Phase Contrast Microscopy with Soft and Hard X-rays Using a Segmented Detector

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BNL Instrumentation Seminar, 28 March 2007

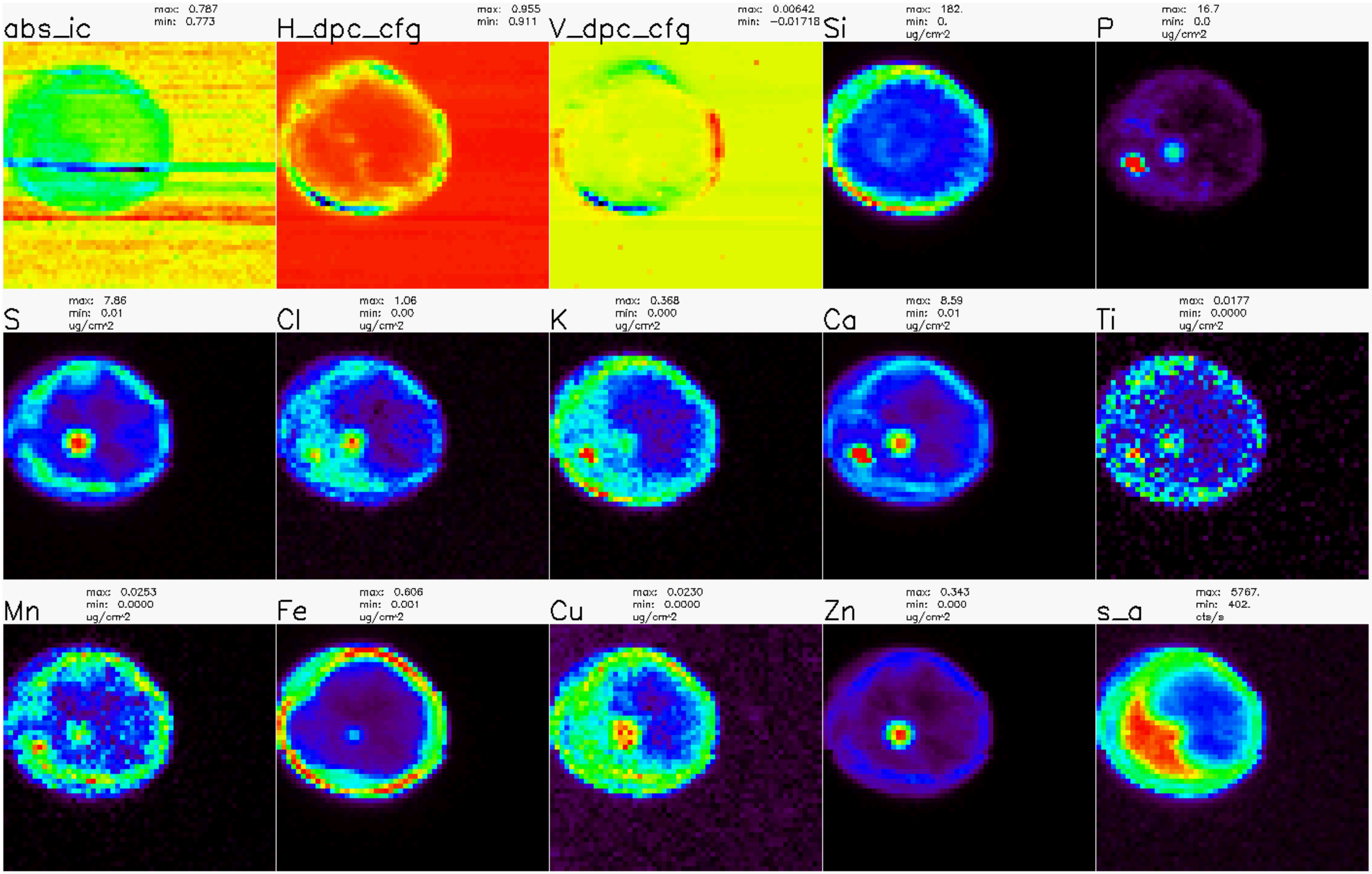
Stony Brook X-ray Microscopy

nsls

Advanced Photon Source
Argonne National Laboratory

CEMS

Fluorescence Trace Element Map of Phytoplankton Cell



10 µm

Sample: Stephen Baines, Stony Brook Marine Sciences

Outline

- Introduction
  - X-ray Microscopy 101
  - Phase Contrast 101
- A Segmented Detector for Hard X-ray Microprobes
  - Segmented Silicon Chip
  - Charge Integrating Electronics
- Differential Phase Contrast (DPC)
  - Comparison with Amplitude Contrast and DPC Examples
  - Integration of the DPC Signal
- Quantitative Amplitude and Phase Reconstruction
  - Reconstruction Scheme
  - Simulations and Experiments with Soft and Hard X-rays
- Summary and Outlook

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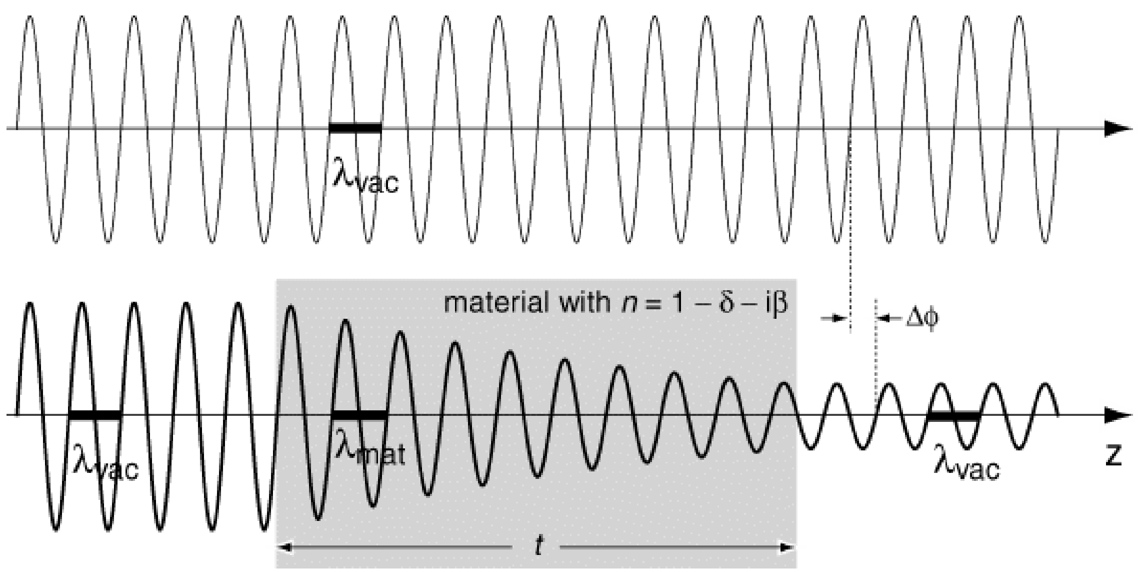
X-ray Interactions: Wave Propagation

