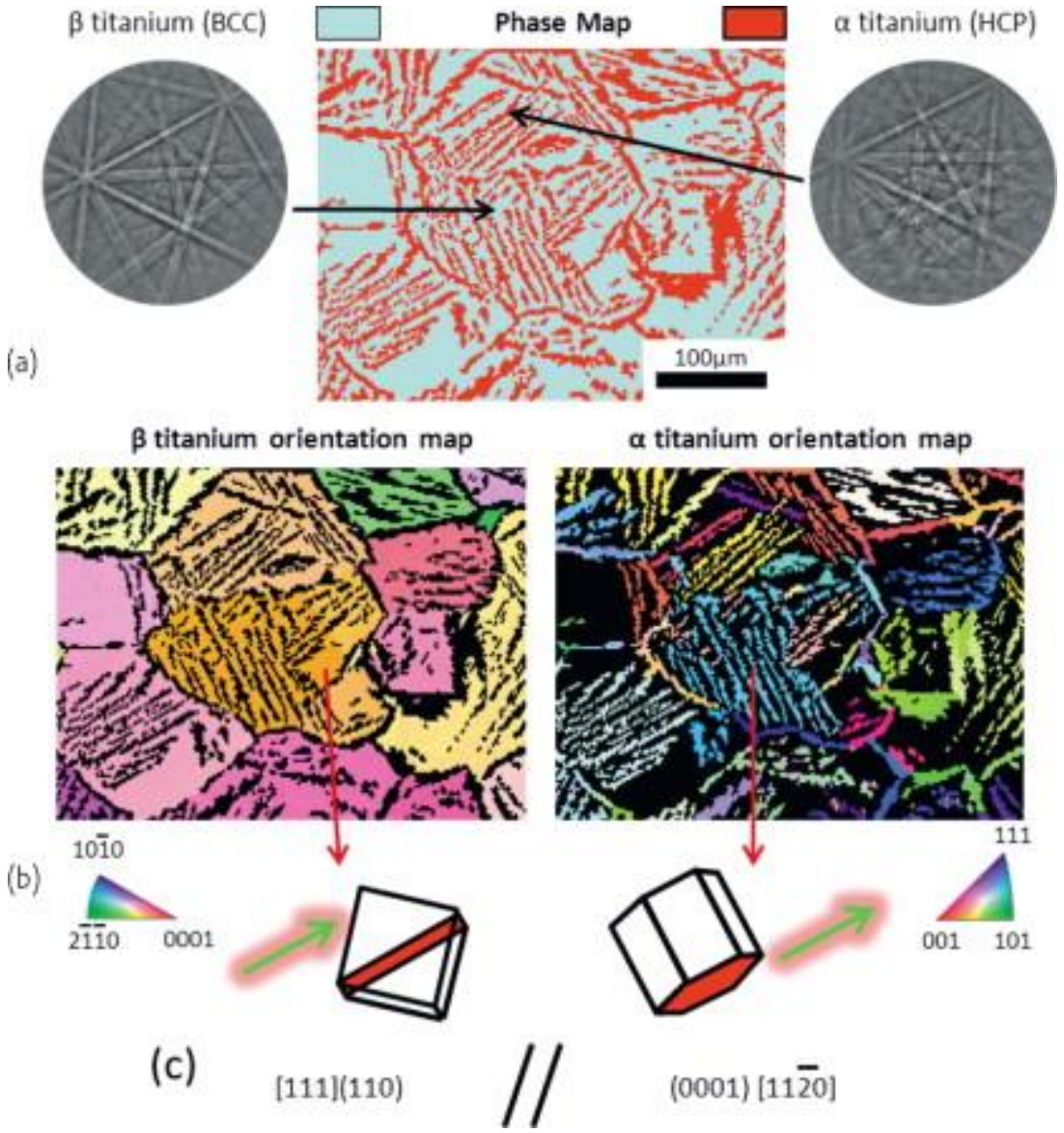
SEM examples: EDX mapping



Tang et al, Crystals **6**(11), p. 156, 2016

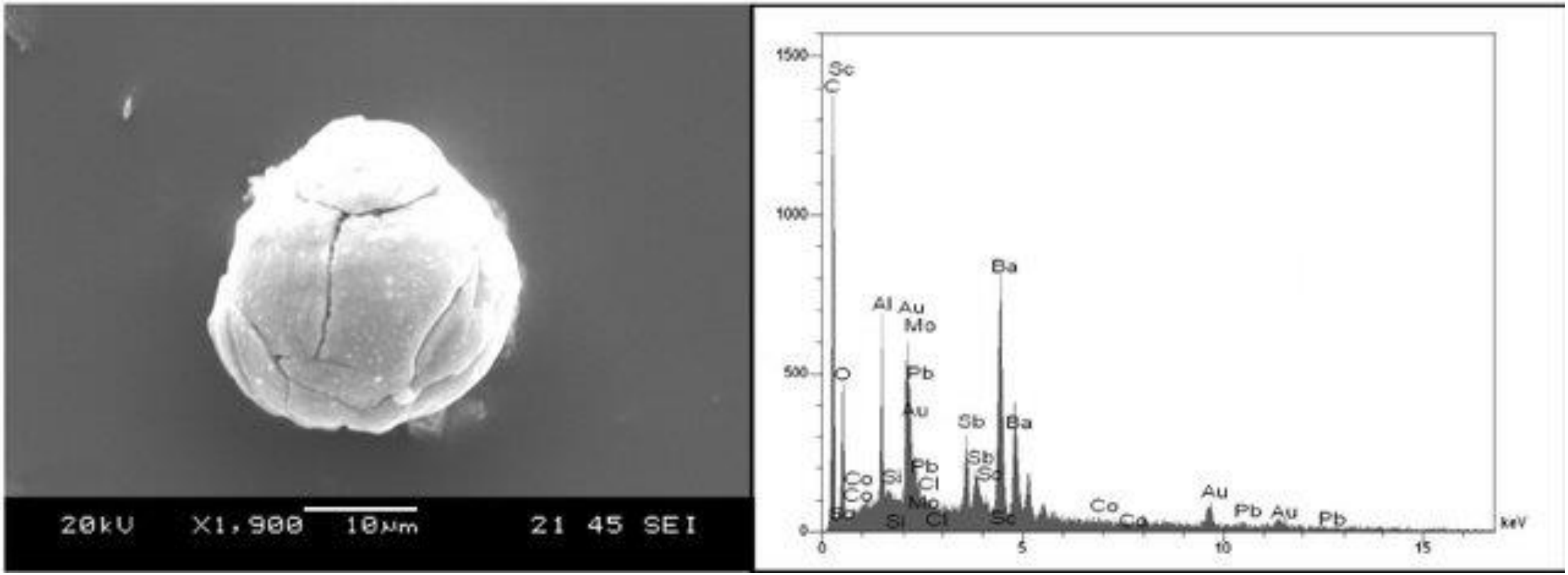
SEM examples: EBSD

- Electron backscatter diffraction (EBSD)
- Collect backscattering w/ detector
- Map grain size, phase, orientation
- Example: Titanium Alloy
- Geology, metallurgy, nuclear energy, organic films, etc



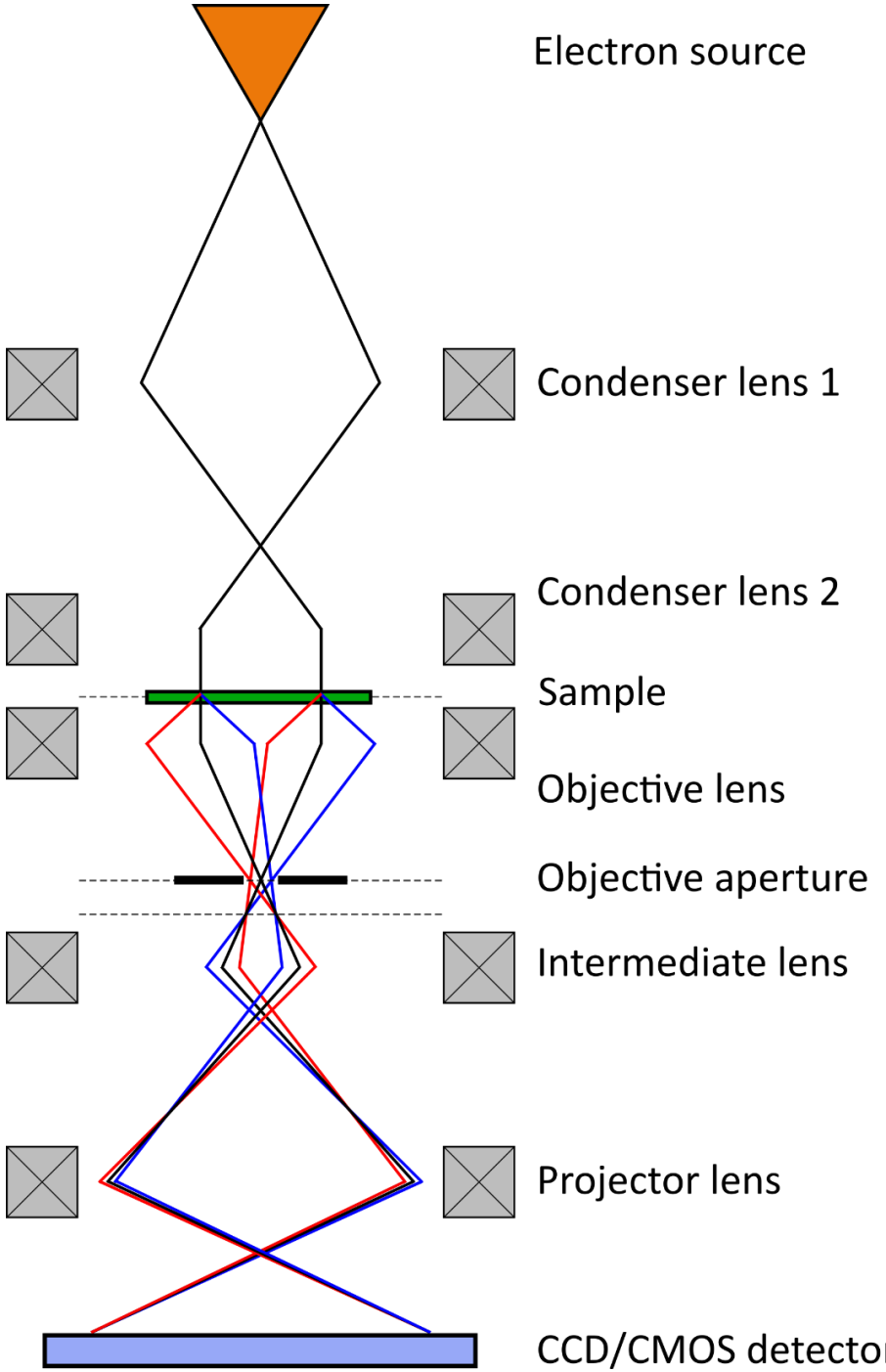
*Wilkinson & Britton, Materials Today **15**(9), pp. 366-376, 2012*

SEM examples: forensics



Silva et al, J. Braz. Chem. Soc. 20 (10), 2009

TEM

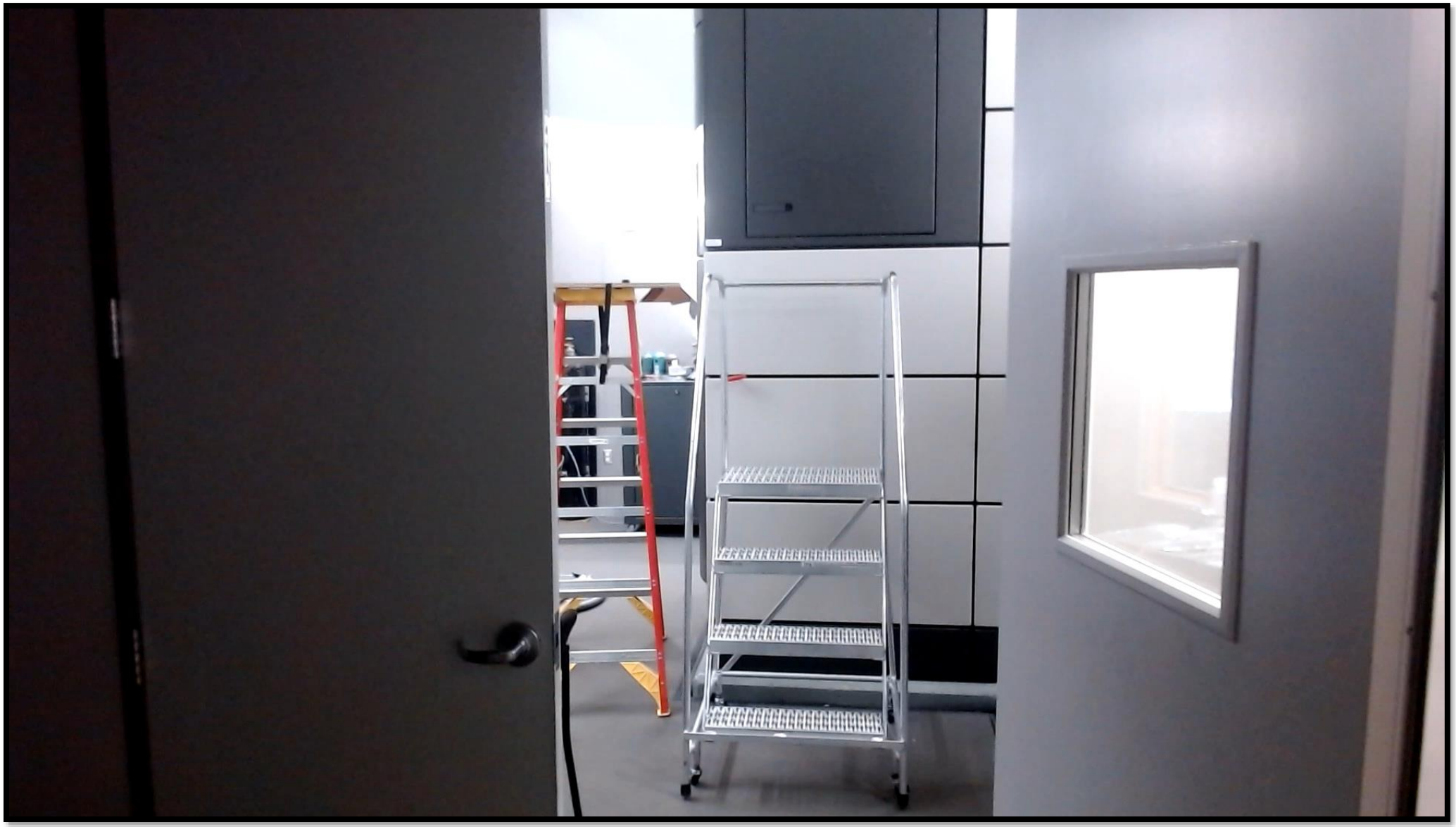

Electron source
Condenser lens 1
Condenser lens 2
Sample
Objective lens
Objective aperture
Intermediate lens
Projector lens
CCD/CMOS detector

S/TEM - Instrument

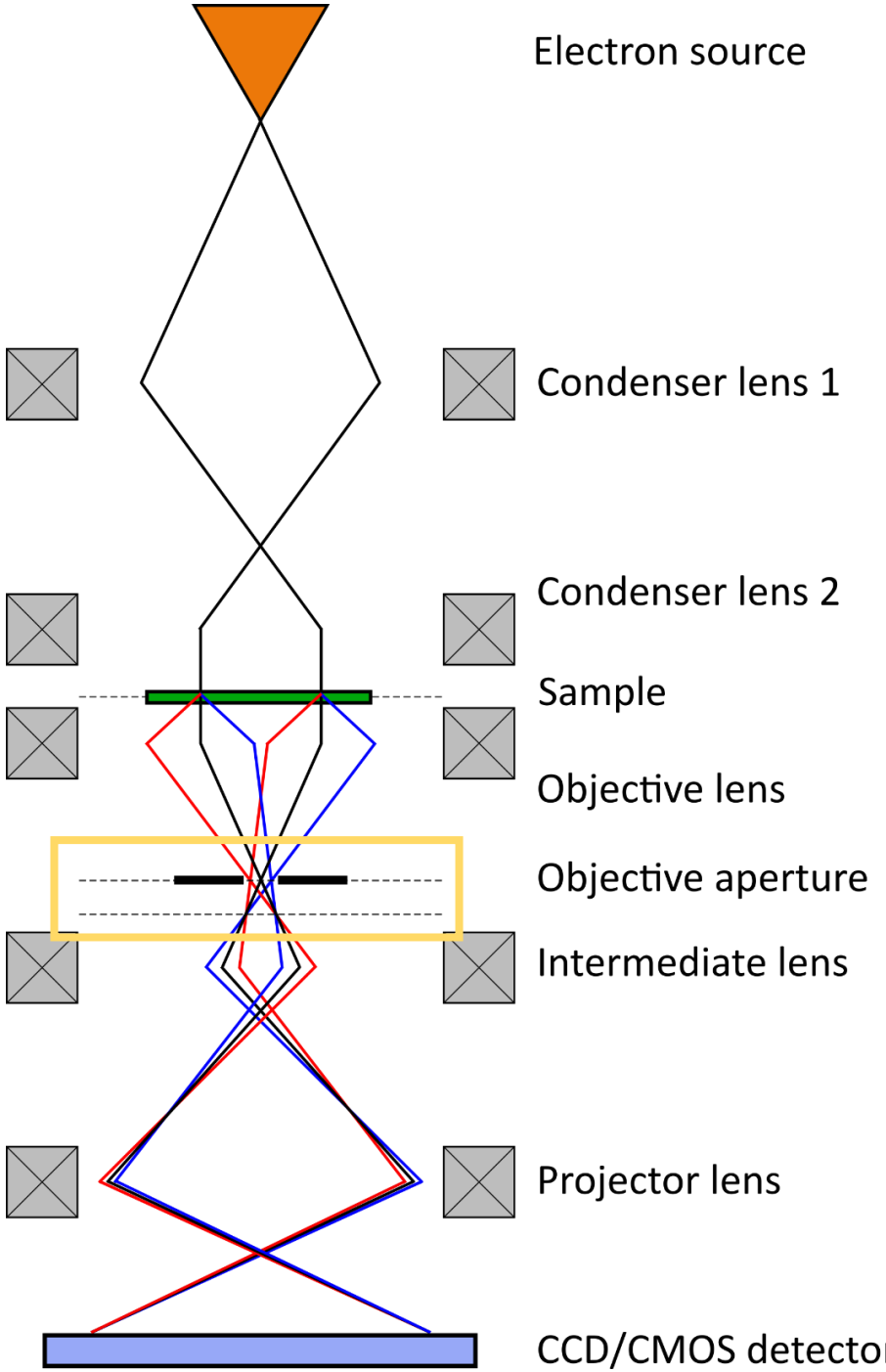
FEI Tecnai

TEAM I

TEAM I



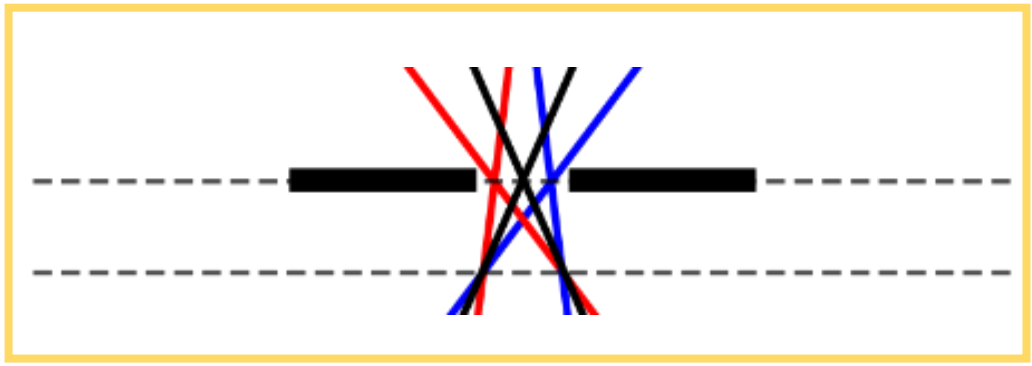
TEM


Electron source
Condenser lens 1
Condenser lens 2
Sample
Objective lens
Objective aperture
Intermediate lens
Projector lens
CCD/CMOS detector