- Complex **index of refraction:** $n = 1 - \delta - i\beta$
- δ, β: small positive numbers (10^{-4}, ..., 10^{-9}, tabulated values)
- Wave propagation through material with refractive index *n*:

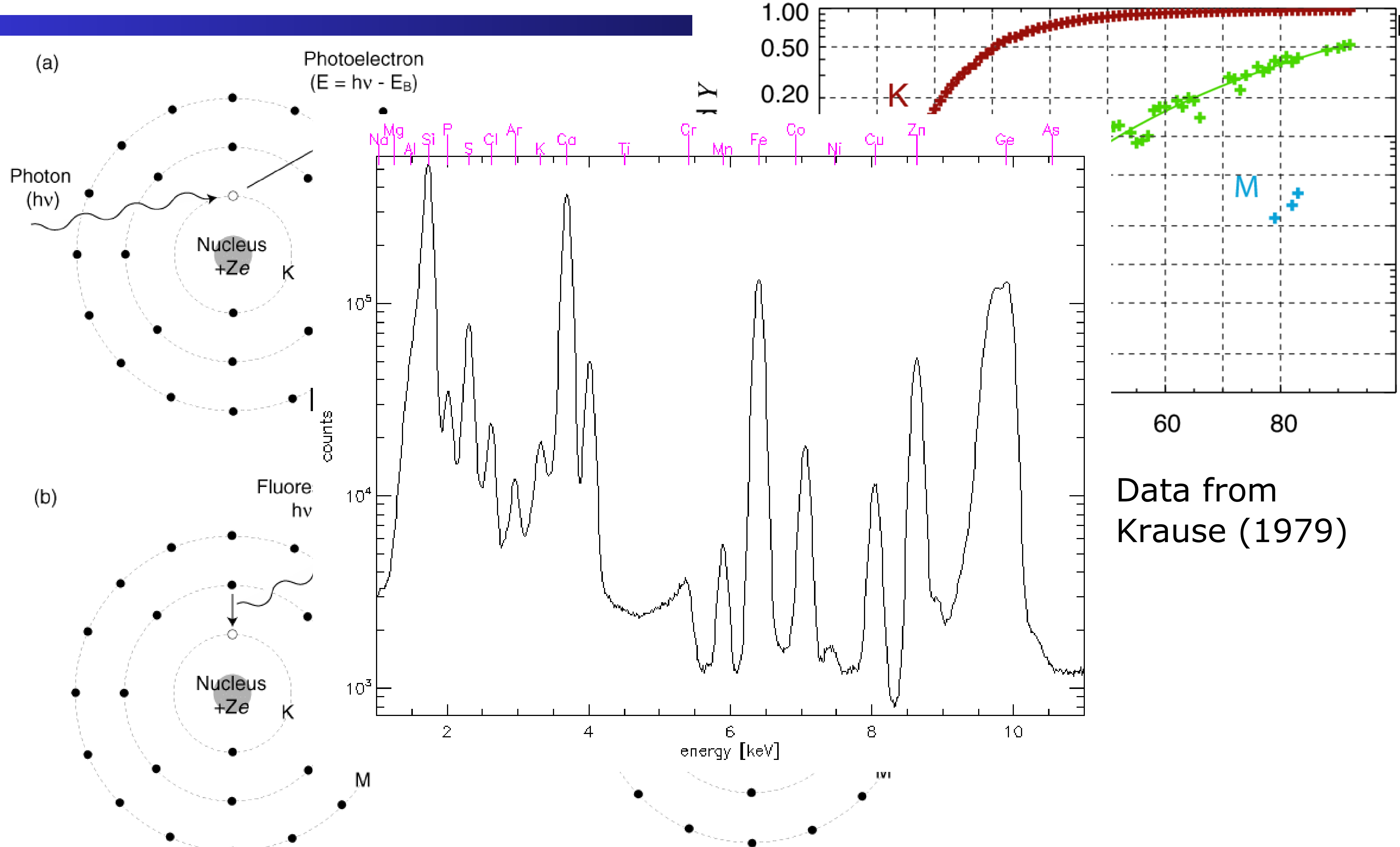
$$\psi(z) = \psi_0 \exp(-inkz) = \psi_0 \exp(-ikz)\,\exp(+i\delta kz)\cdot\exp(-\beta kz)$$

Vacuum propagation — Phase Advance — Absorption

Complex specimen function



X-ray Interactions: Fluorescence



Data from Krause (1979)