TEM imaging and diffraction

Back-focal plane

Selected area plane



Reciprocal space

Real space

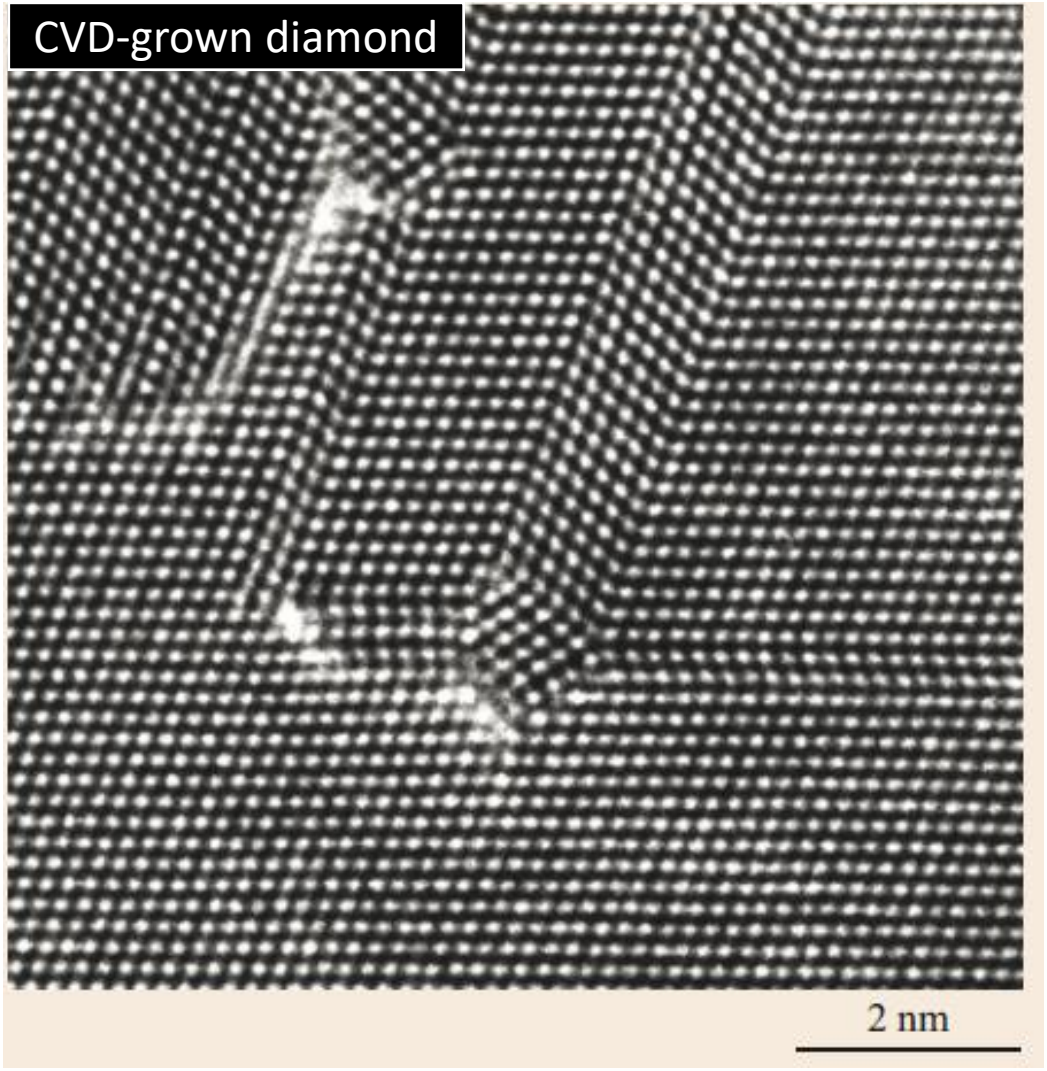
- **Back-focal plane**: Objective aperture controls acceptance angle of imaging electrons.

- **Selected area plane**: Insert aperture to select region for diffraction studies.

TEM examples – imaging

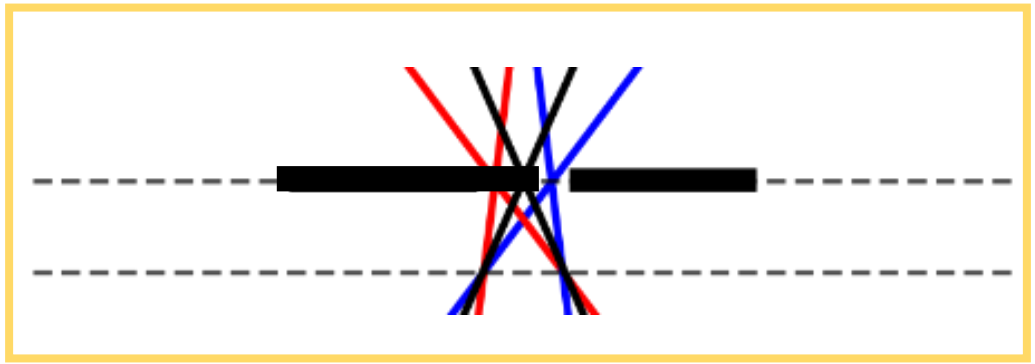
- HR imaging of crystal grains, boundaries and imperfections.

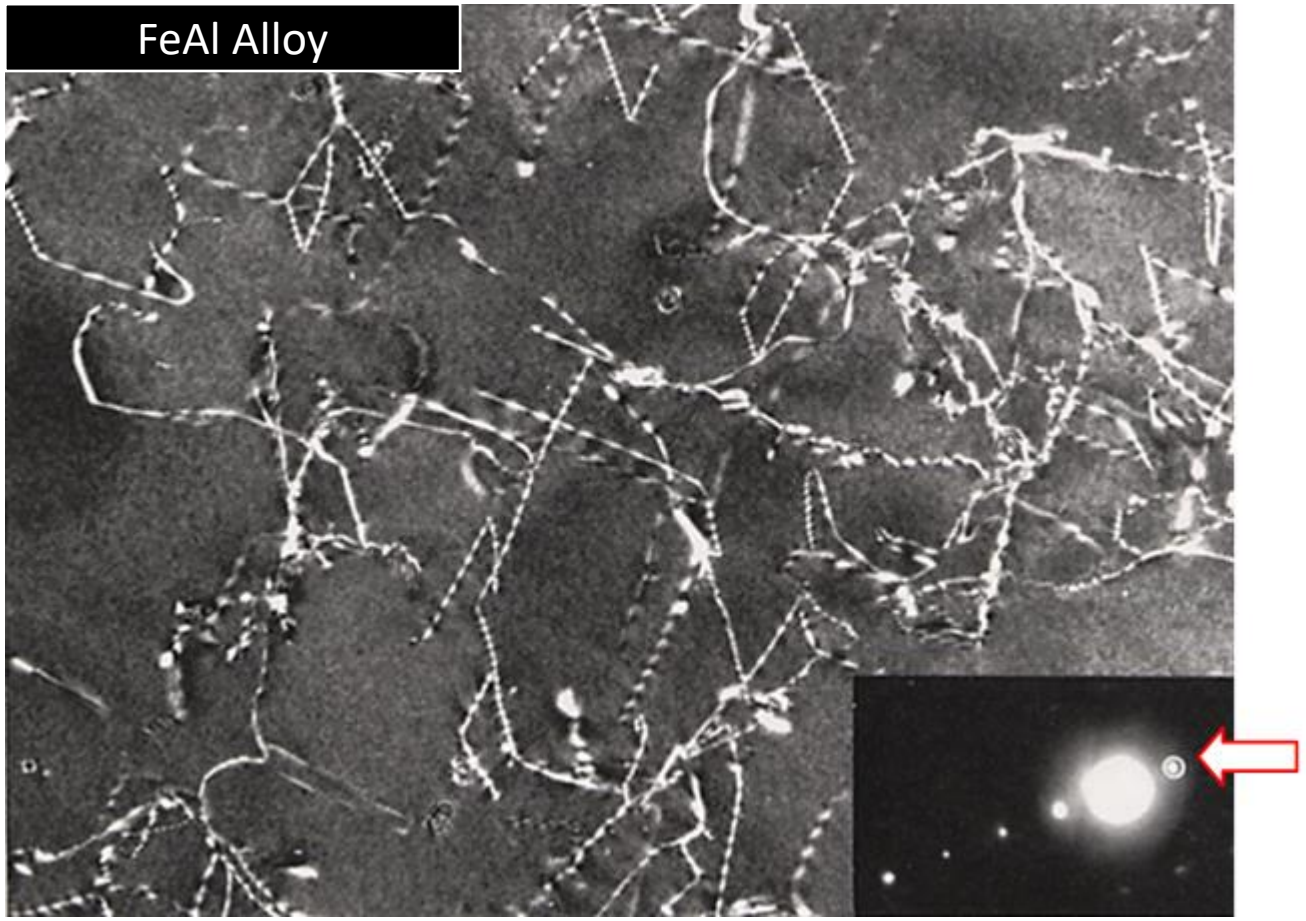
- Metallurgy, semiconductors, aerospace, catalysis, nuclear science, renewable energy, etc.



TEM examples – imaging

- Dark-field imaging for analysis of defects in a material.



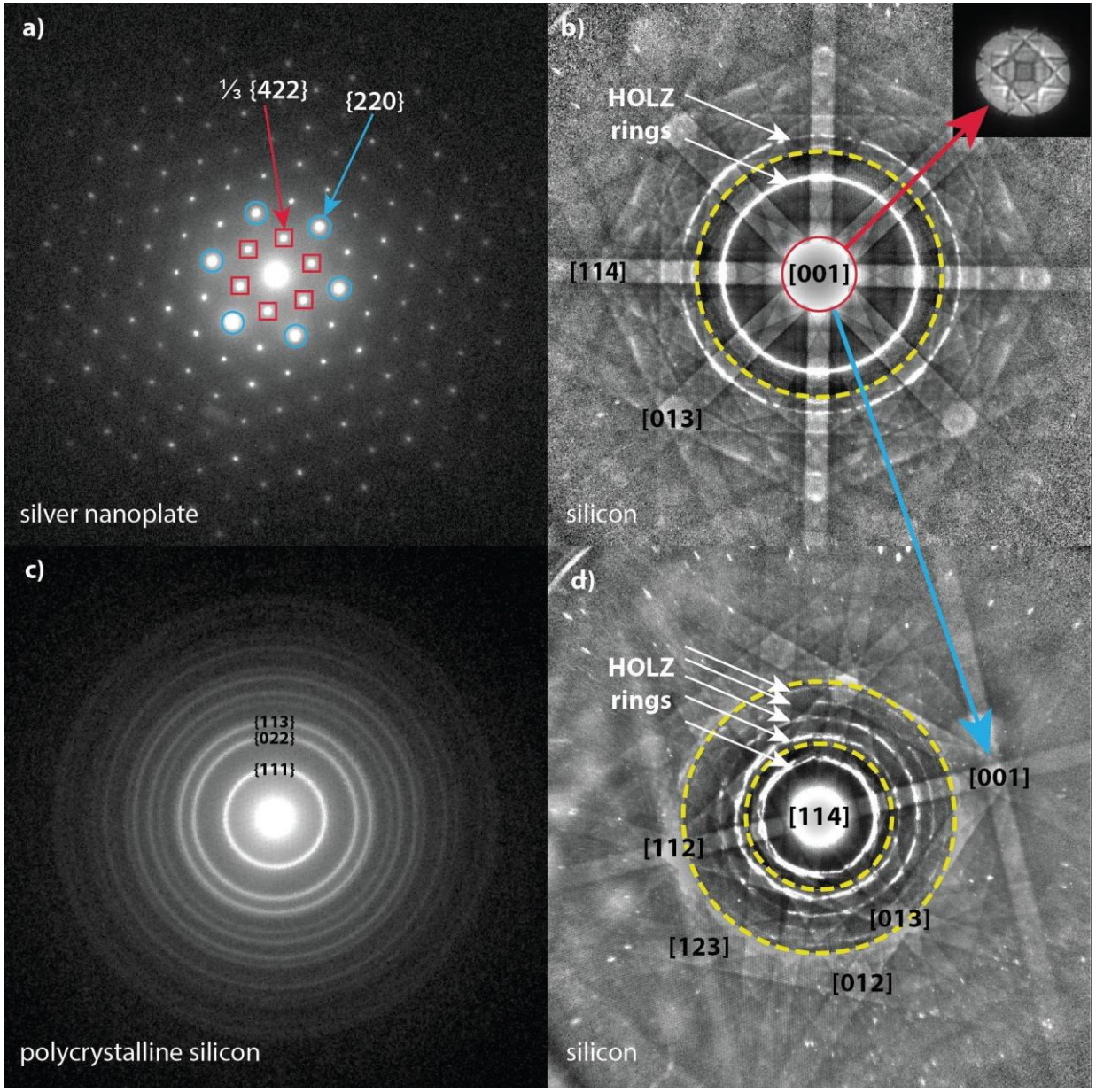


From: JEOL Glossary: Dark-field TEM, https://www.jeol.co.jp/en/words/emterms/search_result.html?keyword=dark-field%20image

TEM examples – diffraction

- Characterization of crystalline, polycrystalline and amorphous materials.

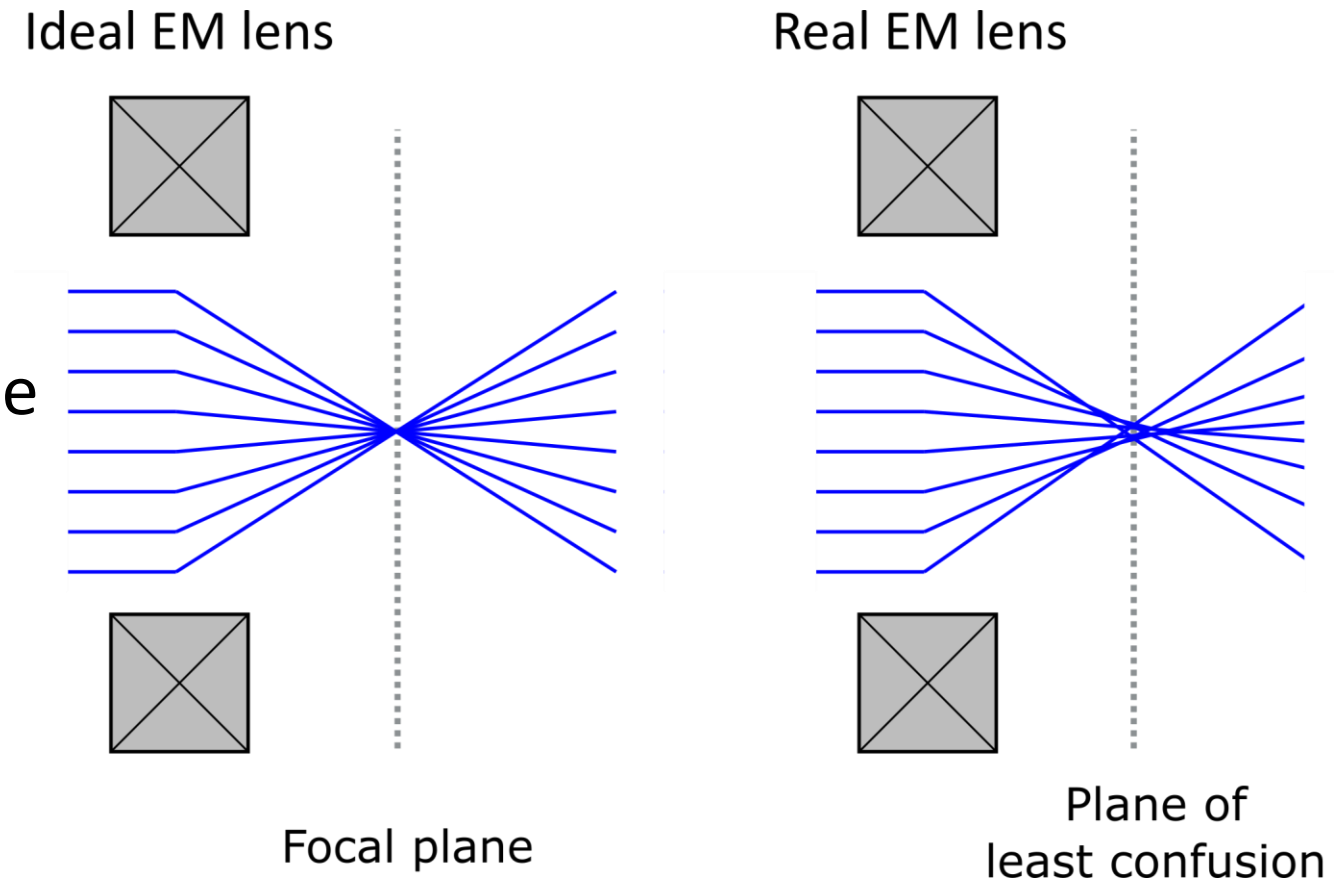
- Atomic resolution information over larger area than imaging.



BW Caplins, NIST, Analytical Transmission Scanning Electron Microscopy
https://www.nist.gov/programs-projects/analytical-transmission-scanning-electron-microscopy

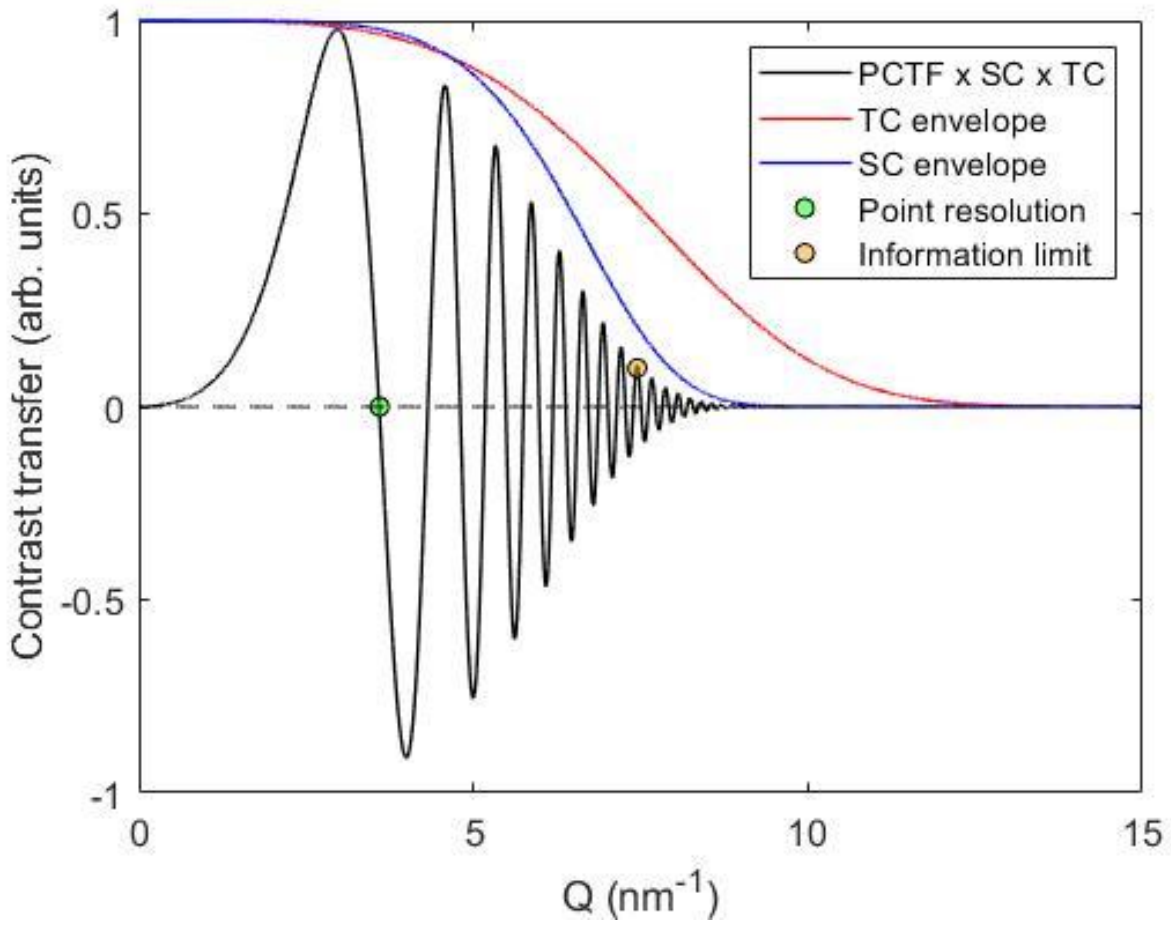
Electron lenses

- Resolution is hindered by inherently poor electron lenses.
- Electrons closer to lens reach the optic axis sooner.
- Causes a blurring effect, i.e. looking through a soda bottle.