Fluorescence emission

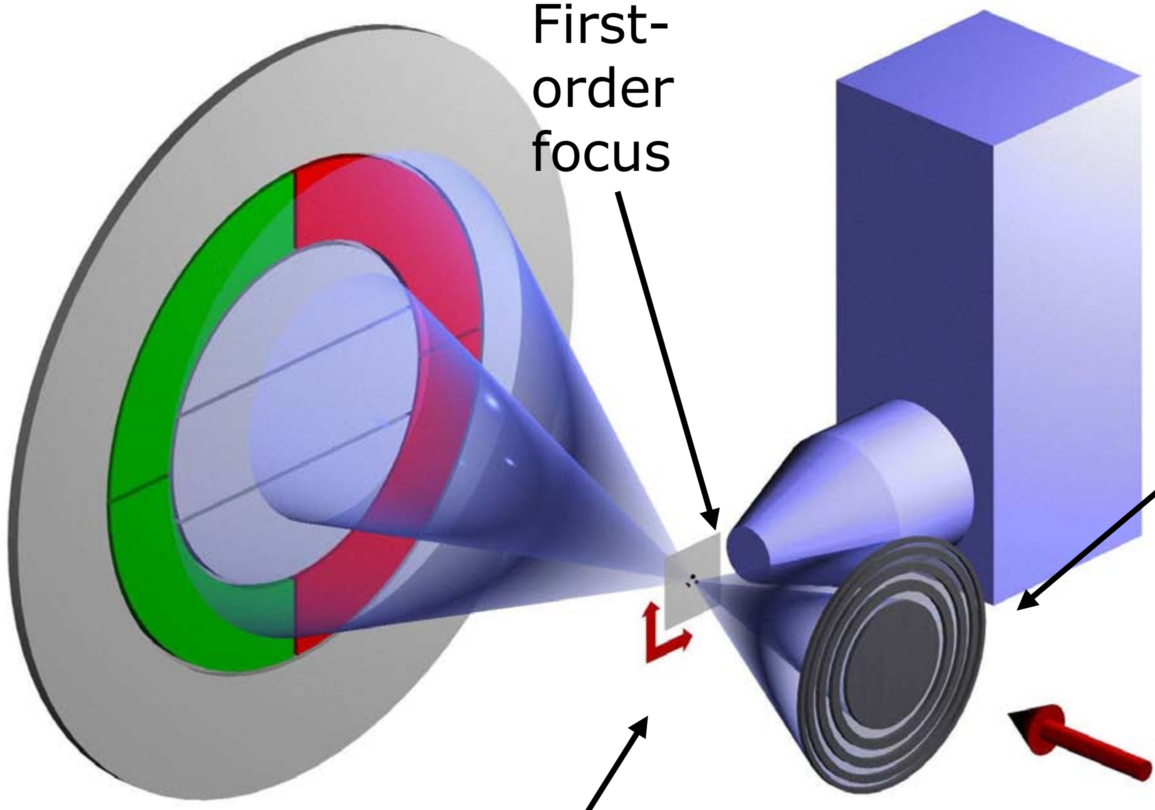
Auger emission

Synchrotrons

Advanced Photon Source (APS), Argonne Nat'l Lab, Illinois

National Synchroton Light Source (NSLS), Brookhaven Nat'l Lab, New York

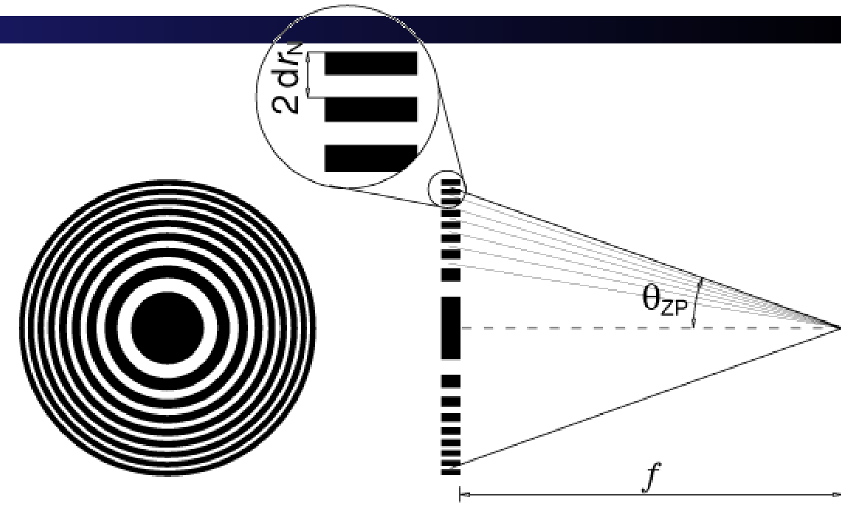
Scanning Transmission X-ray Microscope (STXM) and Fluorescence Microprobe

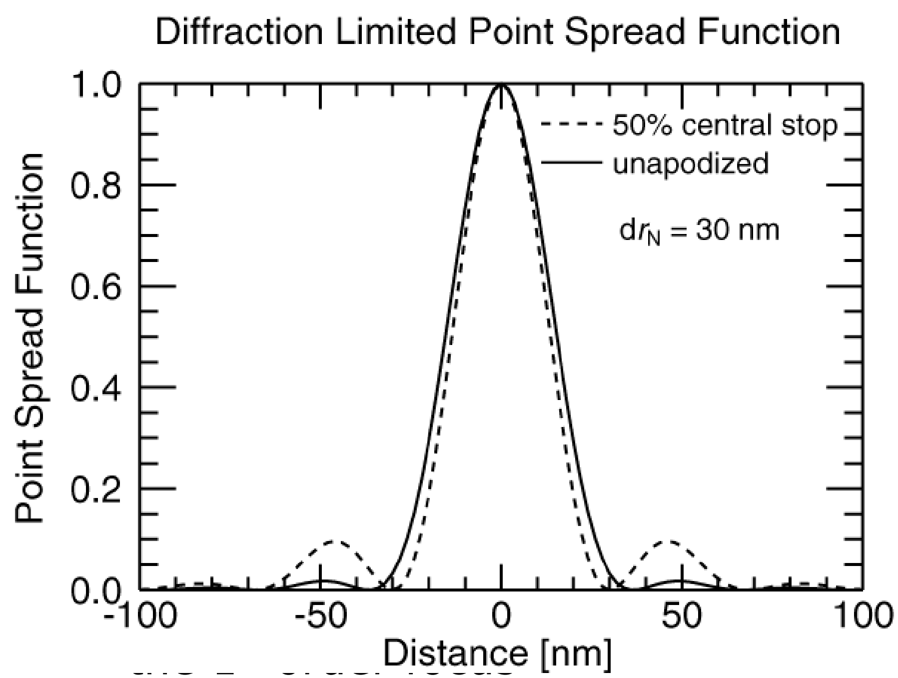


Spatial resolution:
NSLS X1A: 40 nm (sub-keV)
APS 2-ID-B: 55 nm (1-4 keV)
APS 2-ID-E: 250 nm (7-17 keV)

Fresnel Zone Plates

- Circular diffraction gratings with radially decreasing line width
- **Spatial resolution:** 1.22 x outermost zone width
- Usually produced by electron-beam lithography / etching / plating



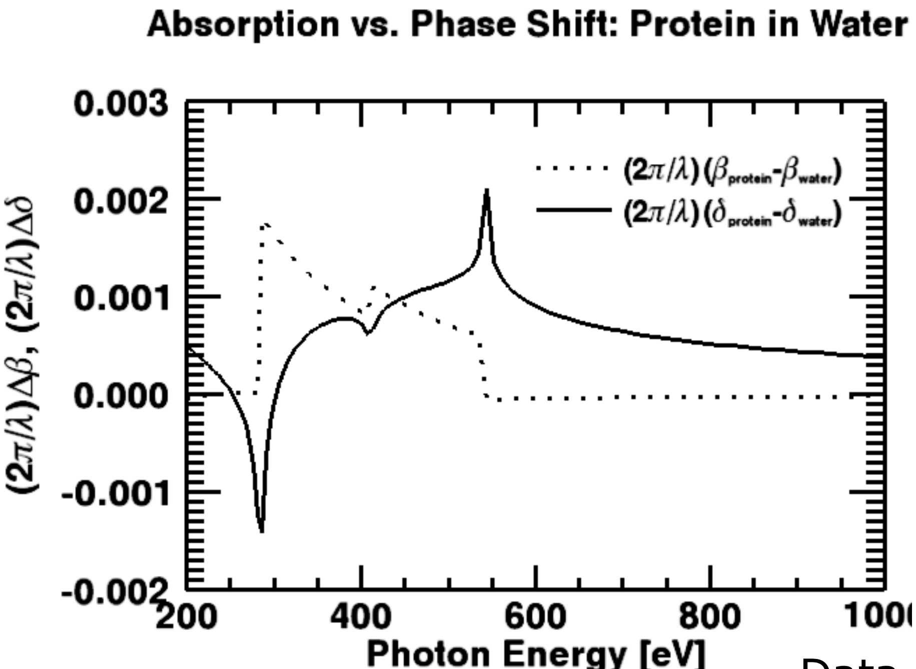


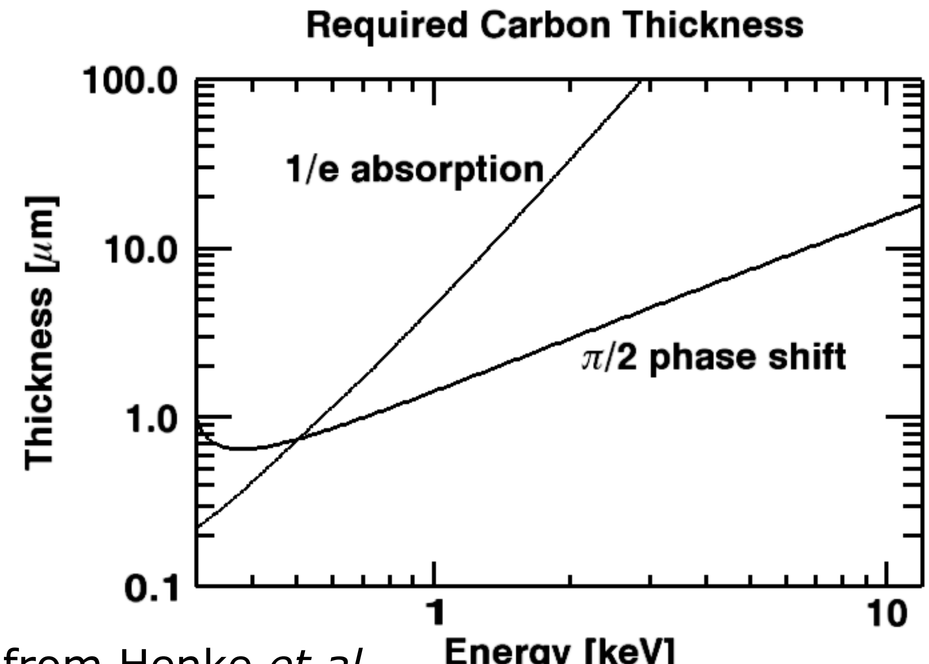
Energy	500 eV	4 keV	10 keV
Wavelength	2.5	0.31	0.12
Diameter	160 um	160 um	320 um
Out. zone wid.	30 nm	50 nm	100 nm
Focal length	1.9 mm	26 mm	270 mm
Thickness	200 nm	450 nm	1600 nm
Material	Nickel	Gold	Gold
Efficiency	12%	15%	30%

Phase Contrast Motivation

- Lower energies: Imaging at the low energy side of an absorption edge can **lower the radiation dose**

- At higher energies: **Phase contrast dominates** $\delta / \beta \propto E^2$
  - Combine with fluorescence
  - PC to image ultrastructure
  - Quantitative PC $\rightarrow$ thickness $\rightarrow$ trace element concentrations

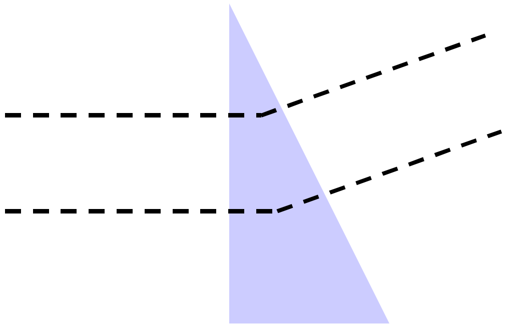


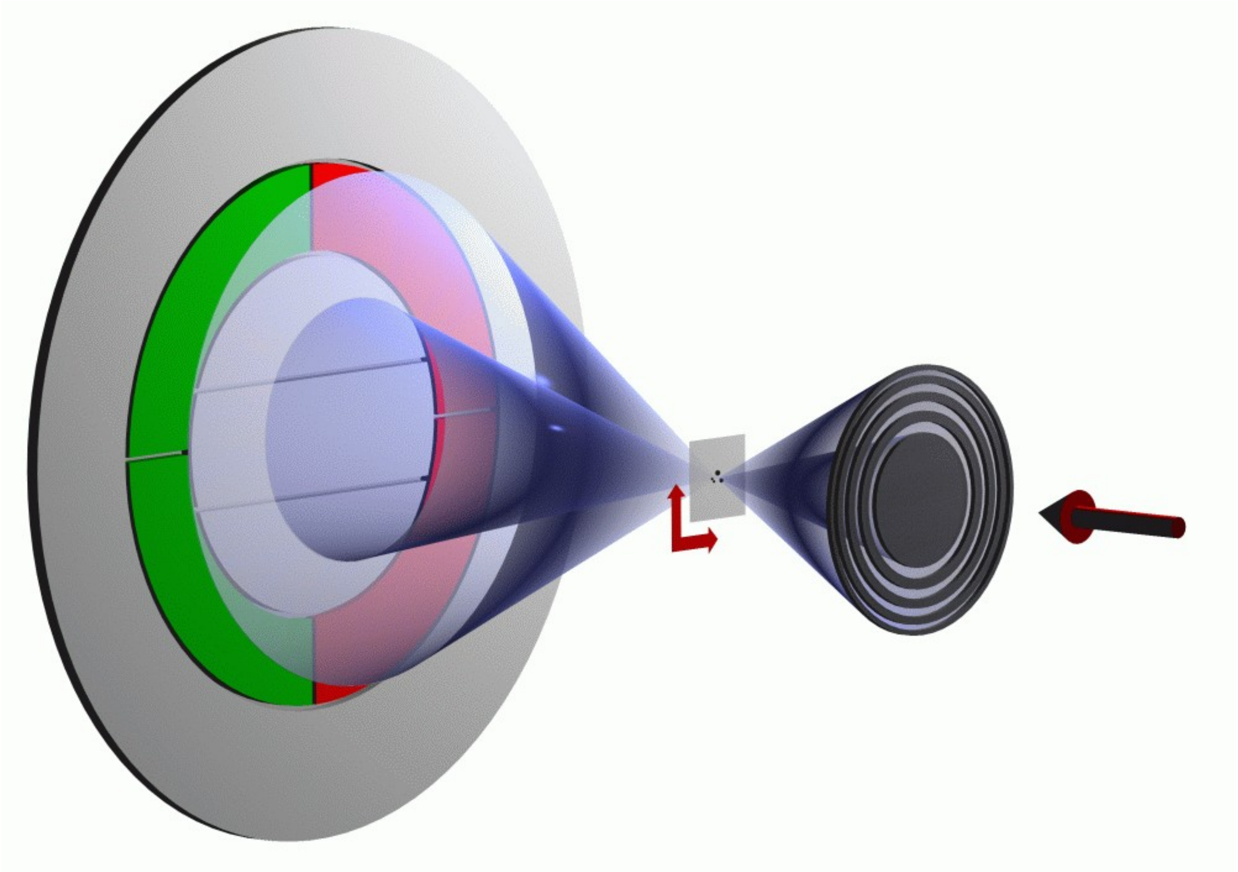


Data from Henke *et al.*

Differential Phase Contrast

- Refraction model – effect of **phase gradient** (like prism for visible light) :