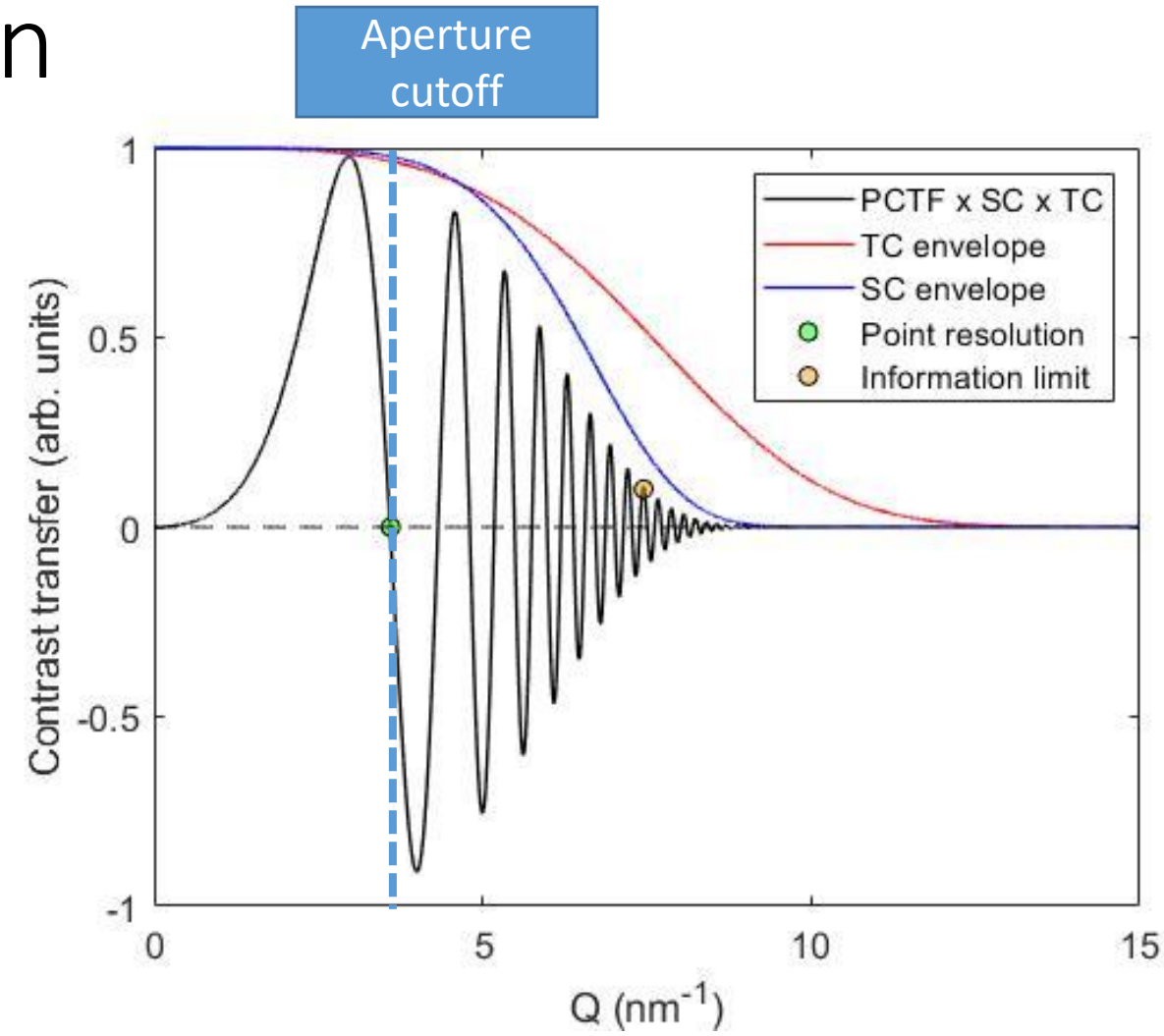
Contrast transfer function

- Positive: bright background, dark atomic features.
- Negative: dark background, bright atomic features.
- Zero: no contrast.



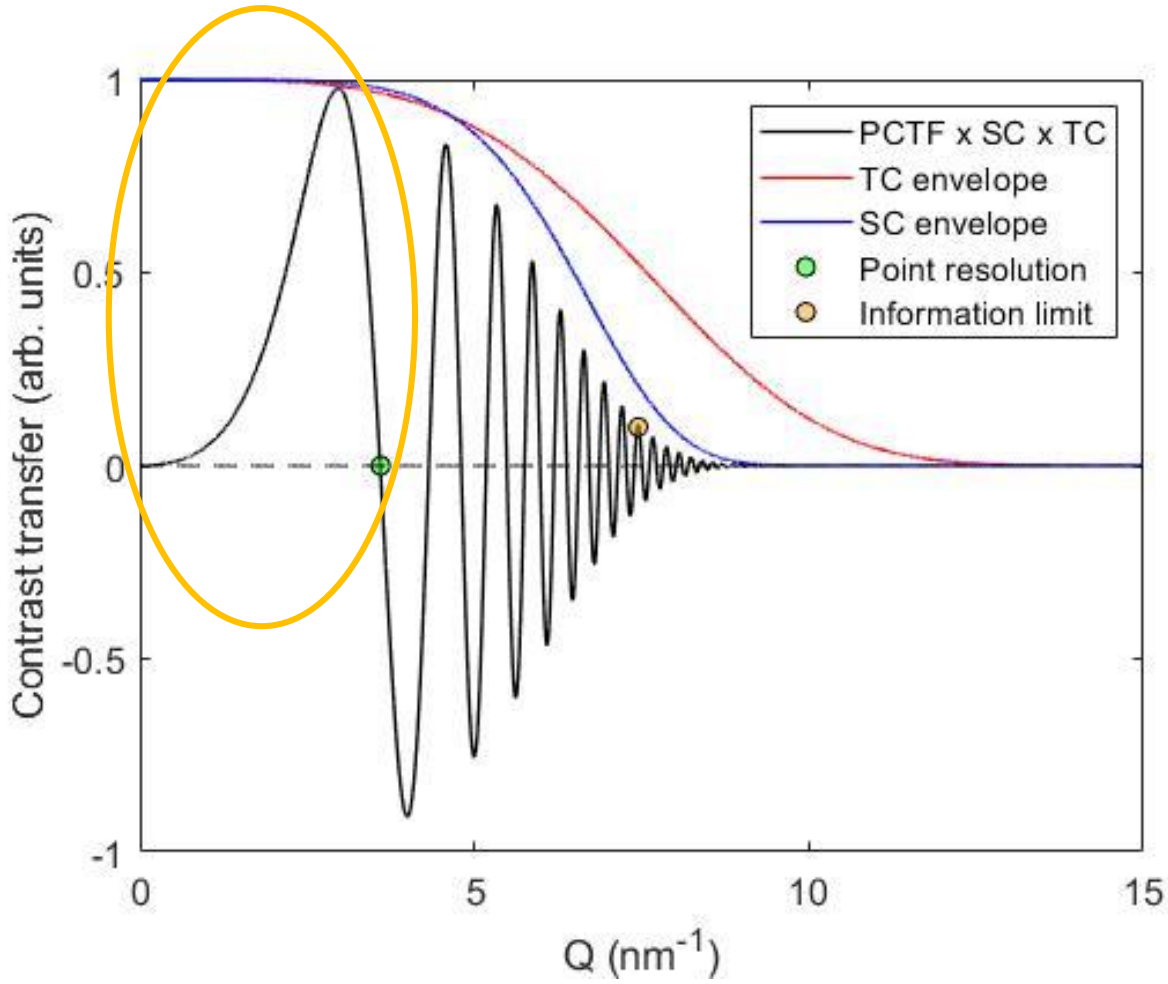
Contrast transfer function

- Insert objective aperture to block off oscillations



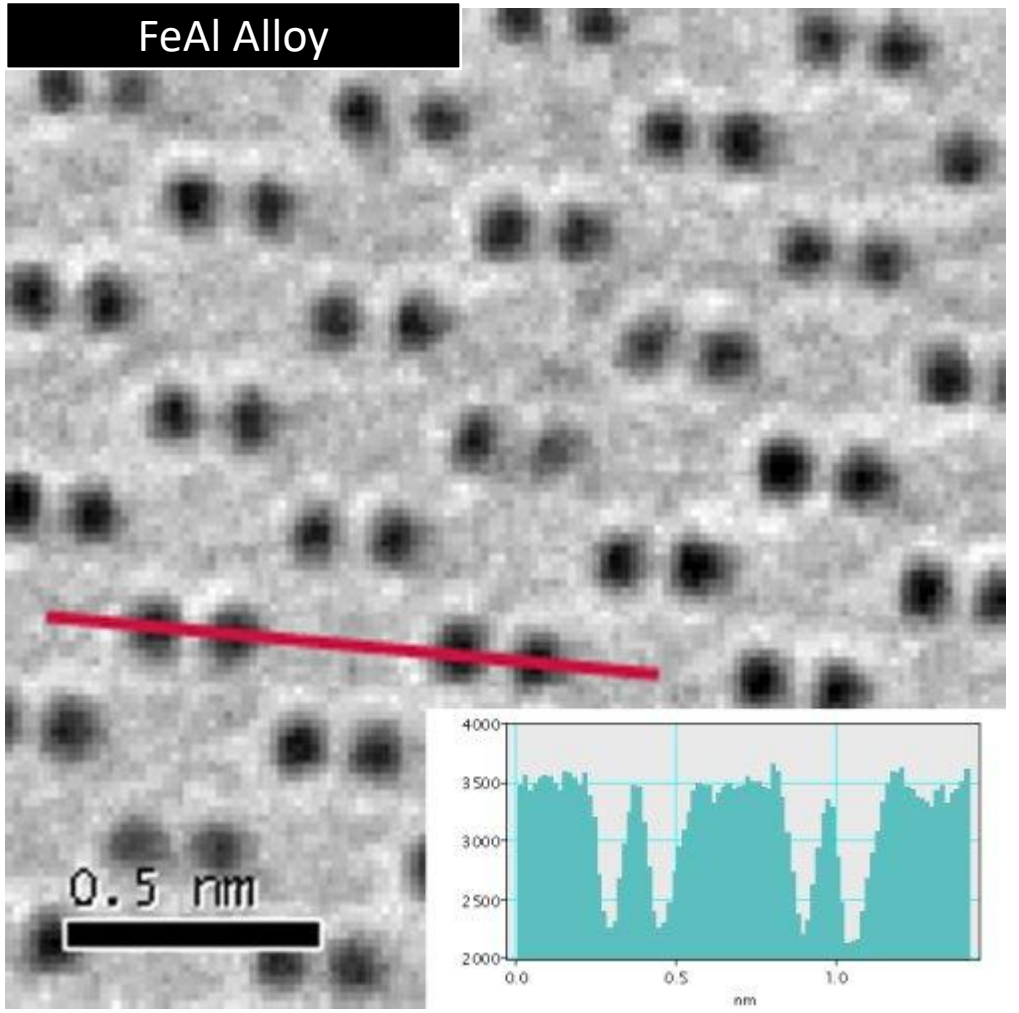
Contrast transfer function (CTF)

- Low contrast at low spatial frequencies.
- Can change the aberrations.
- Phase plates can be inserted.
- Aberration correctors.



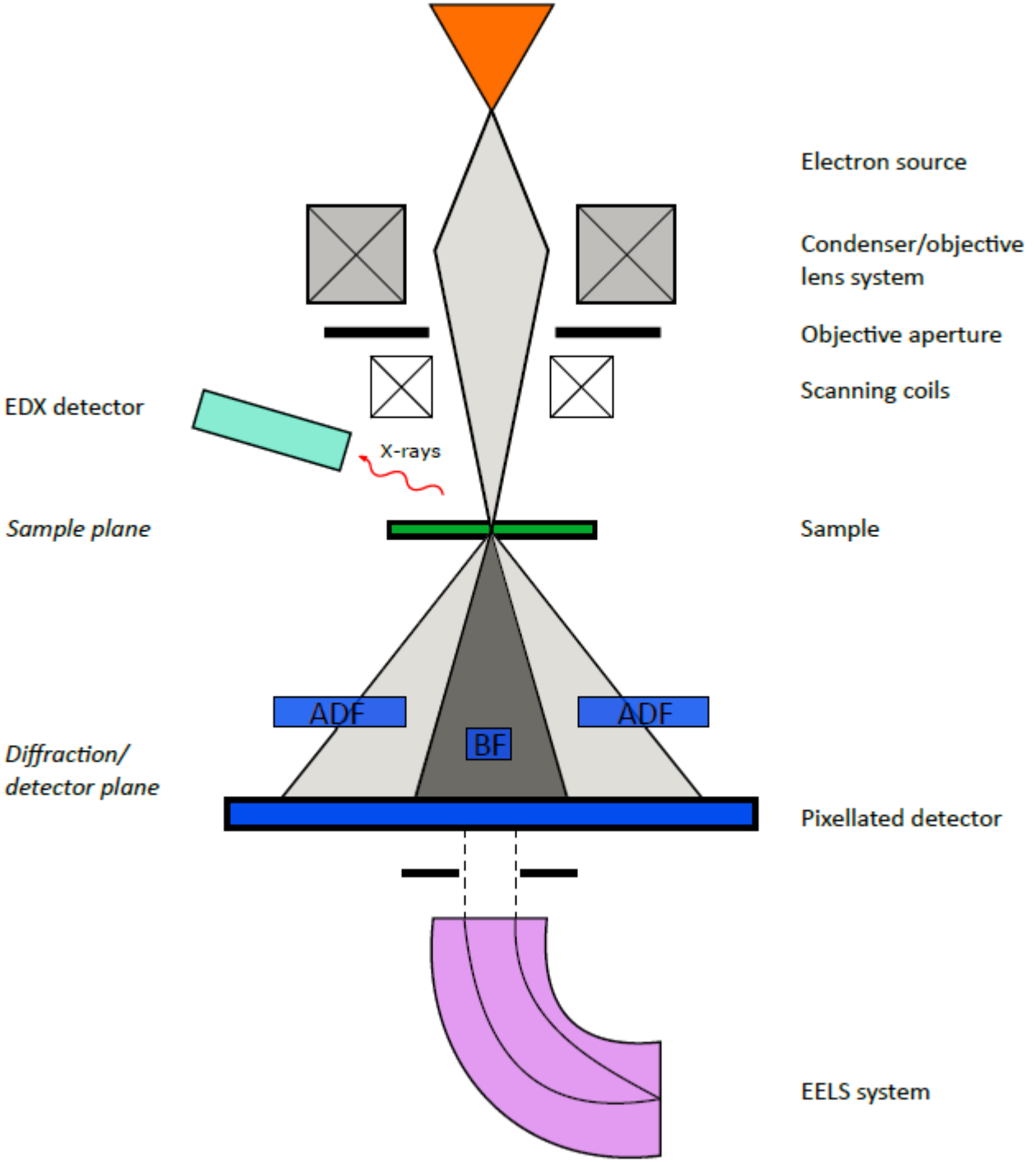
Aberration-corrected electron microscopy

- Use multipole electron lens
- Generate negative spherical aberration
- Atomic-resolution S/TEM.



Physics World, Surfaces and Interfaces Research Update, 2007
Image courtesy of ThermoFisher Scientific

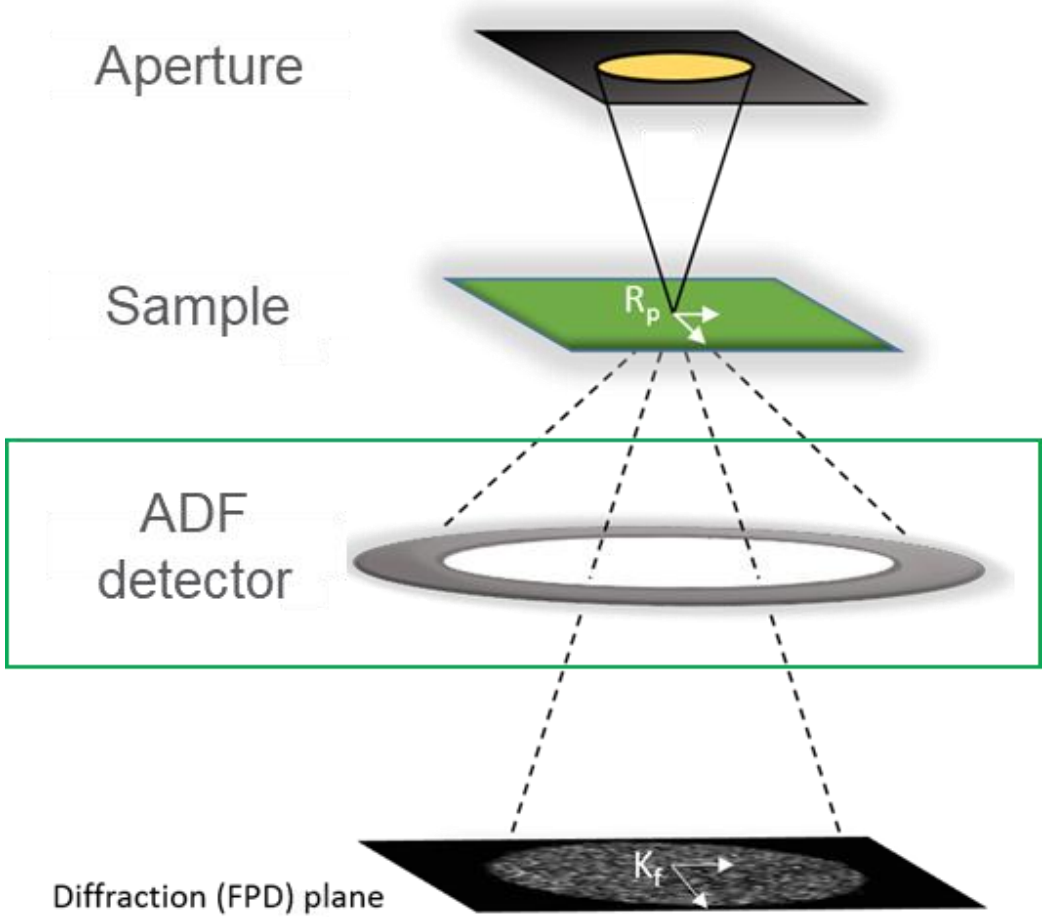
STEM

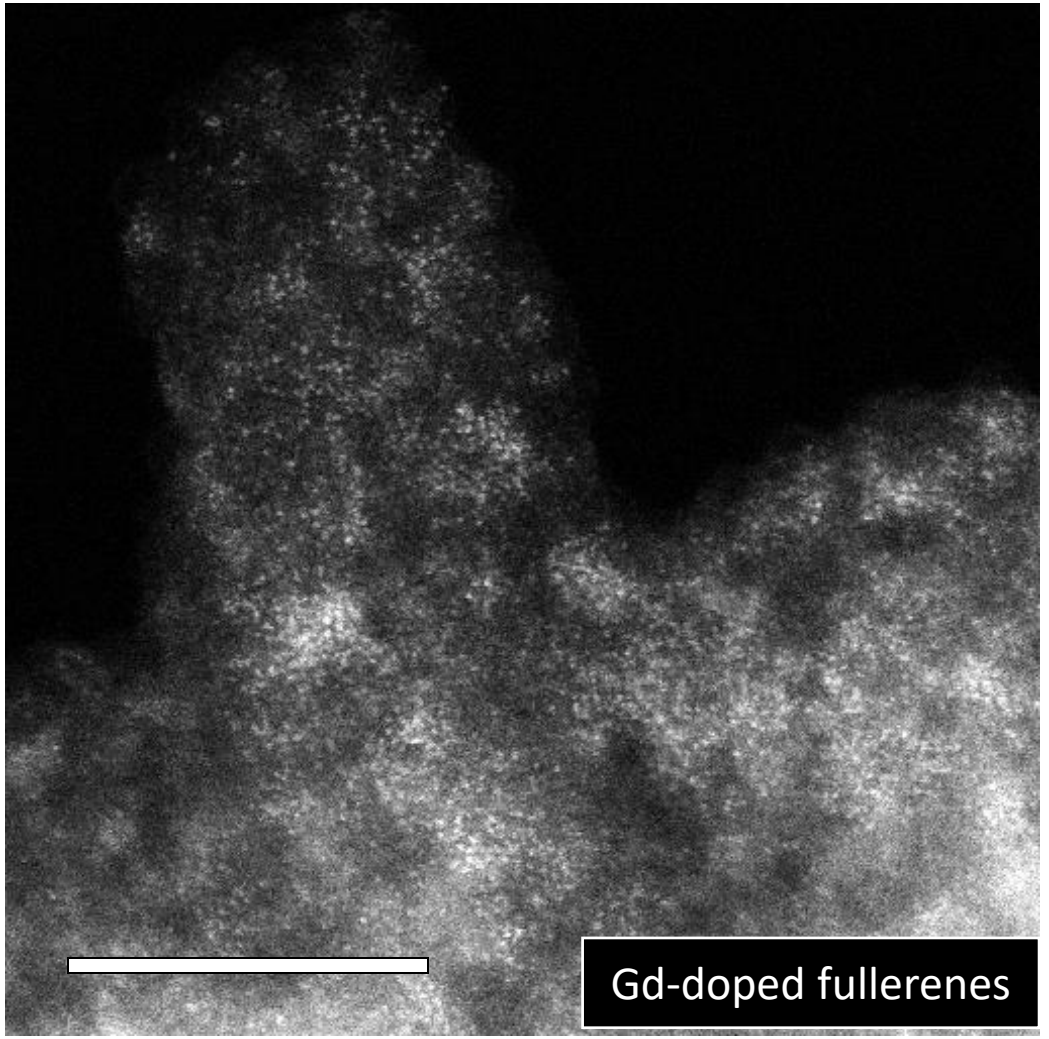

Electron source
Condenser/objective lens system
Objective aperture
Scanning coils
EDX detector
X-rays
Sample plane
Sample
ADF
BF
ADF
Diffraction/ detector plane
Pixellated detector
EELS system

STEM

- A convergent beam is scanned across a thin sample.
- Collect coherent/incoherent/inelastic electrons, x-rays, etc.
- Can be used to determine atomic-resolution structure and composition.
- ‘Upside-down TEM’.
- Typical voltage range: 30 – 300 kV.
- Typical resolution: < 1 Å.