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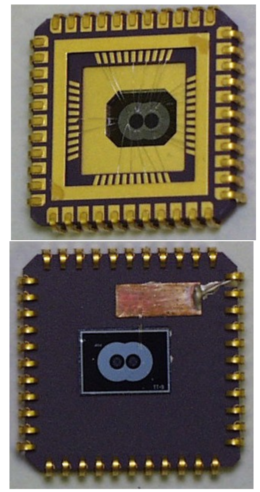
Why not use a CCD?

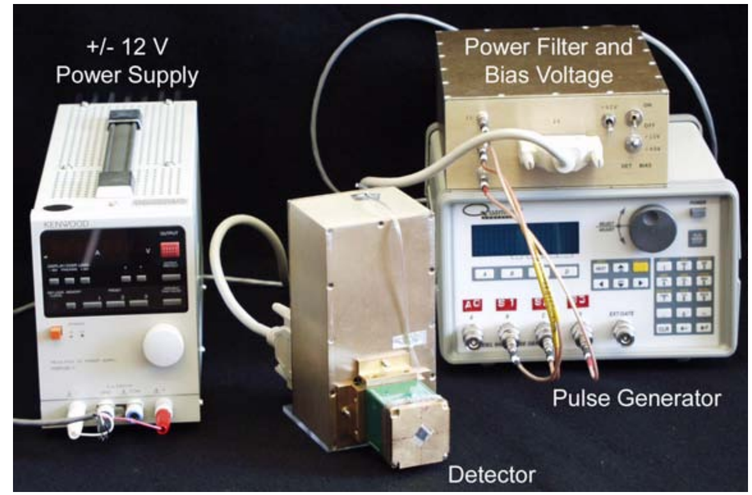
- Slow (serial) readout (tens of ms to sec) vs. ms pixel dwell times
- huge amounts of data
- statistical significance of a single detector pixel

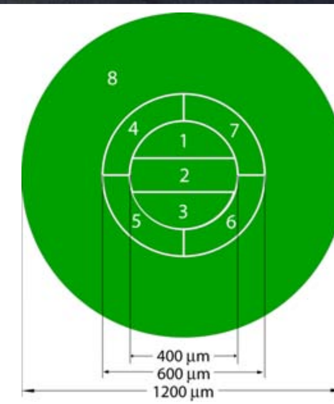
- fast readout pixel detectors in the future?

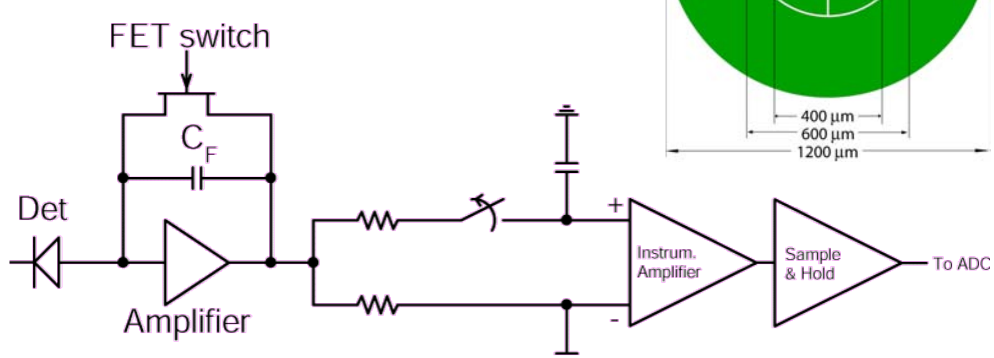
Review: Segmented Detector Version 1

- M. Feser, Ph.D. 2002, Nucl. Instr. Meth. A 565 (2006)
- **Collaboration** with
  - BNL Instrumentation (P. Rehak, G. De Geronimo
  - Max Planck Semiconductor Lab (L. Strüder, P. Holl)
- **For NSLS STXM:** 200-800 eV, 10^6 photon/sec
- **Segmented silicon chip** (high quantum efficiency)
  - rotational **symmetry**
- **Charge integrating** electronics (high count rates)
  - **Simultaneous** recording of all segments (**various contrast modes**)









Electronics: 10 channels

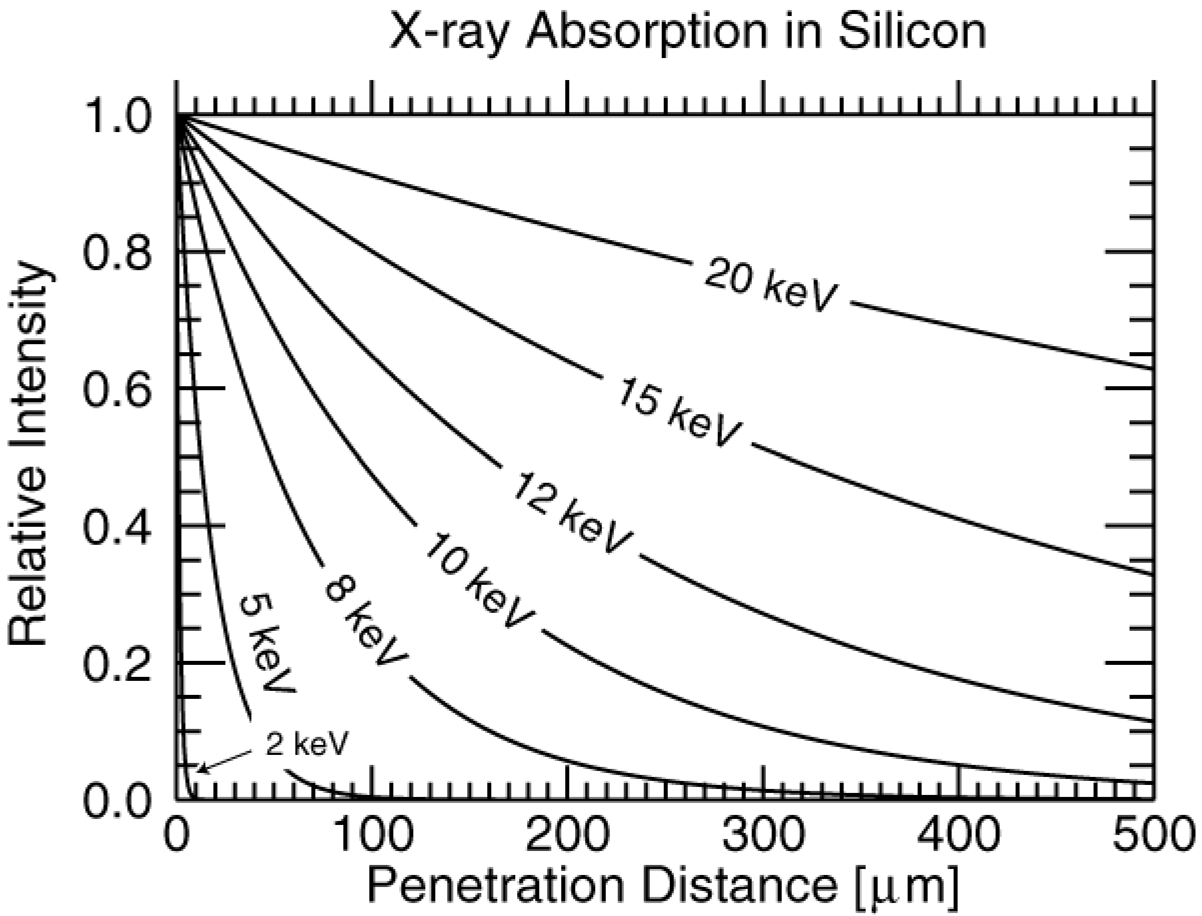
Modifications for Hard X-Rays (APS)