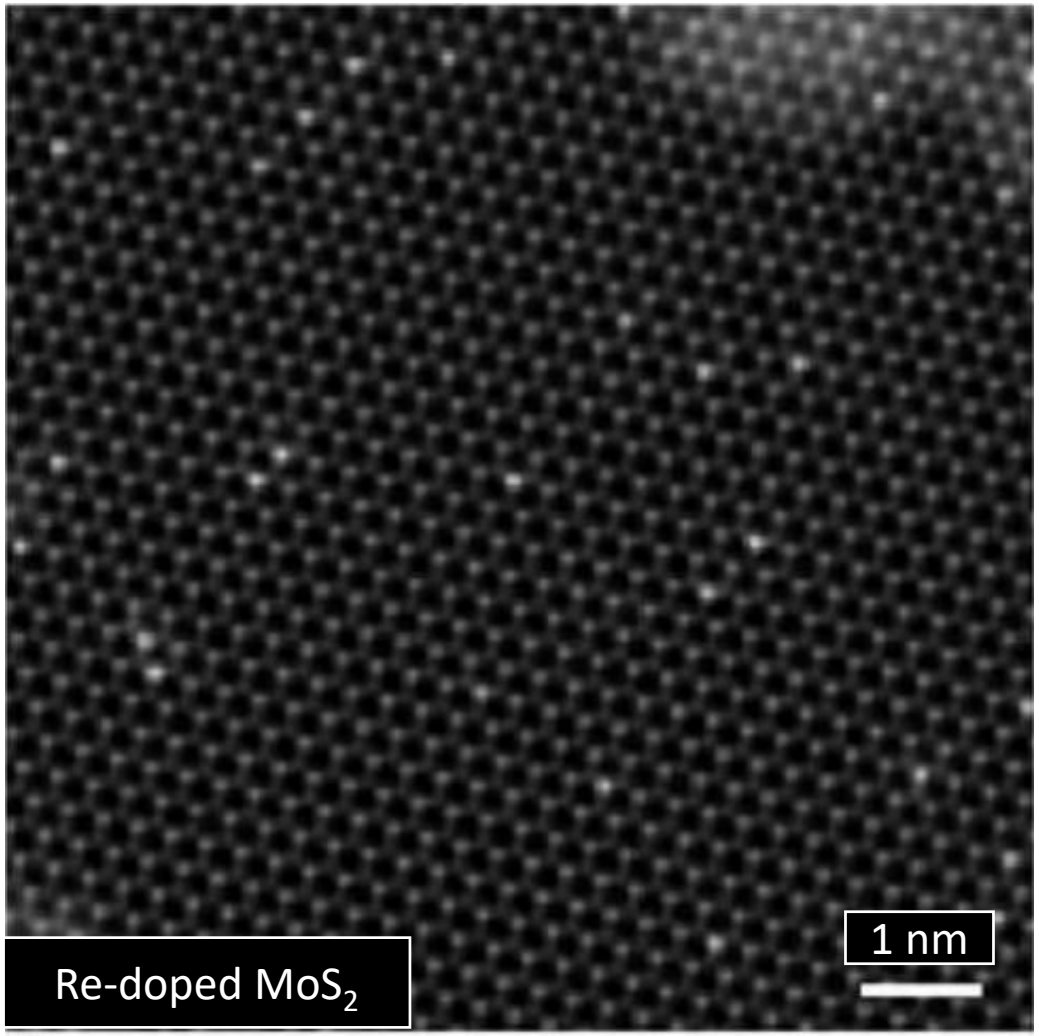
STEM examples: incoherent imaging





STEM examples: incoherent imaging

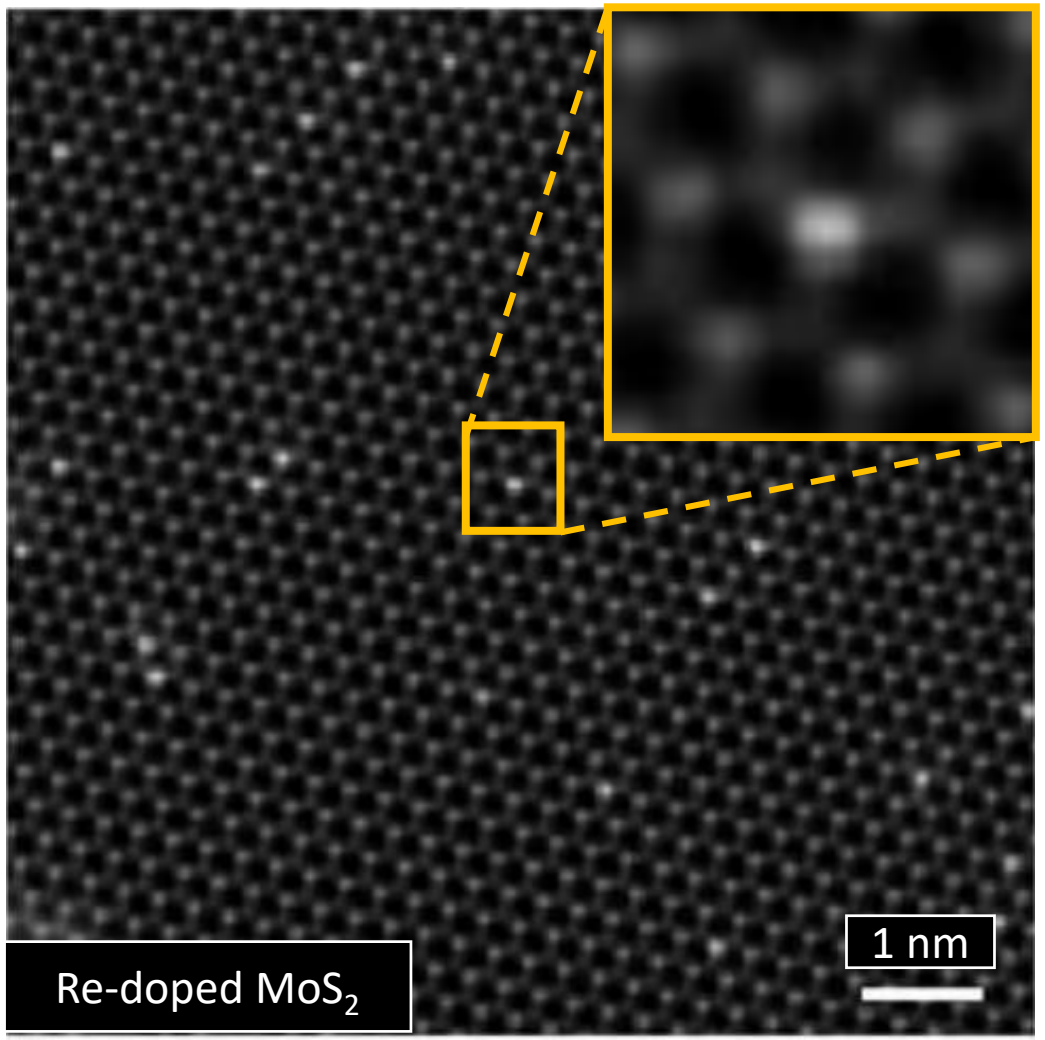
- ‘Z-contrast imaging’.
- Coherent contributions avoided using annular detector.
- Interpretable contrast
  - Can identify different elements.



*Tian et al, Nature Materials **19**, pp. 867–873, 2020*

STEM examples: incoherent imaging

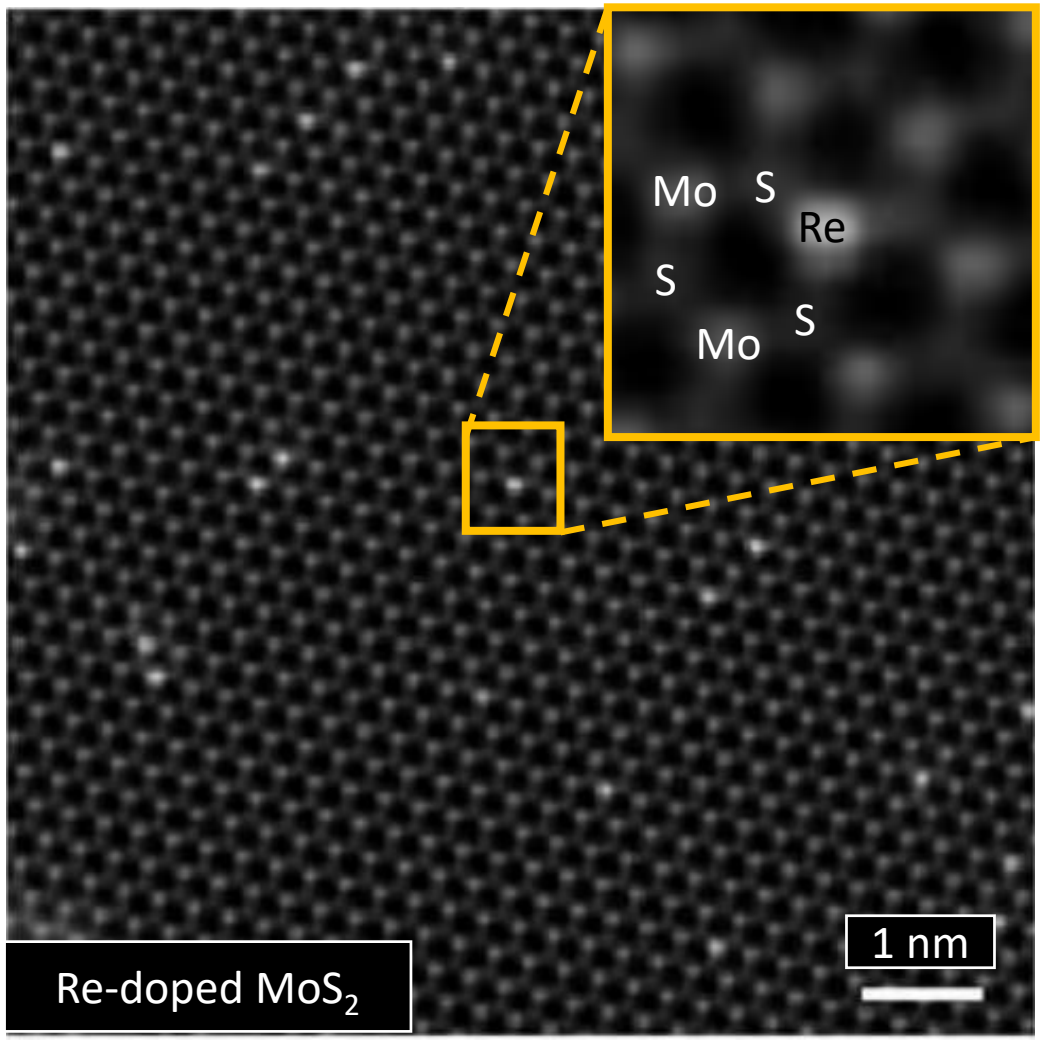
- ‘Z-contrast imaging’.
- Coherent contributions avoided using annular detector.
- Interpretable contrast
  - Can identify different elements.



*Tian et al, Nature Materials **19**, pp. 867–873, 2020*

STEM examples: incoherent imaging

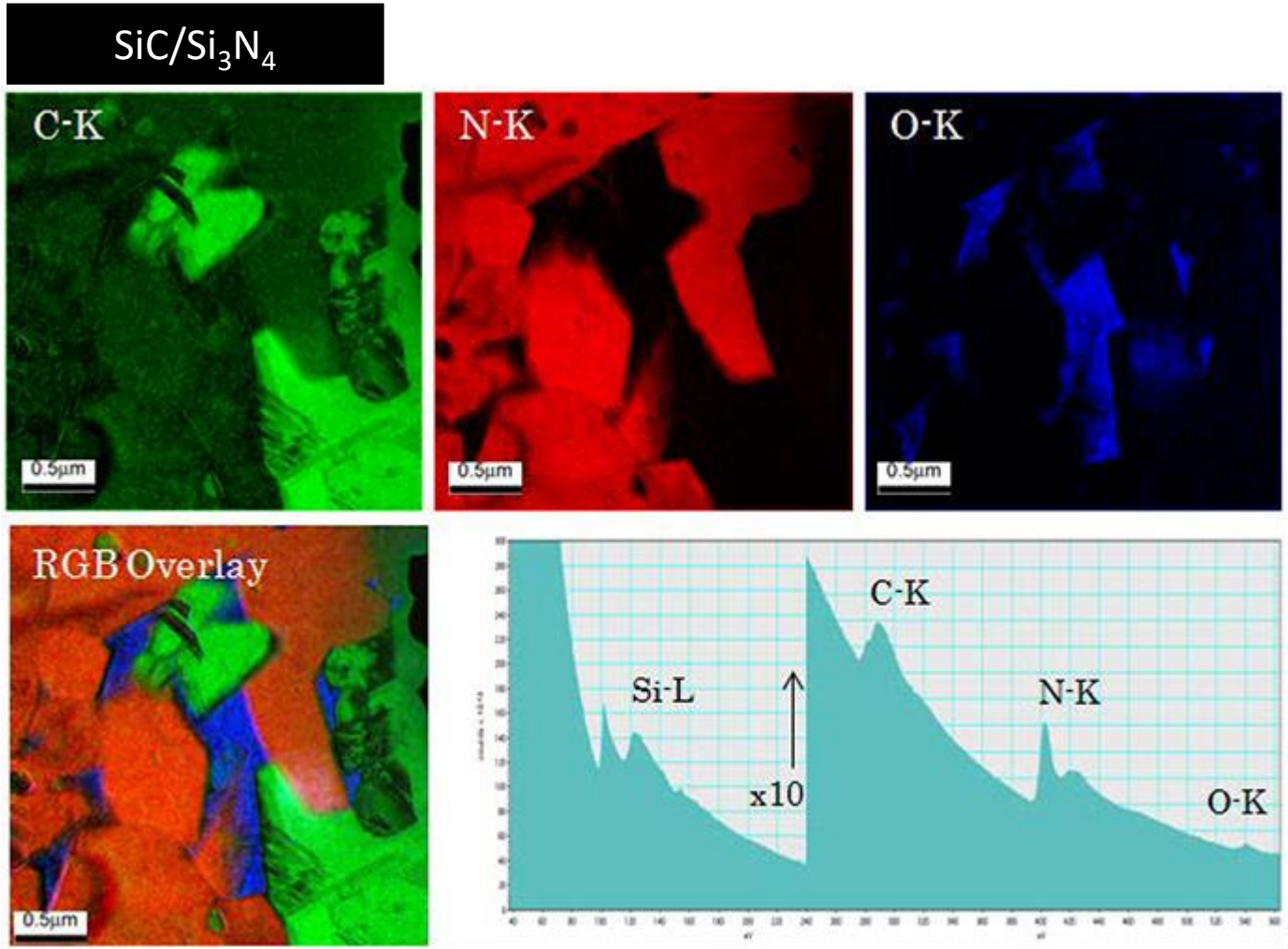
- ‘Z-contrast imaging’.
- Coherent contributions avoided using annular detector.
- Interpretable contrast
  - Can identify different elements.