Beamline	Flux	Photon Energy	Current	Dwell Times
NSLS X-1A	10^6/s	200 – 800 eV	1-20 pA	1-10 ms
APS 2-ID-B	10^8/s	1 – 4 keV	1-100 nA	0.5-5 ms
APS 2-ID-E	10^9/s	7-17 keV	0.1-1 µA	sub-ms – sec
Nanoprobe	10^{10}/s	10 (- 30) keV	0.5-5 µA	sub-ms – sec

- **APS 2-ID-B:**
  - One NSLS detector modified with larger feedback capacitance
- **APS 2-ID-E:**
  - Used 15-20 layers of Al foil in front of detector to absorb > 99.5 % of the photons
  - Decouple detector integration time and pixel dwell time

X-ray Absorption in Silicon

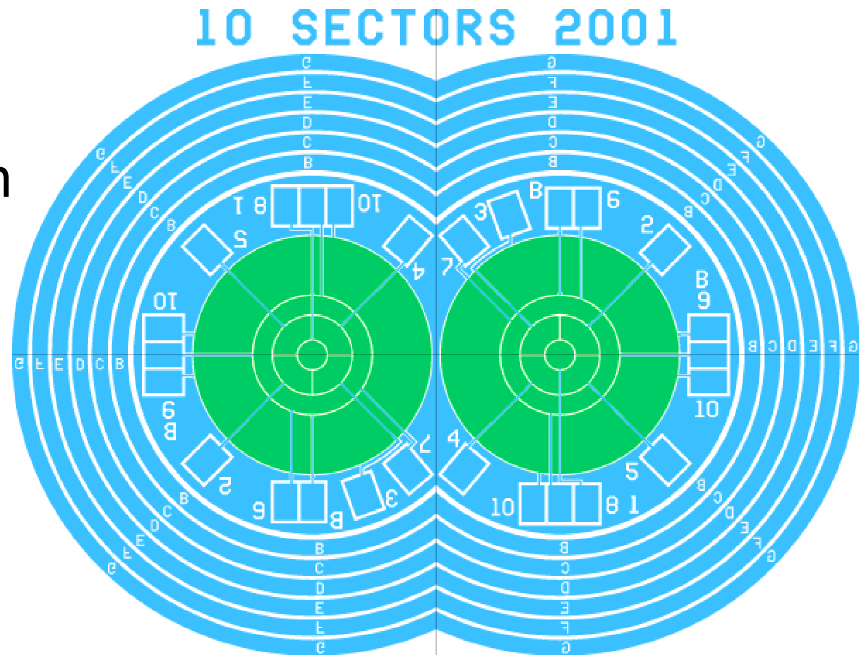
- To be detected, photons must be absorbed in (active region of) chip
- At higher energies, thickness limits quantum efficiency
- At lower energies (< 1 keV), absorption effects in surface oxide layer

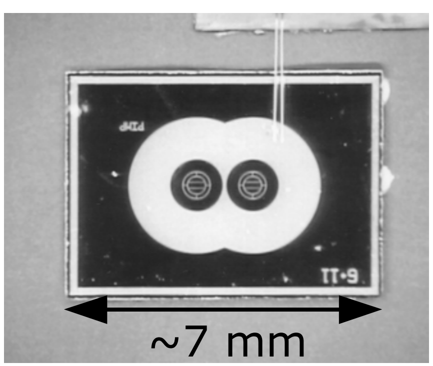


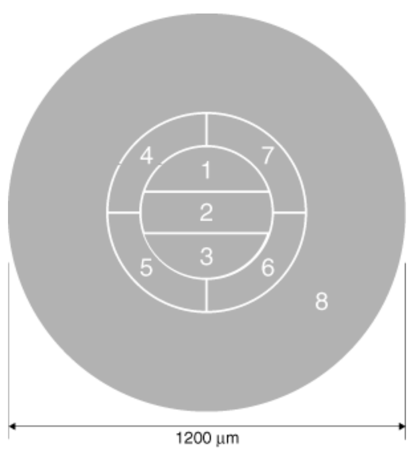
Data from Henke *et al.*

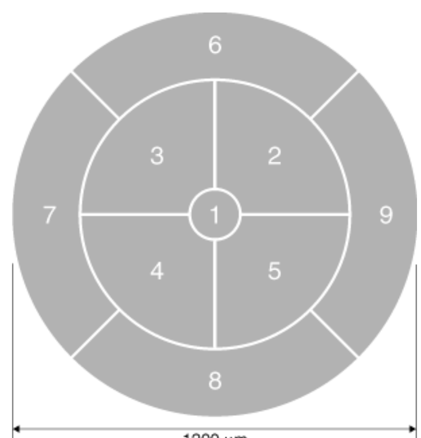
Segmented Silicon Chip

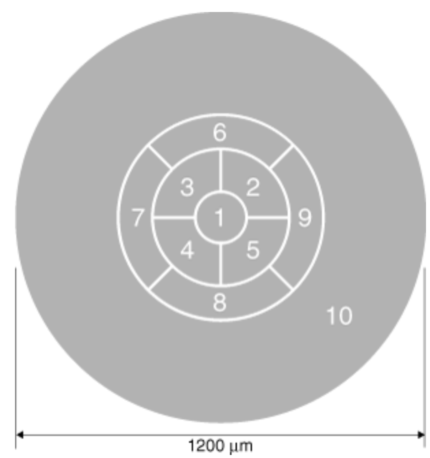
- Produced by Max Planck Semiconductor Lab
- 300 to 450 µm thick n-type silicon
- segments: shallow p-implant with current readout
- Ohmic junction on back side for bias voltage
- Can illuminate front or back side
- Extremely low leakage current





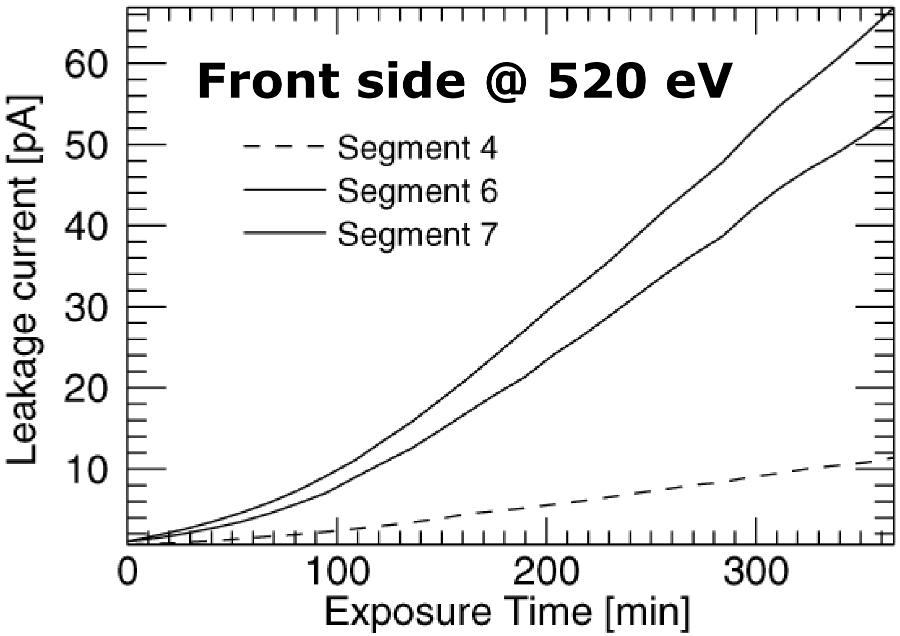






Radiation Damage

- Front side is radiation-sensitive
- Increase of leakage current with exposure
- Repair by annealing
- **Problems:**
  - Adds to signal → Calibration
  - Uses up part of dynamic range
- **Solution:**
  - Soft x-rays: Back side Illumination
  - Hard x-rays: Regular annealing

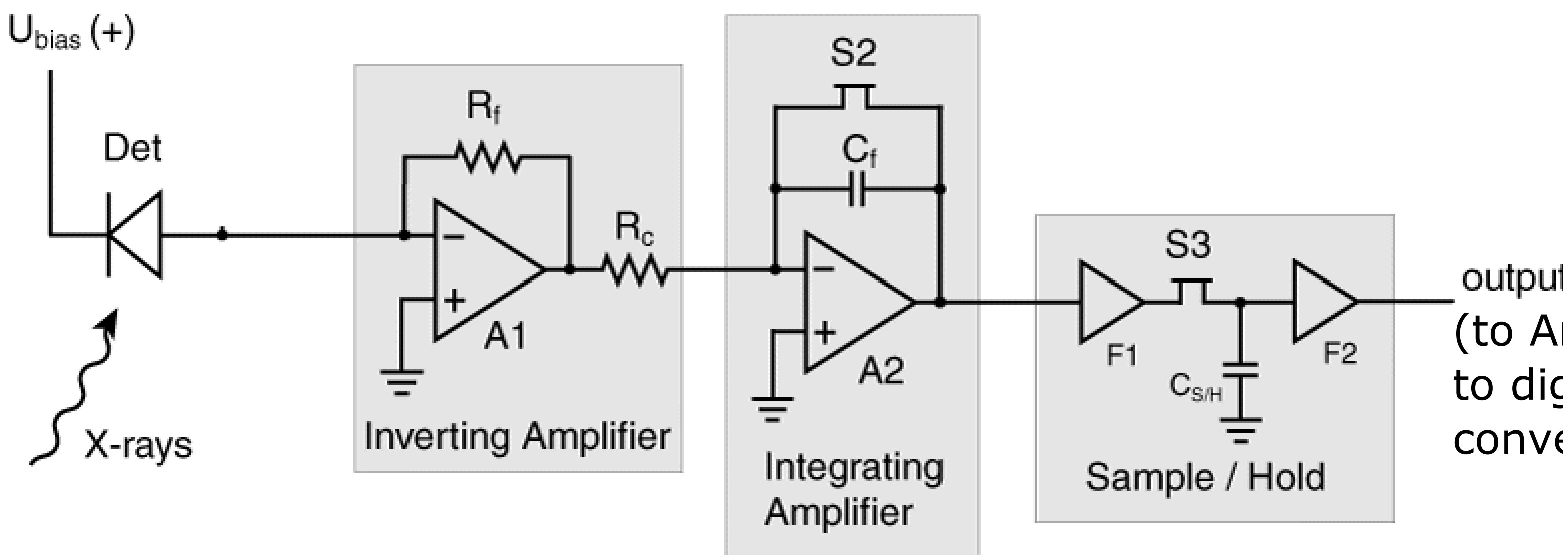


Back side @ 10 keV

Seg.	Leakage Current (pA)		
	Initial	3 days exp.	annealed
4	2	15	0.7
5	1.9	14	2
7	1.1	7.1	0.5

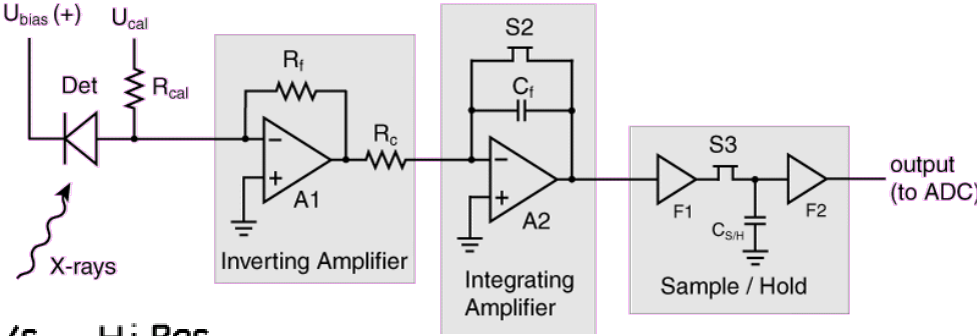
Charge Integrating Electronics

- 10 channels for up to 10 segments
- Current amplifier (adjusted to signal rate)
- Integrator (adjusted to dwell time)
- Sample and hold for readout
- Dead time ca. 10 µs