*Tian et al, Nature Materials **19**, pp. 867–873, 2020*

STEM examples: EELS

- Lighter elements than EDX
- Distinguish different phases same elemental composition
- Low-loss spectra (plasmons, phonons) can be used to study the band structure



From: JEOL Ltd.
https://www.jeol.co.jp

Capabilities in electron microscopy

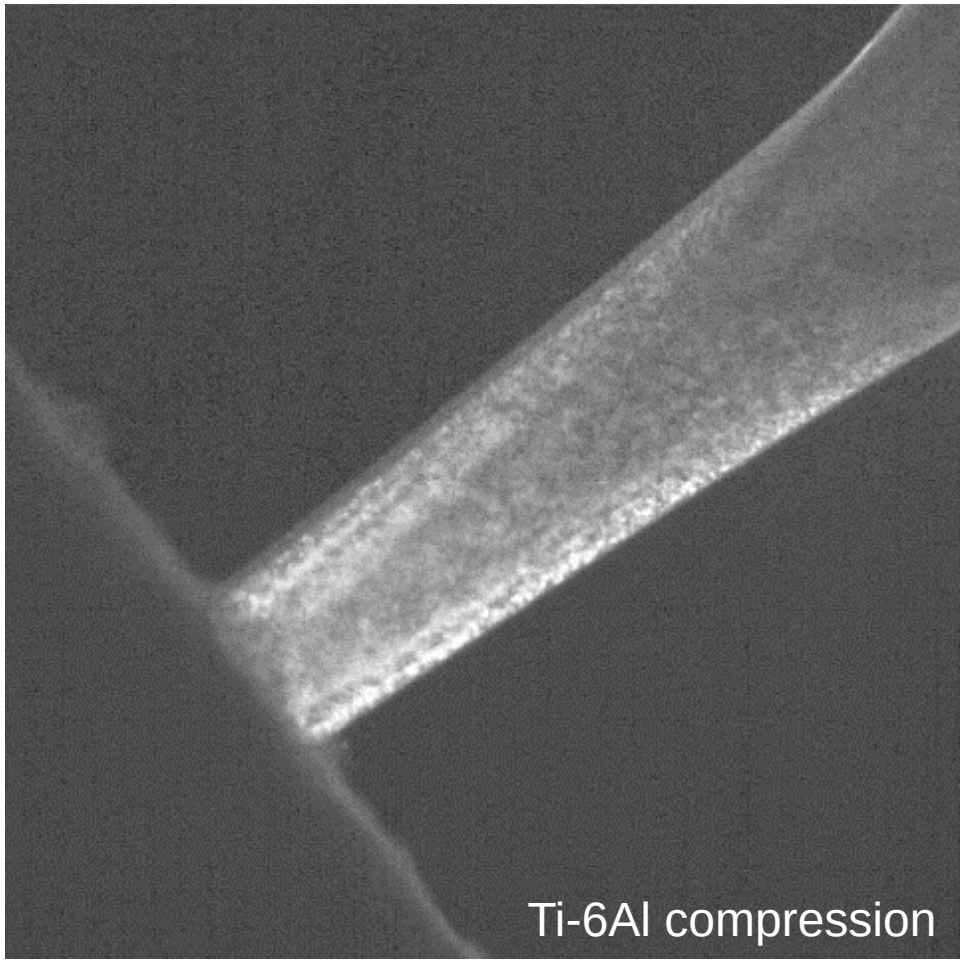
	SEM	TEM	STEM
Atomic resolution	✗	✓	✓
Thick samples	✓	✗	✗
X-ray analysis	✓	✓	✓
Ultra-fast imaging	✗	✓	✗
Energy loss studies	✗	✓	✓
Diffraction studies	✓	✓	✓

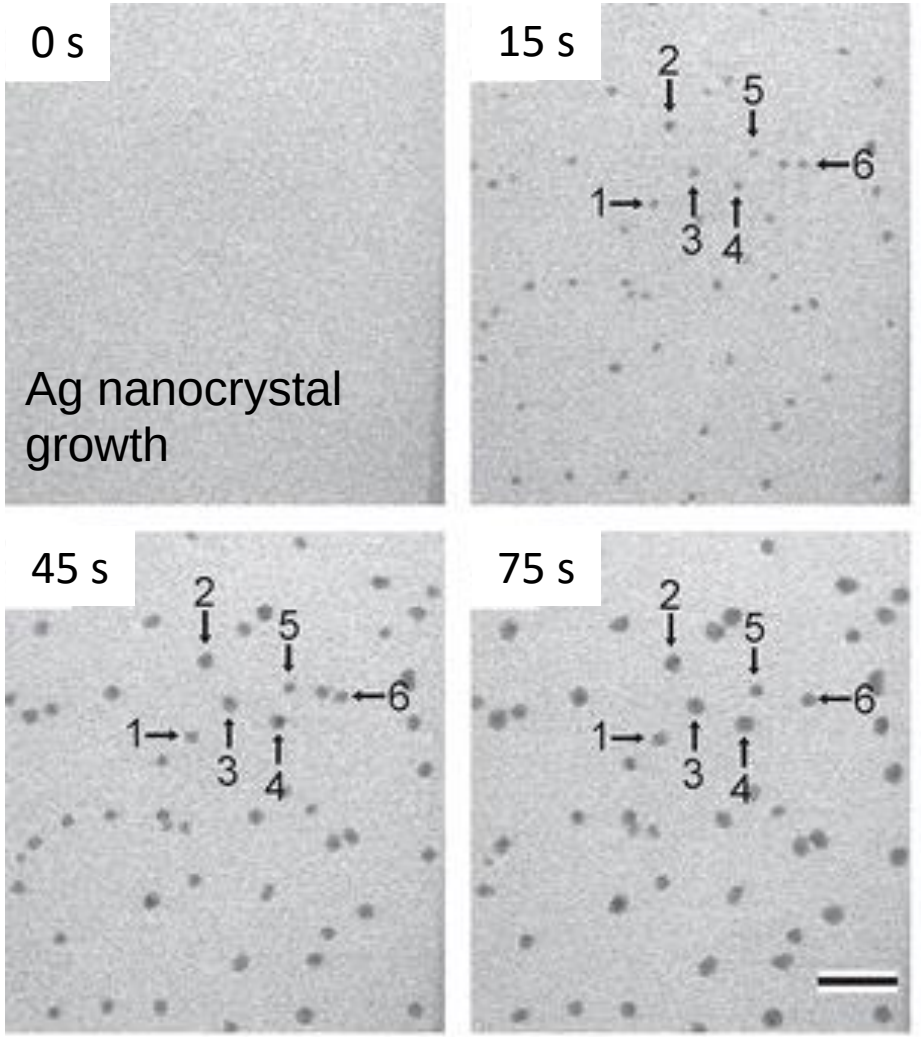
Challenges

- Small field of view
- Beam damage to materials
- Partial coherence
- Experiments in vitro
- Sensitivity to light elements
- 2D projection

Applications of electron microscopy

Time-series/In-situ TEM

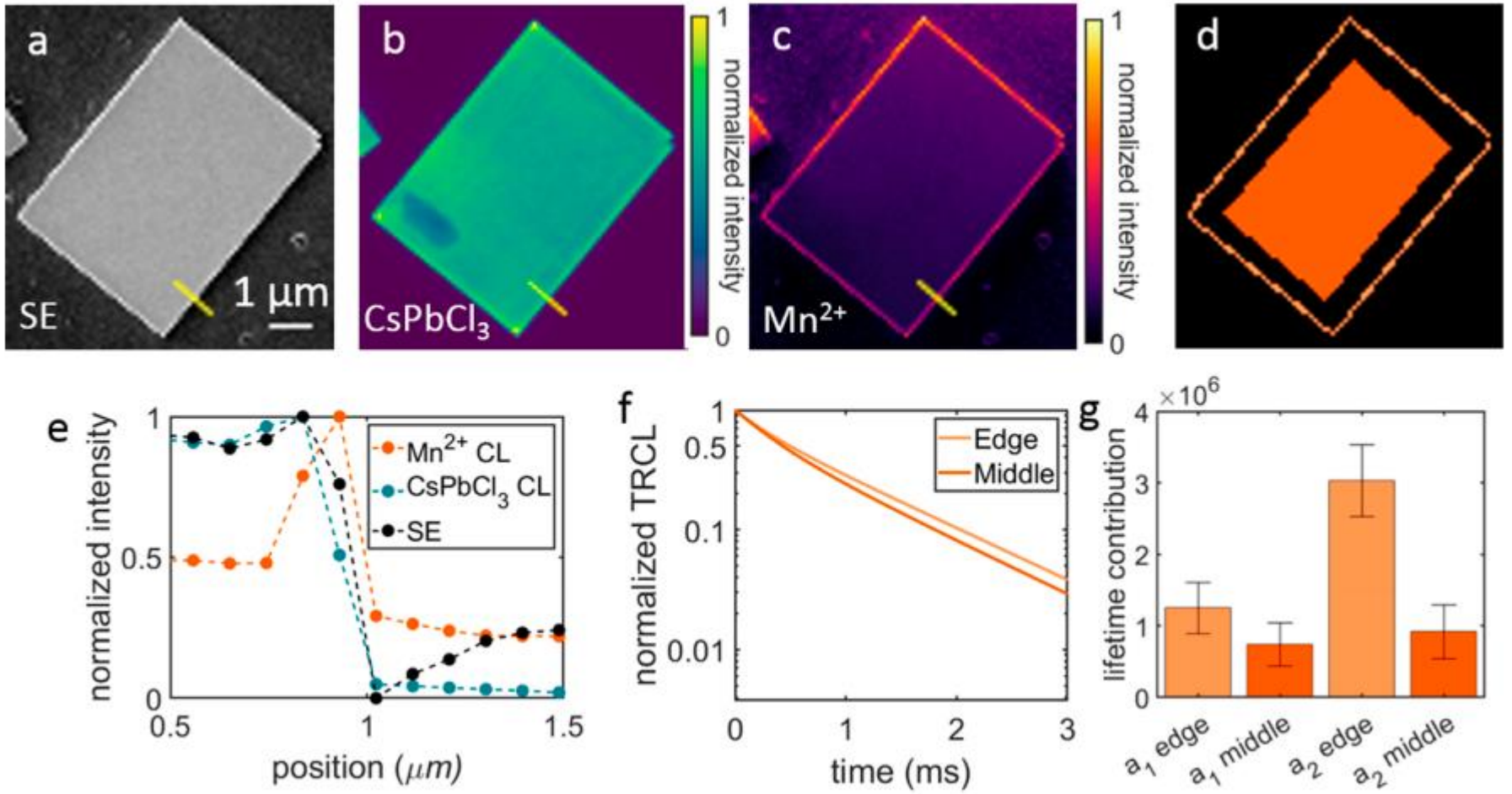




*Zhang et al., Science **5**(12), p.eaax2799, 2019*

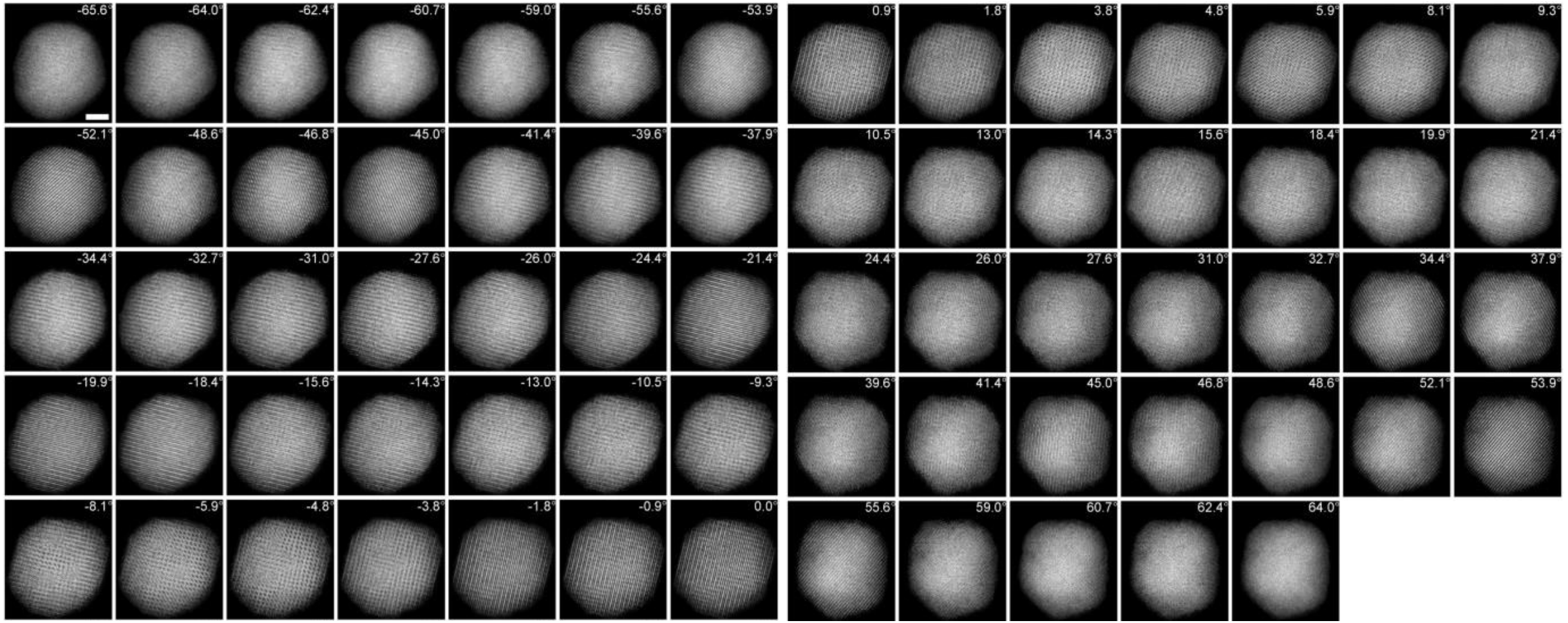
ACS Nano 2012, 6, 10, 8599-8610

Time-series/CL SEM

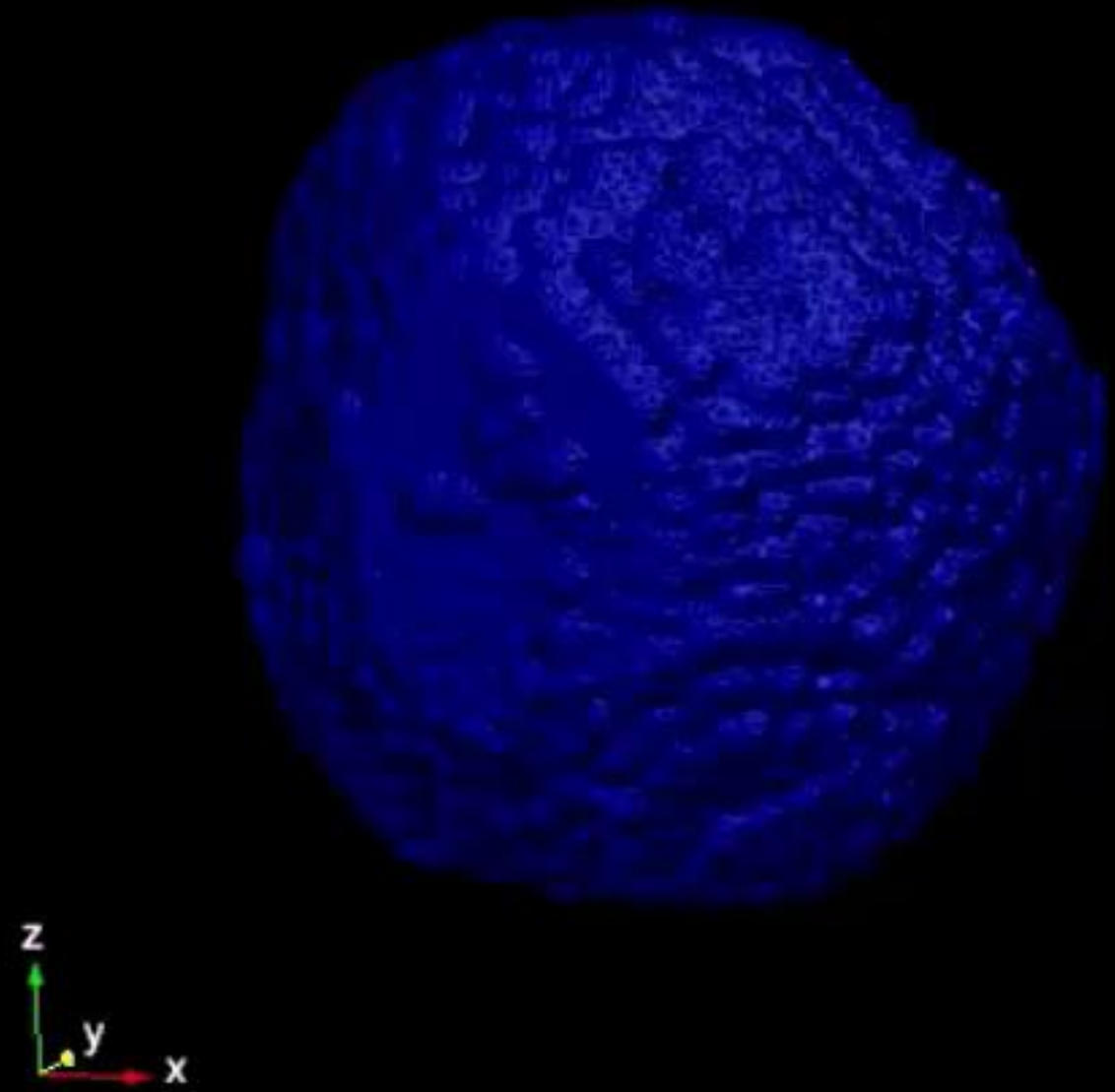


Wai et al, J. Phys. Chem. Lett. ***11****, pp. 2624−2629, 2020*

Atomic electron tomography

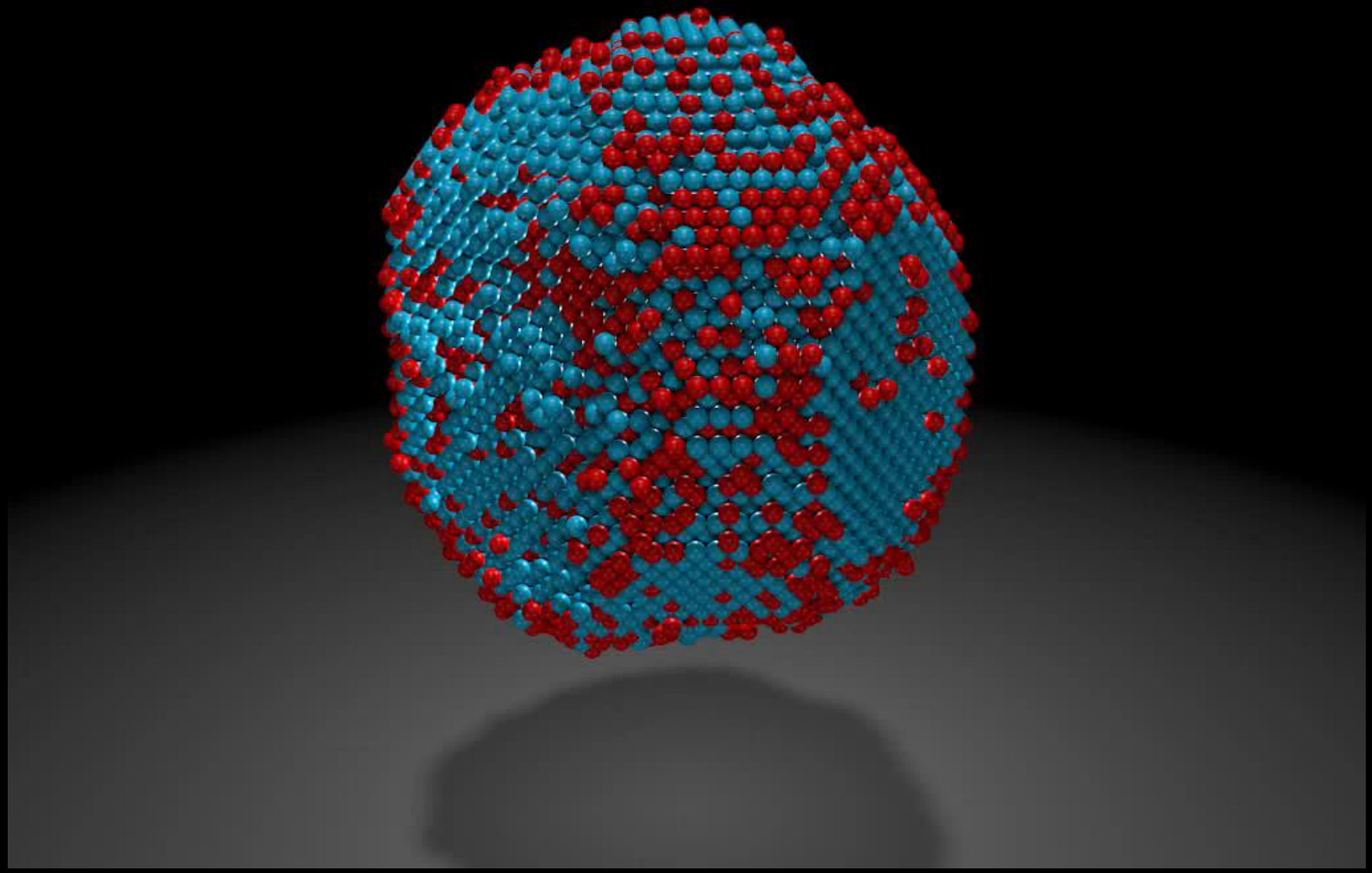


Yang et al. Nature **542**, pp. 75–79, 2017



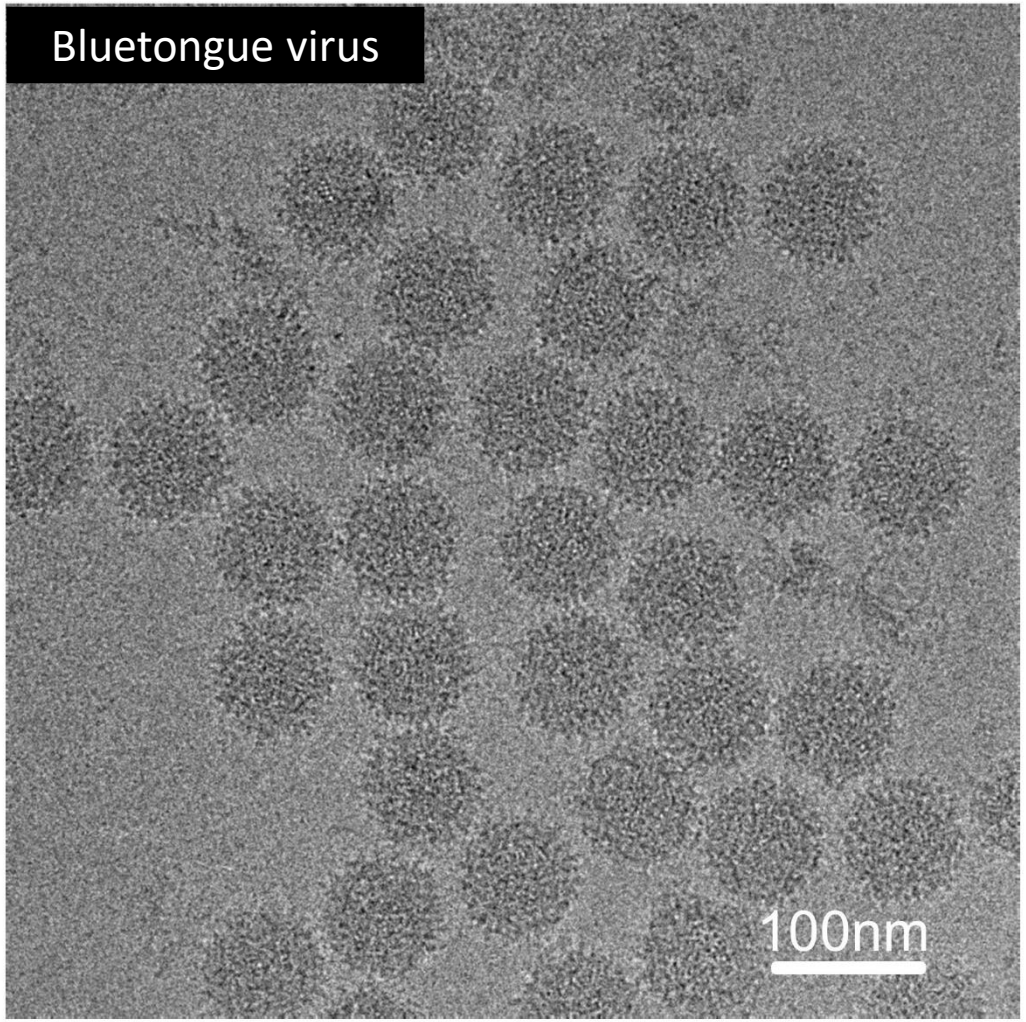
Yang et al. Nature **542**, pp. 75–79, 2017

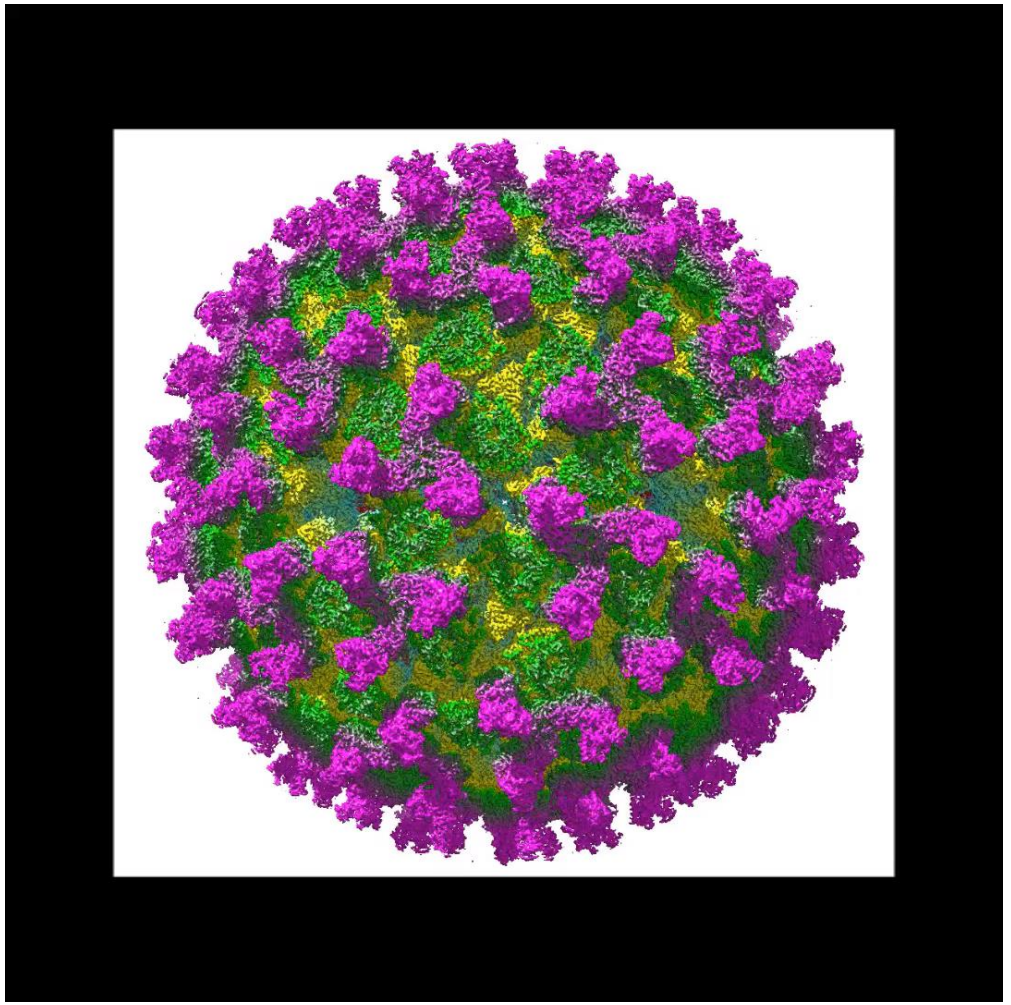
Fe: red
Pt: blue



Yang et al. Nature **542**, pp. 75–79, 2017

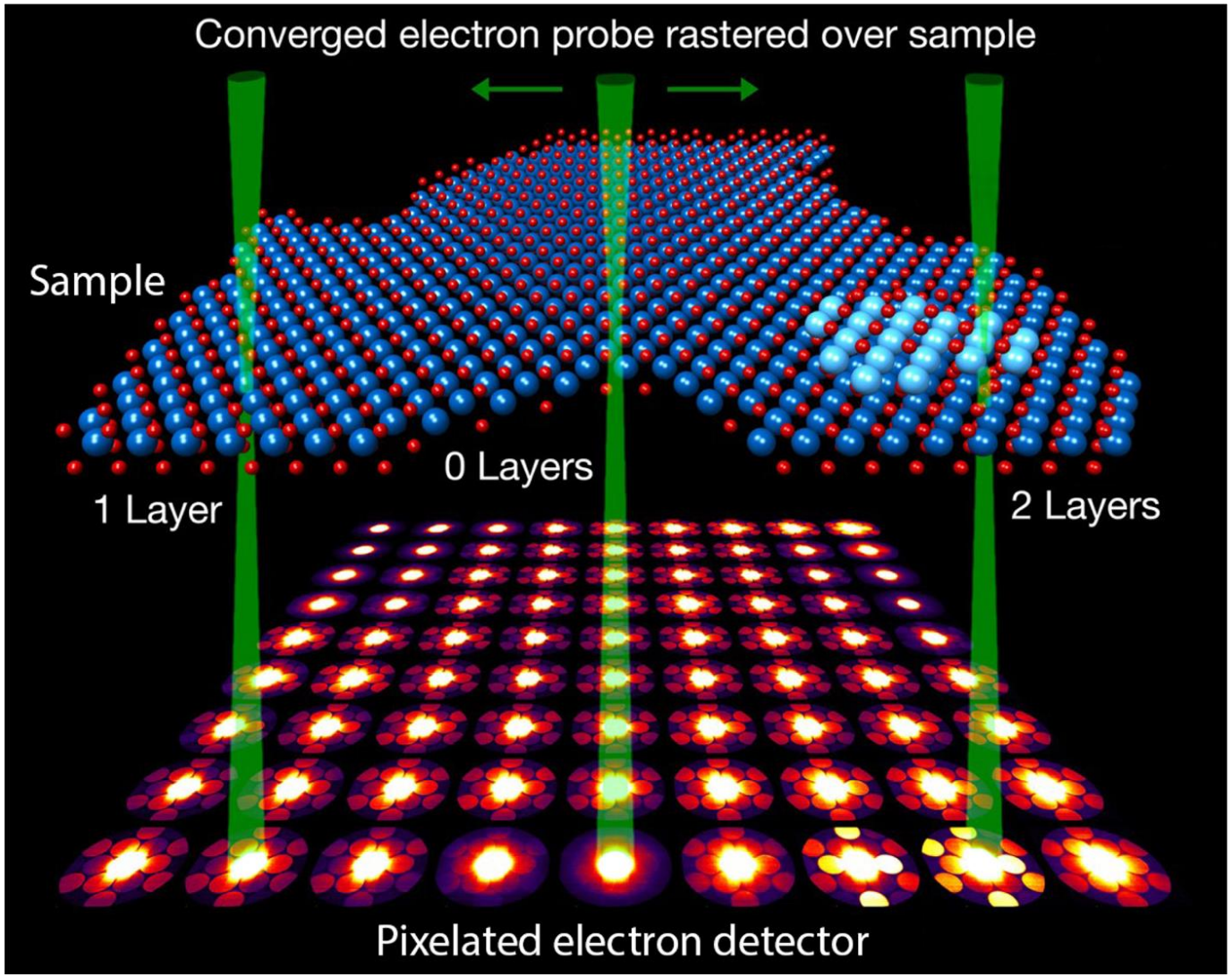
Cryo-EM, single particle reconstruction





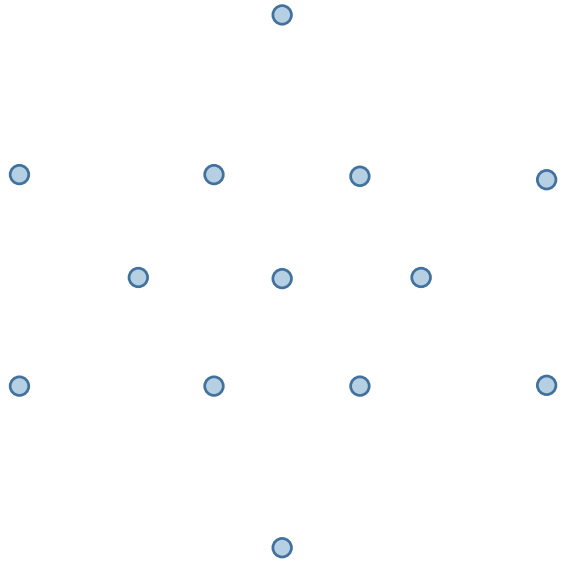
*Zhang et al, Nature Structural & Molecular Biology **23**, pp. 74–80, 2016*

4D-STEM

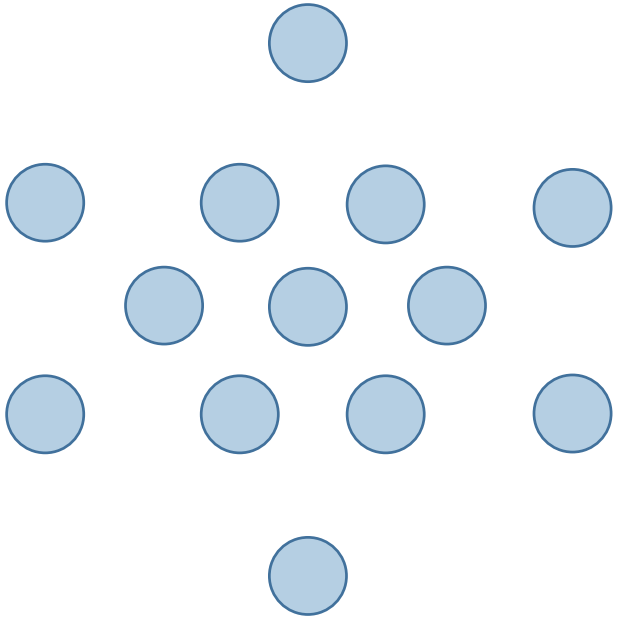


*Ophus, Microscopy and Microanalysis **25**(3), pp. 563-582, 2019.*

Appendix: TEM vs STEM diffraction

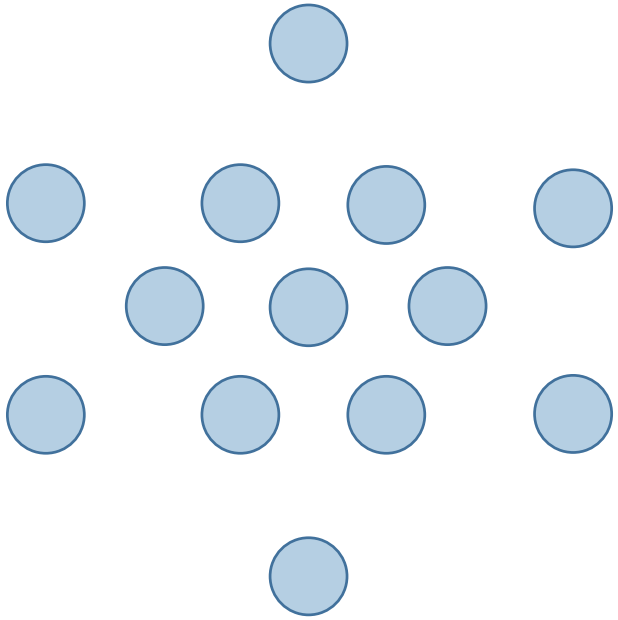


Parallel TEM – diffraction spots

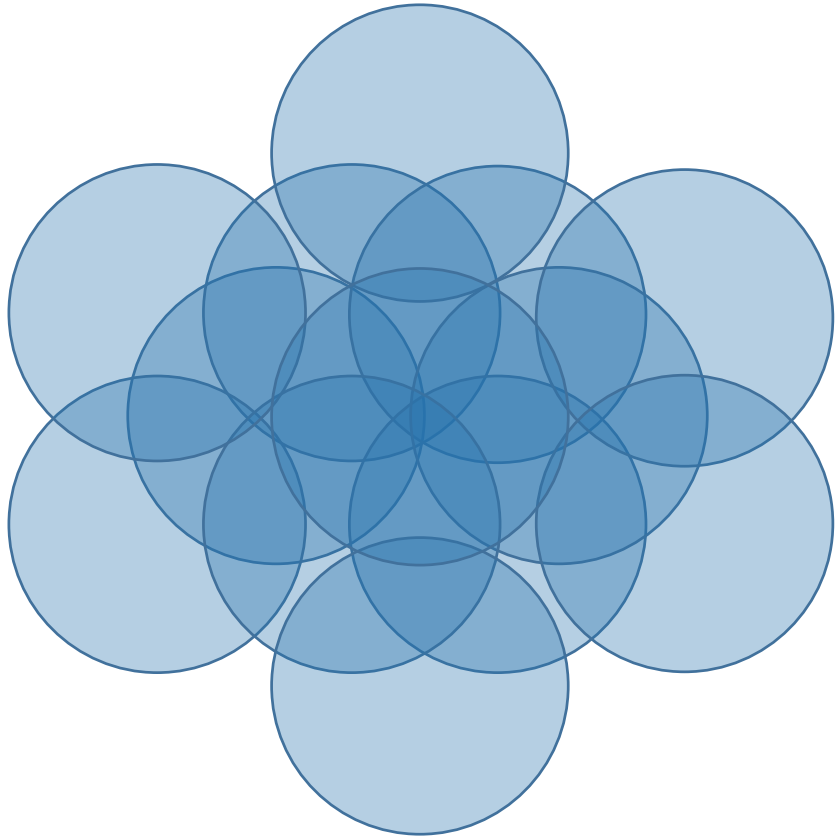


STEM – diffraction disks

Appendix: TEM vs STEM diffraction

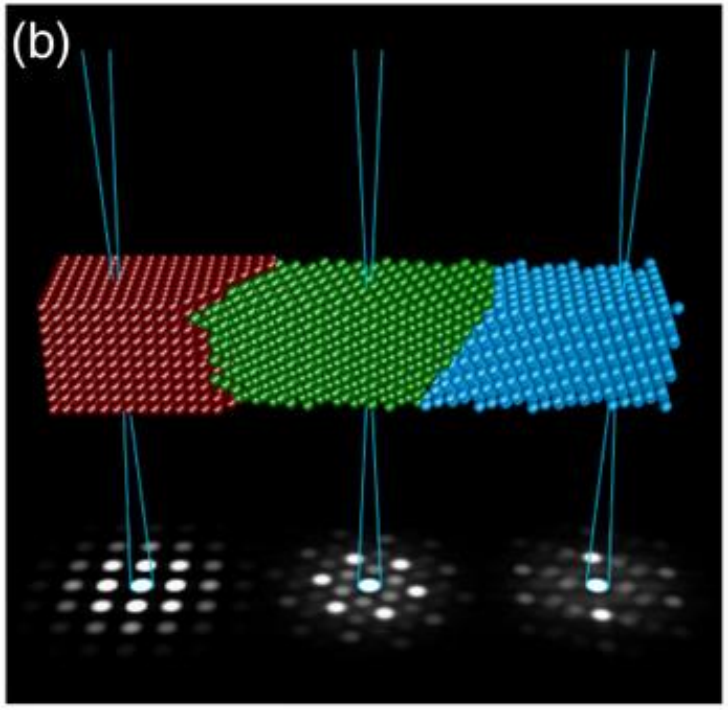


Small convergence: crystal orientation mapping

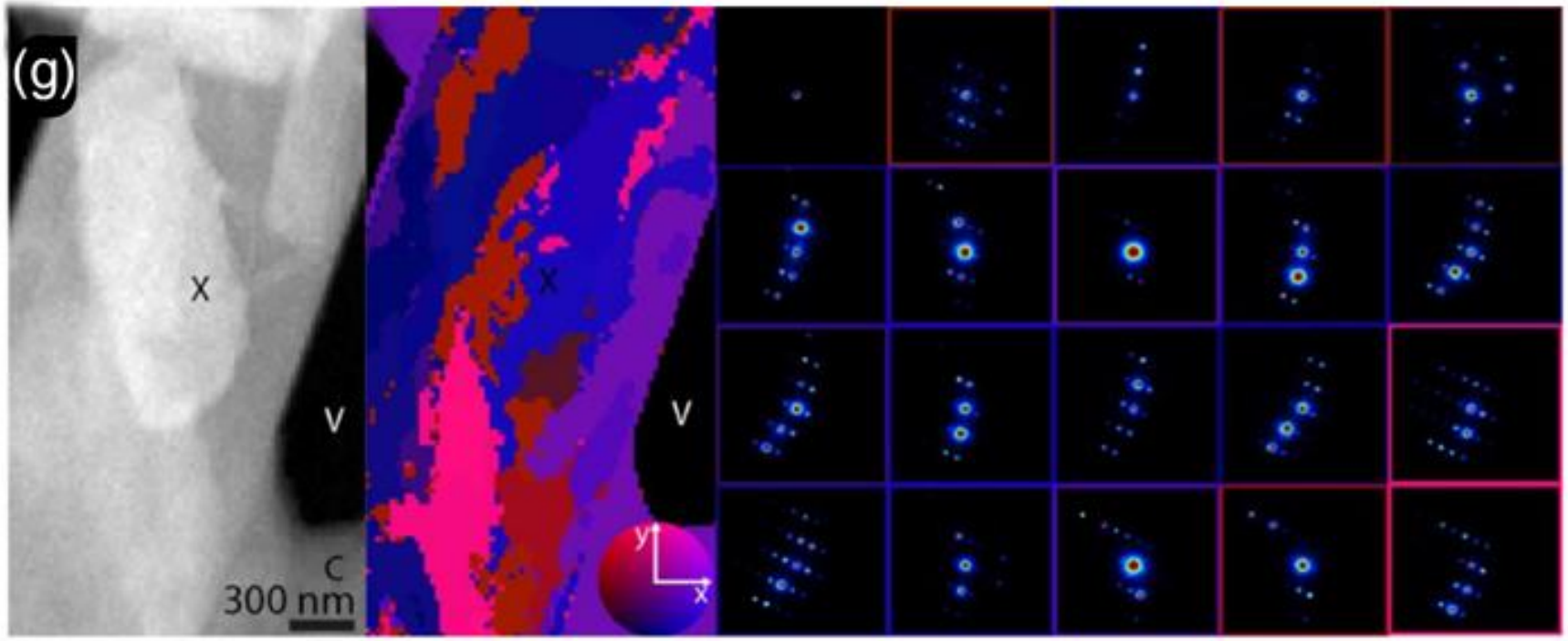


Large convergence – atomic-resolution imaging

4D-STEM: orientation+ strain mapping



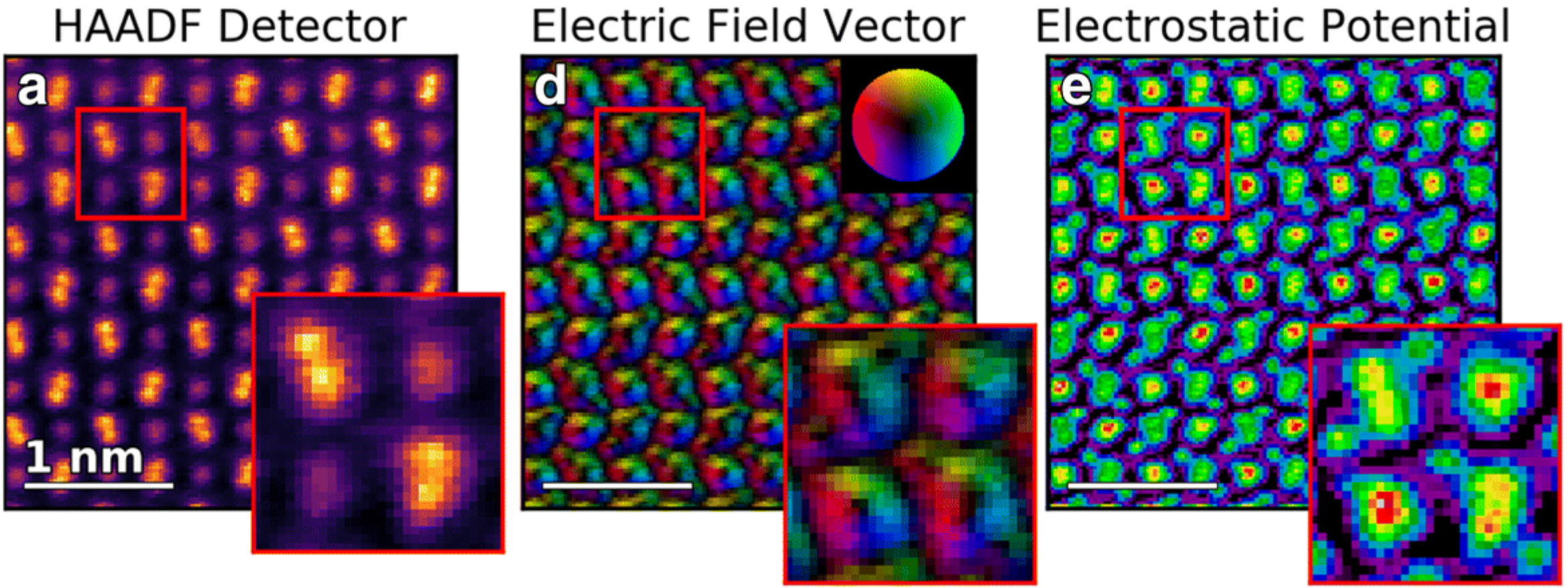
Bragg diffraction



Peptide crystal

Gallagher-Jones et al, Commun. Biol. 2, p. 26, 2019
*Ophus, Microscopy and Microanalysis **25**(3), pp. 563-582, 2019.*

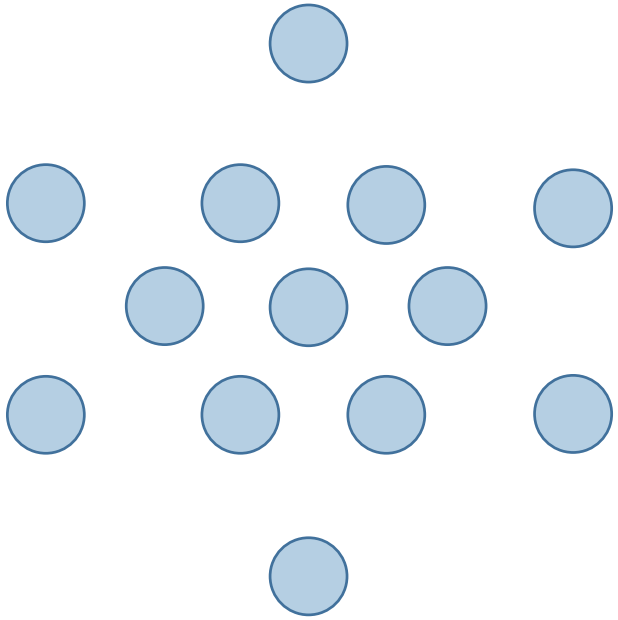
4D-STEM: electric field mapping



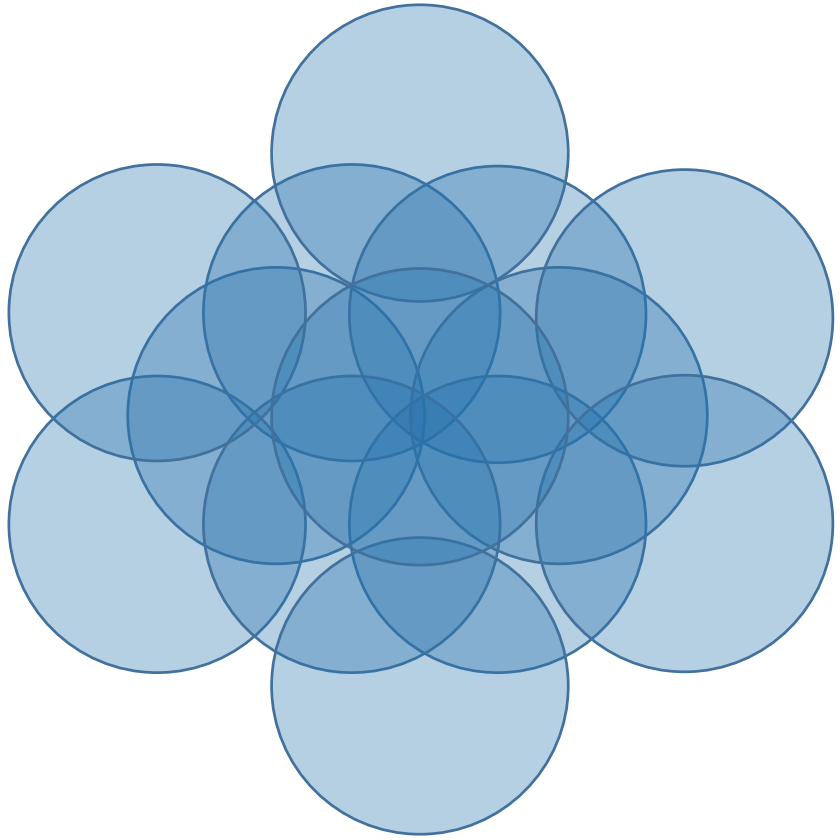
Sample:$DyScO_3$

*Hachtel et al, Advanced Structural and Chemical Imaging **4**(10), 2018*

Interference between overlapping beams

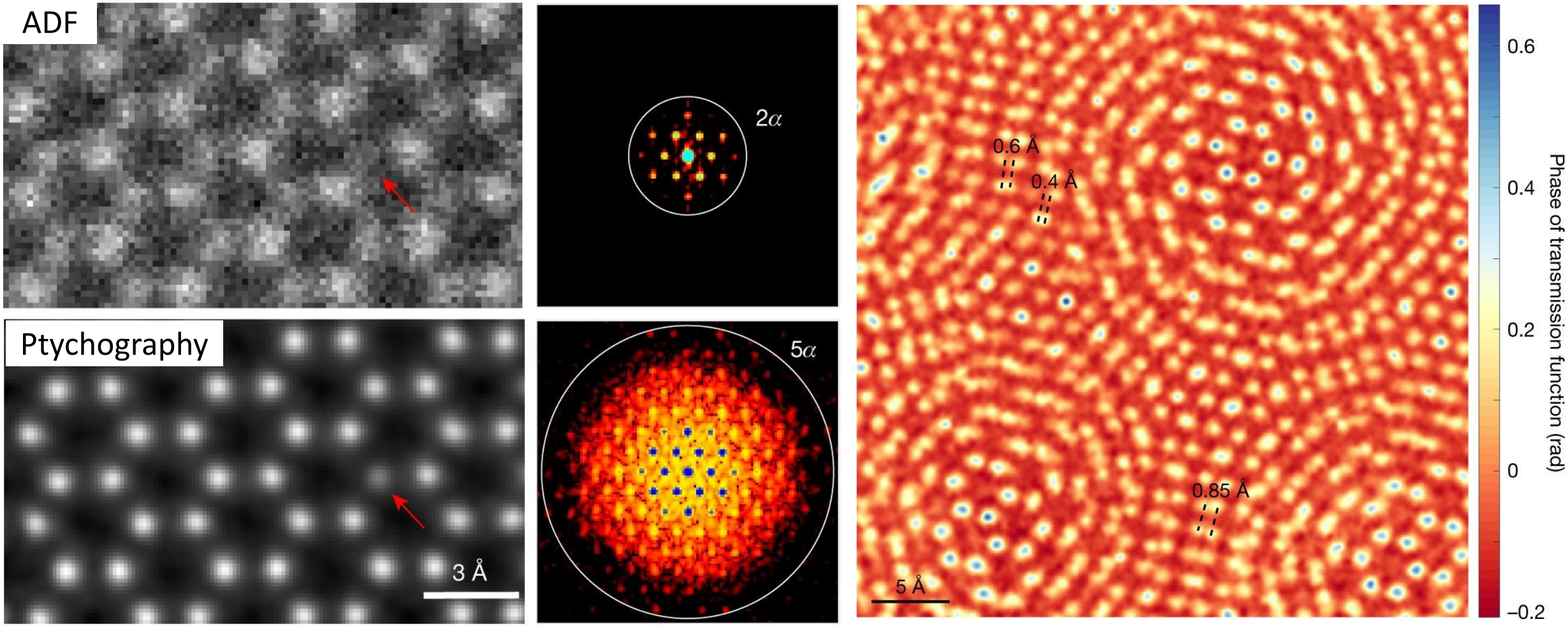


Small convergence: crystal orientation mapping



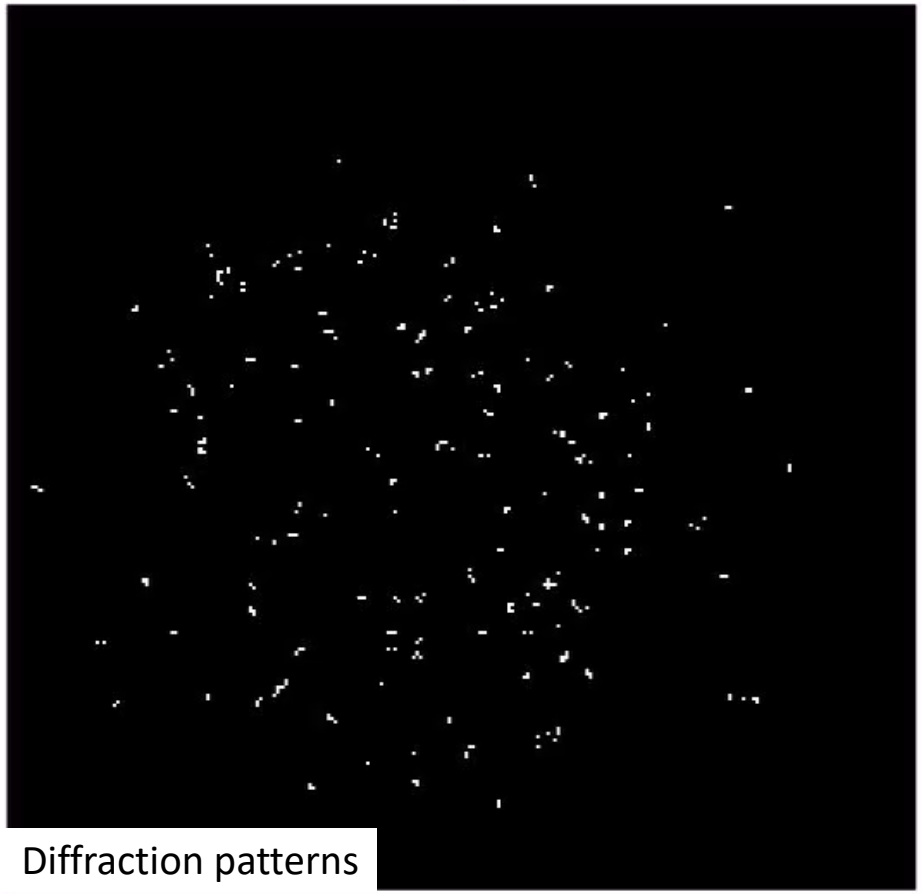
Large convergence – atomic-resolution imaging

4D-STEM: electron ptychography

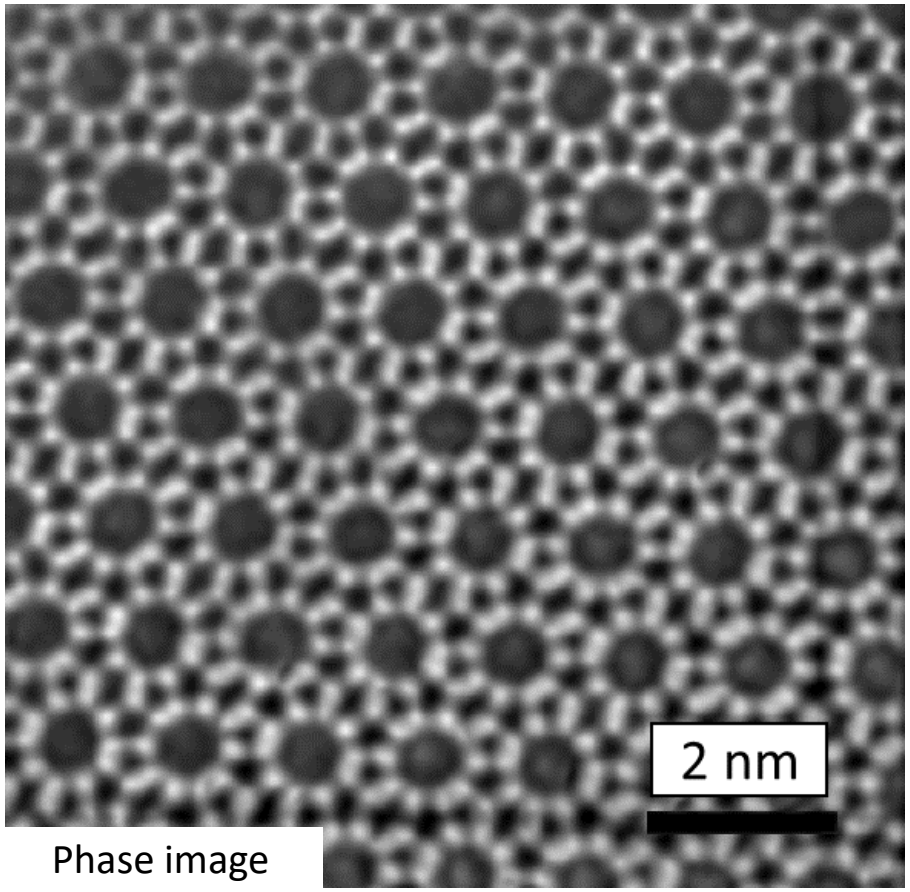


*Jiang et al, Nature **559**, pp. 343–349, 2018*

4D-STEM: Electron ptychography



Diffraction patterns

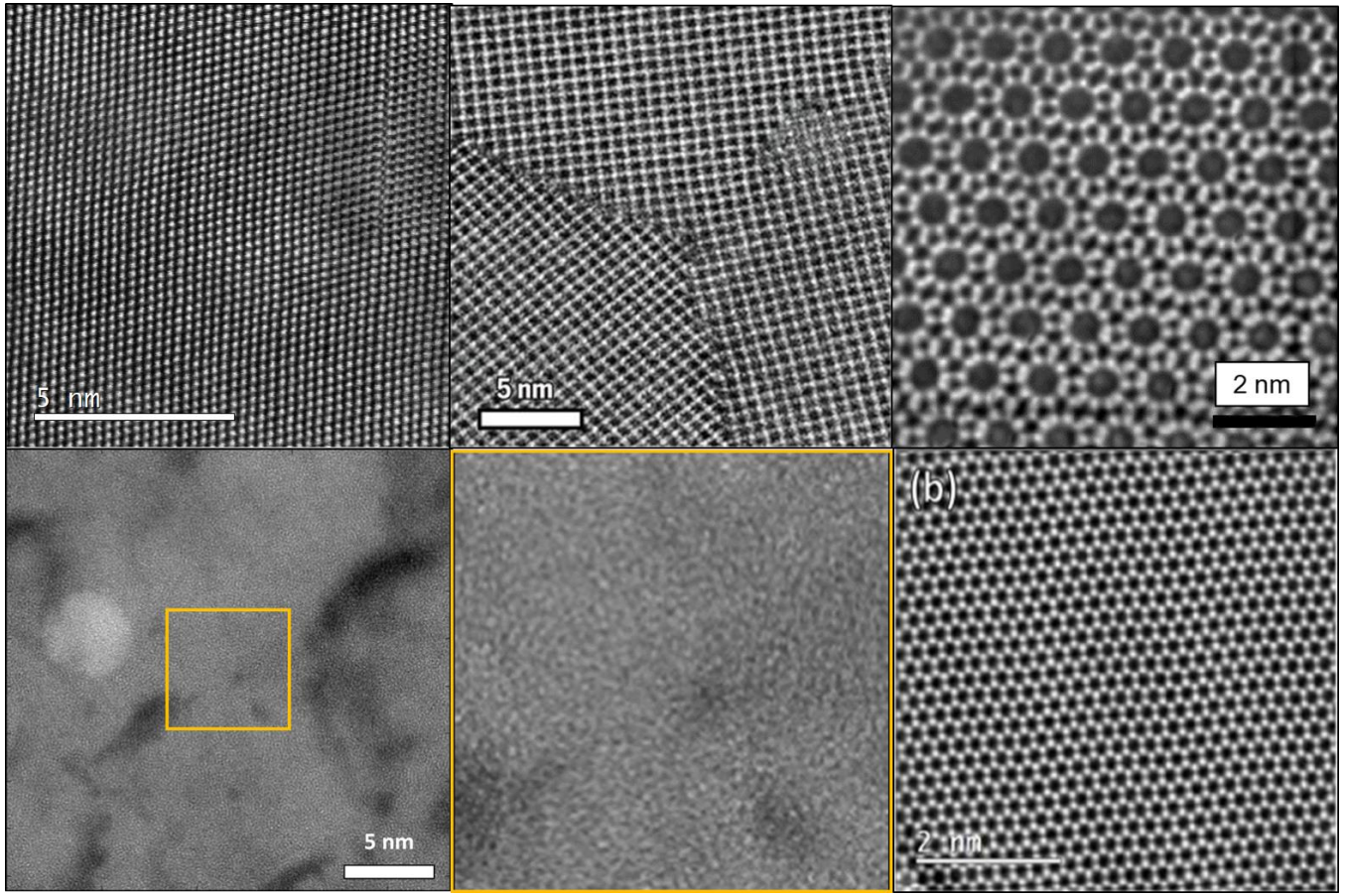


Phase image

O'Leary et al, Appl. Phys. Lett. ***116****, p. 124101, 2020*

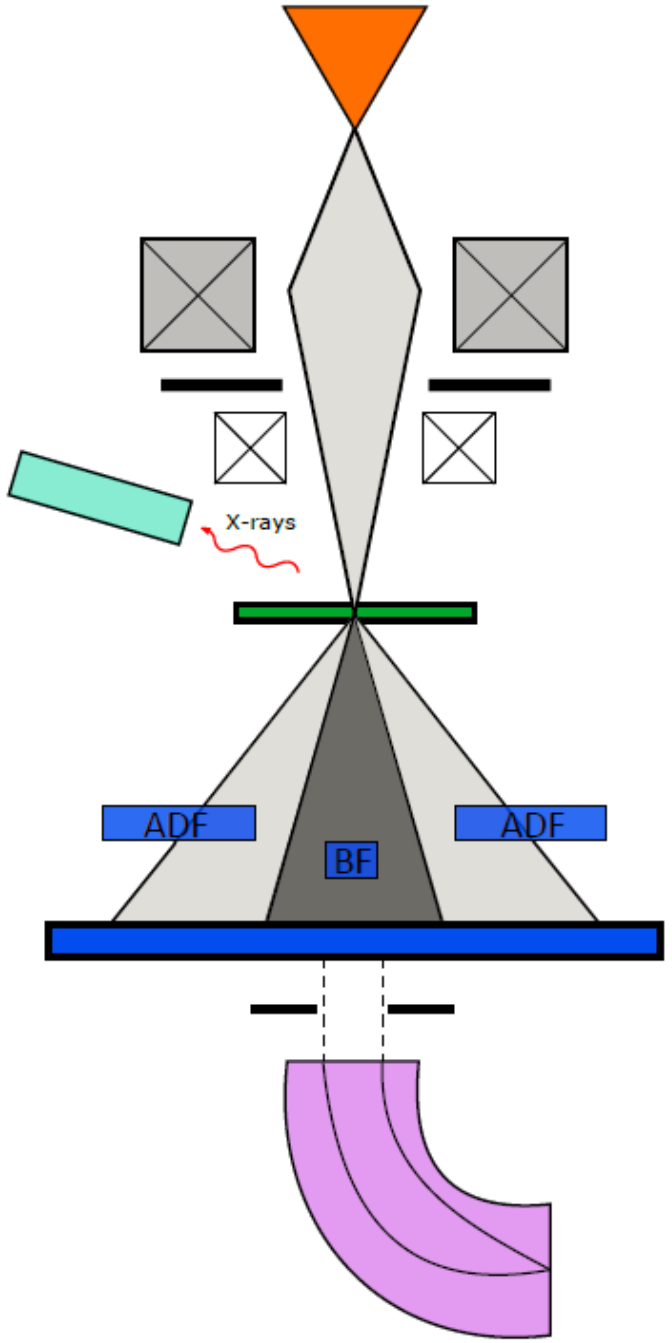
Outline

- **Why use electron microscopy?**
- Modes and applications.
- Challenges and latest developments.



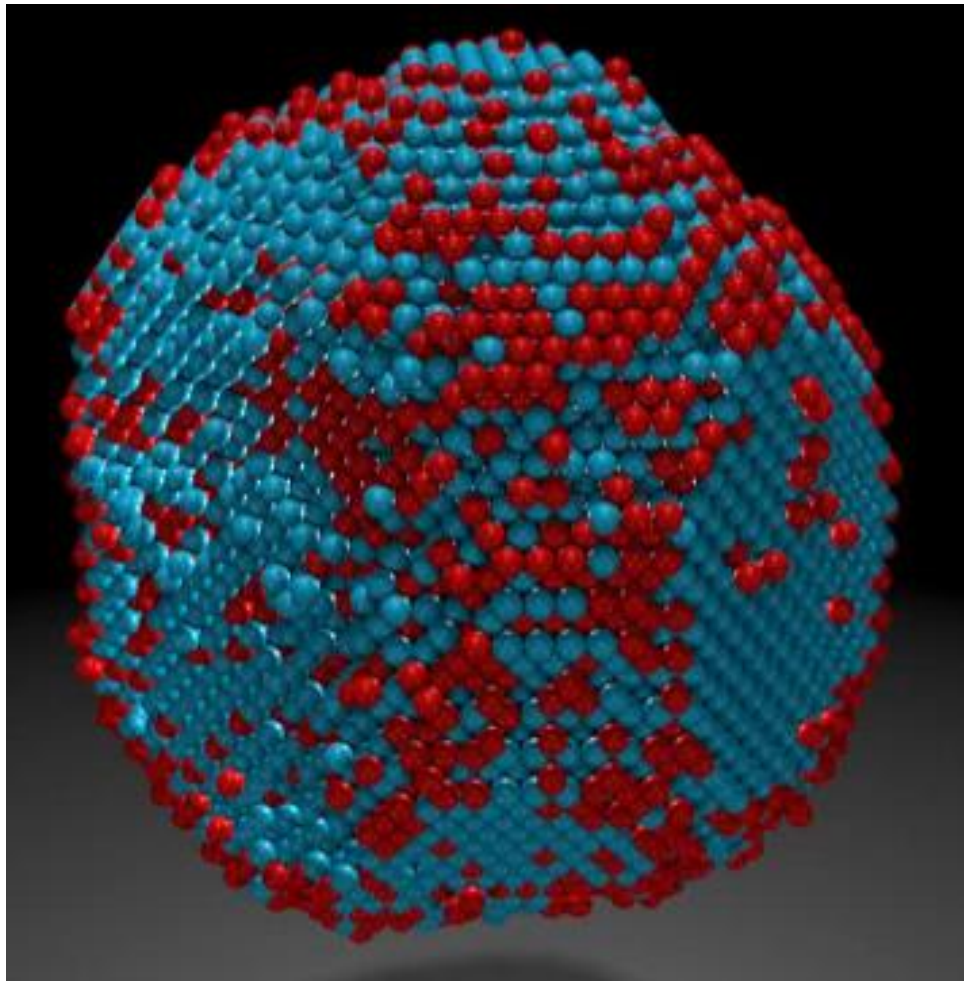
Outline

- Why use electron microscopy?
- **Modes and applications.**
- Challenges and latest developments.



Outline

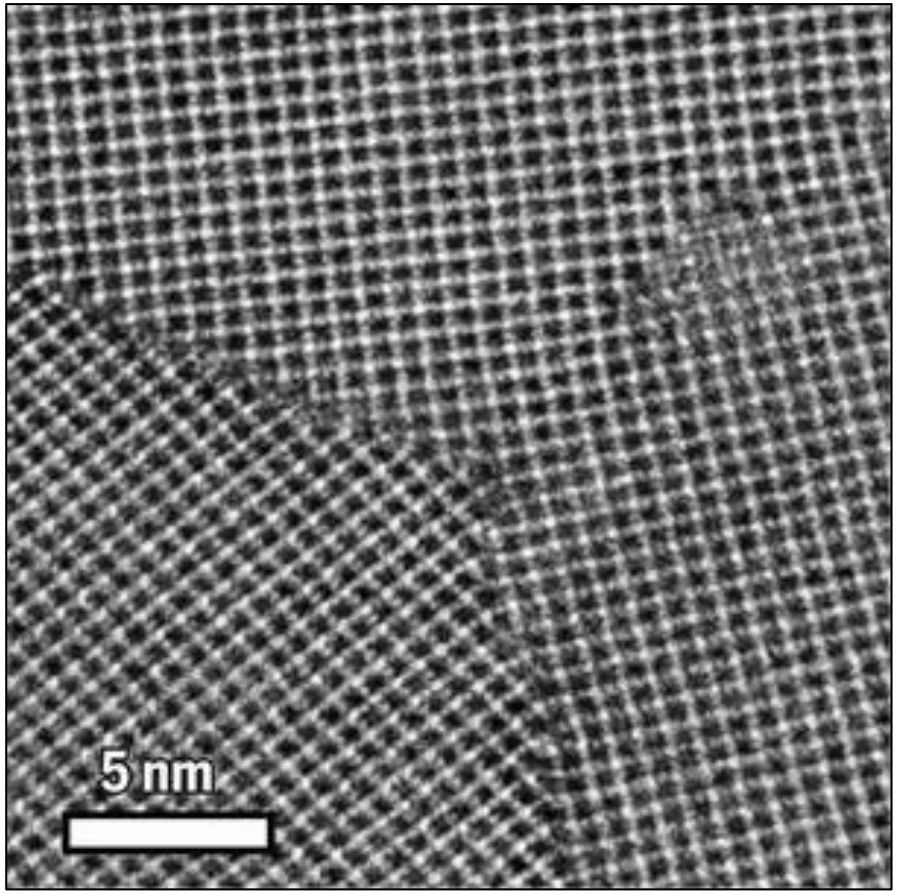
- Why use electron microscopy?
- Modes and applications.
- **Challenges and latest developments.**



Room for improvement

- **Atomic-resolution for large FOV.**
- 4D-STEM analysis (ptychography).
- Low voltage applications.
- Small, atomic-resolution microscopes.

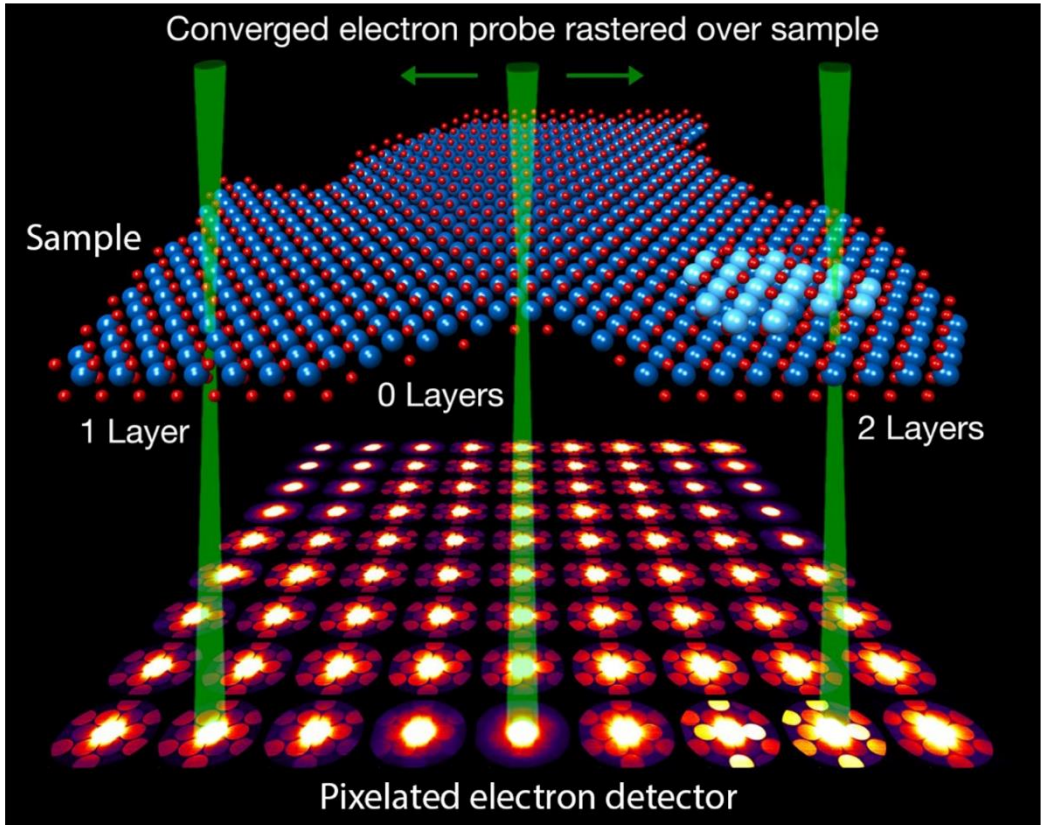
Lead-halide perovskite



Crystal structure and imperfections

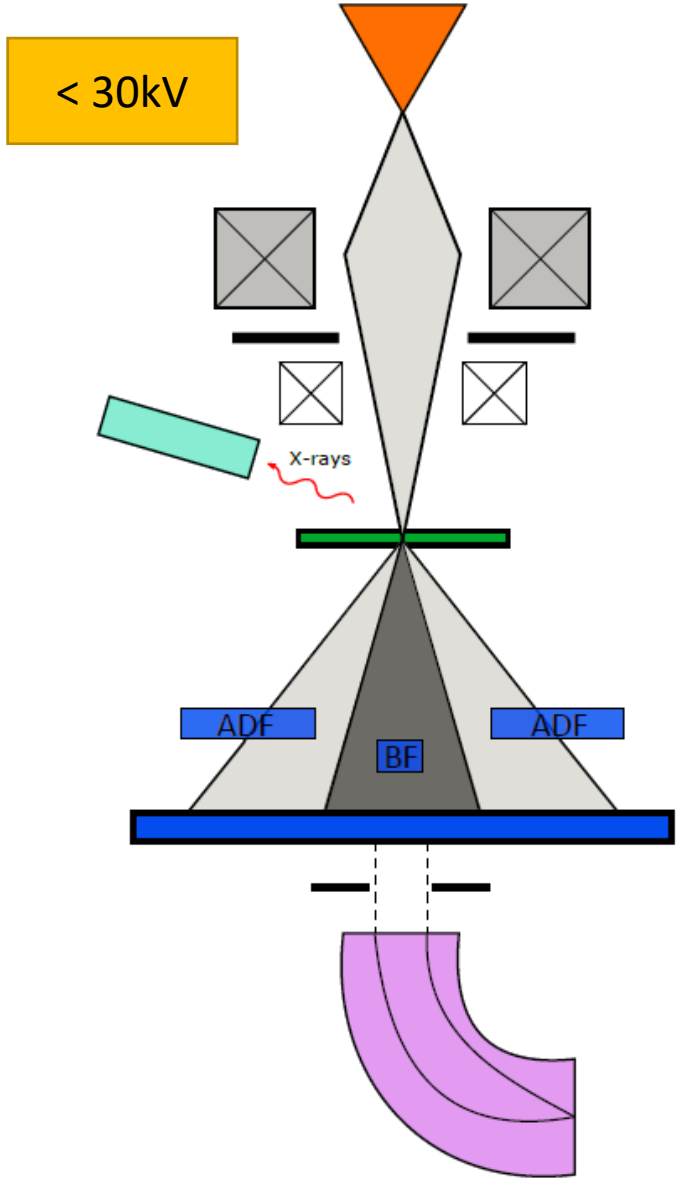
Room for improvement

- Atomic-resolution for large FOV.
- **4D-STEM analysis (ptychography).**
- Low voltage applications.
- Small, atomic-resolution microscopes.



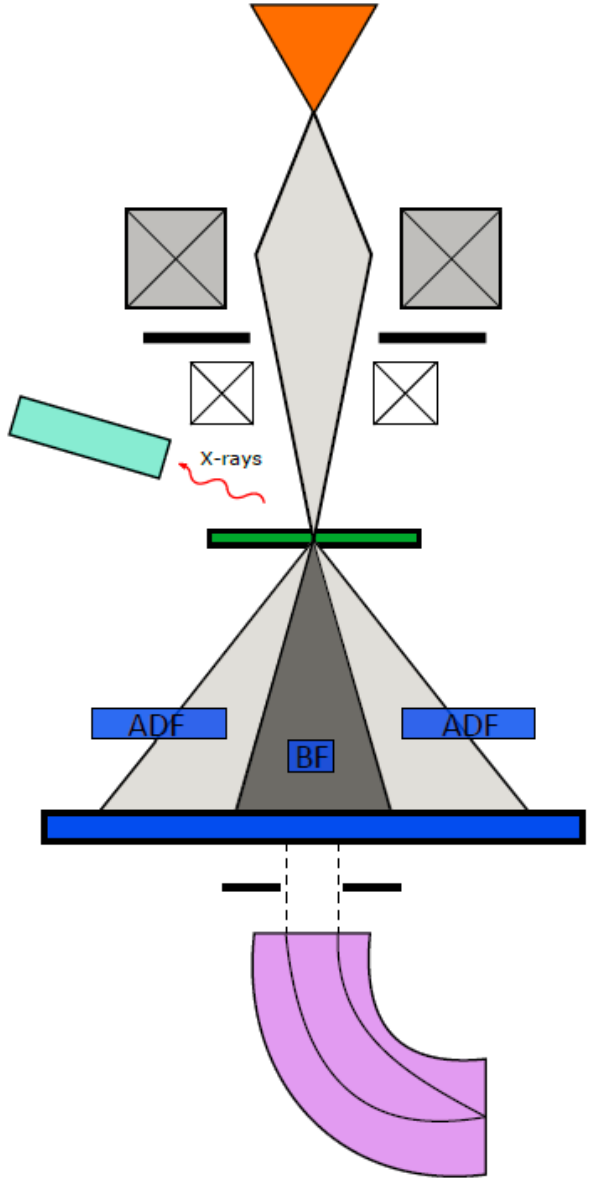
Room for improvement

- Atomic-resolution for large FOV.
- 4D-STEM analysis (ptychography).
- **Low voltage applications.**
- Small, atomic-resolution microscopes.



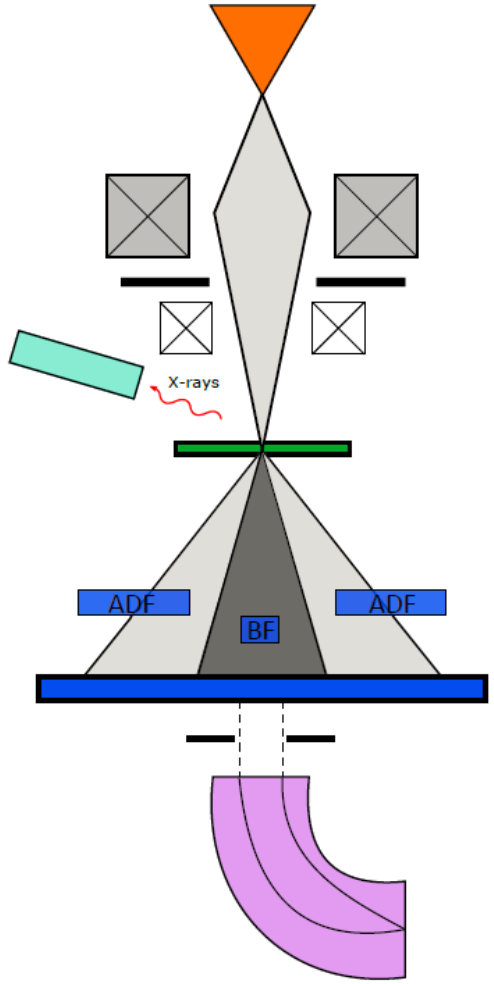
Room for improvement

- Atomic-resolution for large FOV.
- 4D-STEM analysis (ptychography).
- Low voltage applications.
- **Small, atomic-resolution microscopes.**



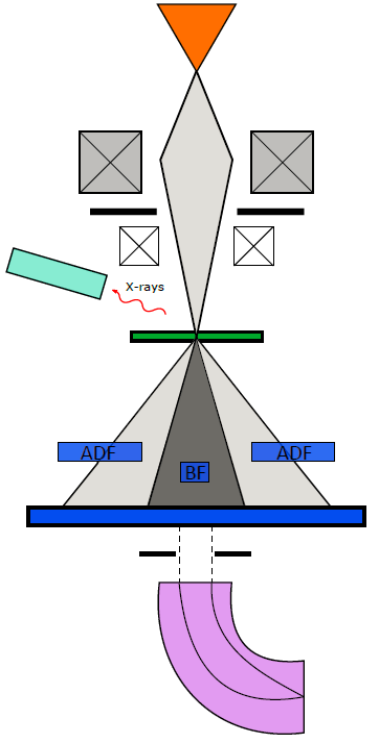
Room for improvement

- Atomic-resolution for large FOV.
- 4D-STEM analysis (ptychography).
- Low voltage applications.
- **Small, atomic-resolution microscopes.**



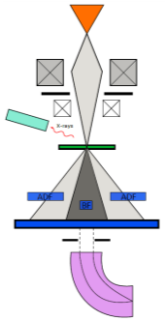
Room for improvement

- Atomic-resolution for large FOV.
- 4D-STEM analysis (ptychography).
- Low voltage applications.
- **Small, atomic-resolution microscopes.**



Room for improvement

- Atomic-resolution for large FOV.
- 4D-STEM analysis (ptychography).
- Low voltage applications.
- **Small, atomic-resolution microscopes.**



Thank You