(to Analog to digital converter)

Integration Cycle

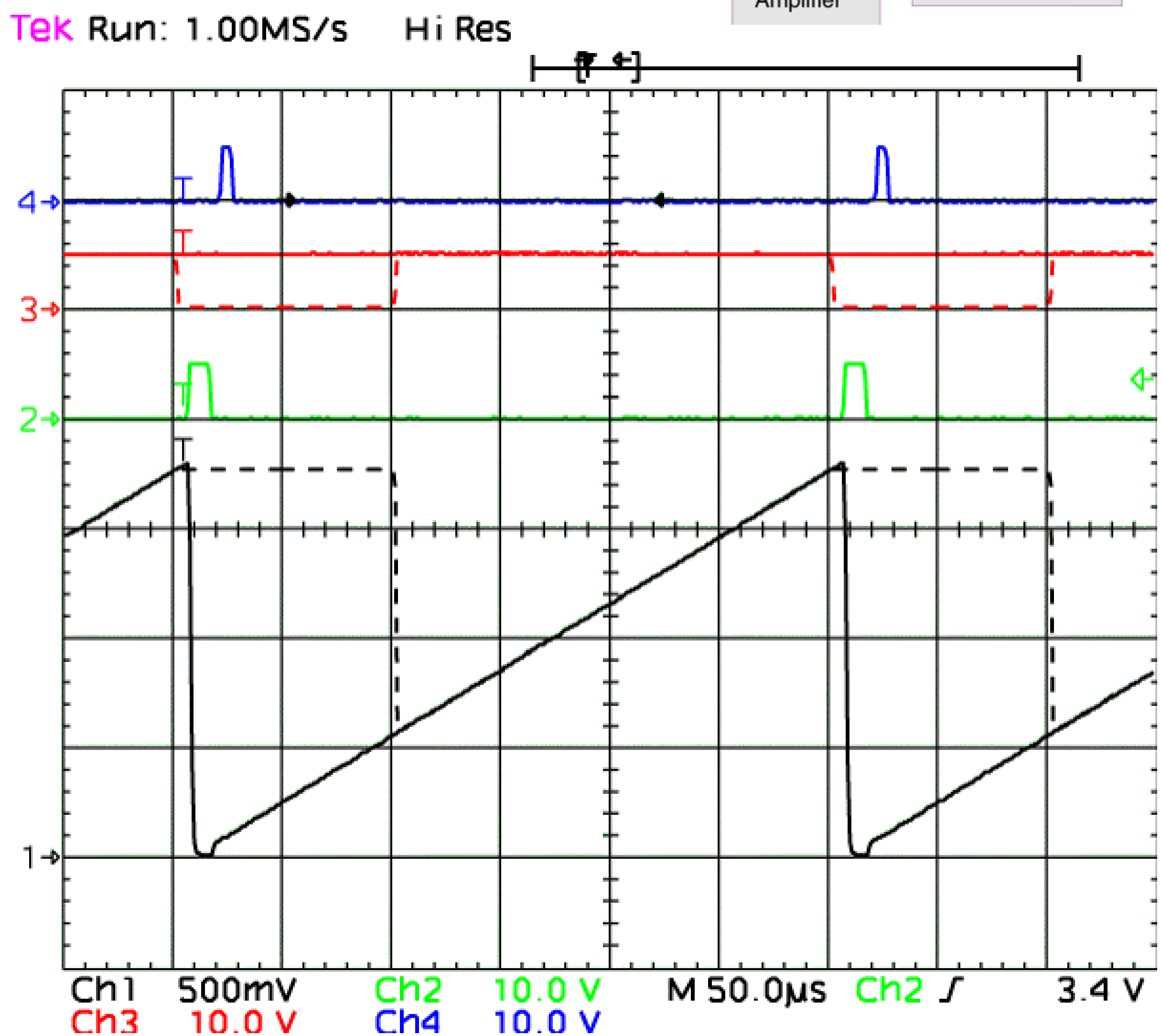


Trigger to ADC

S/H control pulse

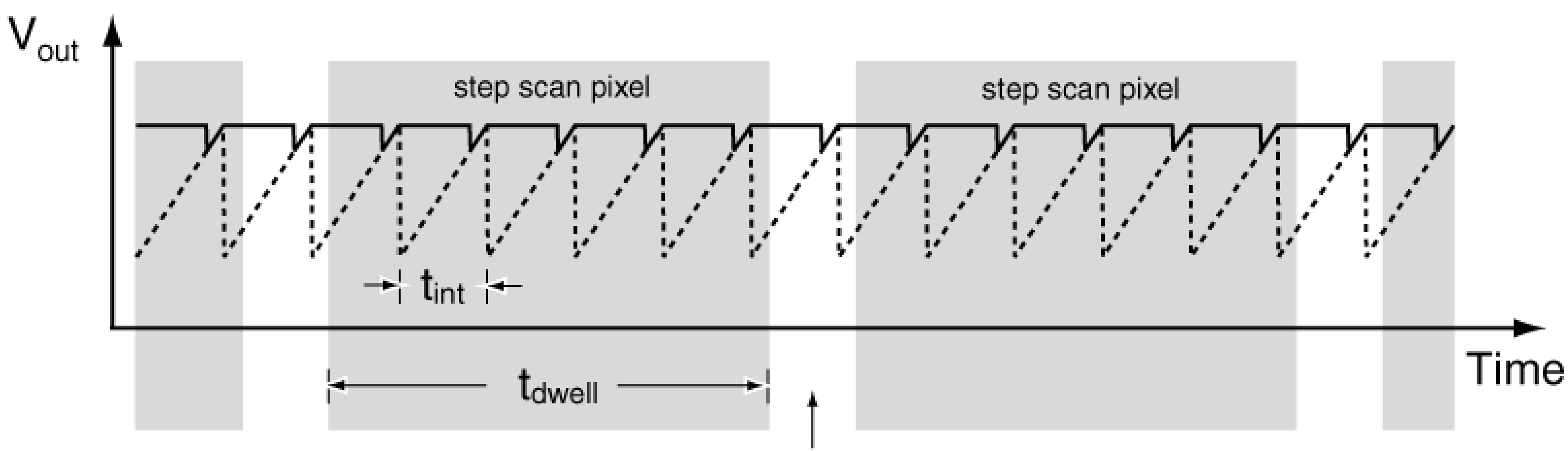
Integrator Reset pulse

S/H output



Interfacing with Microscope Electronics

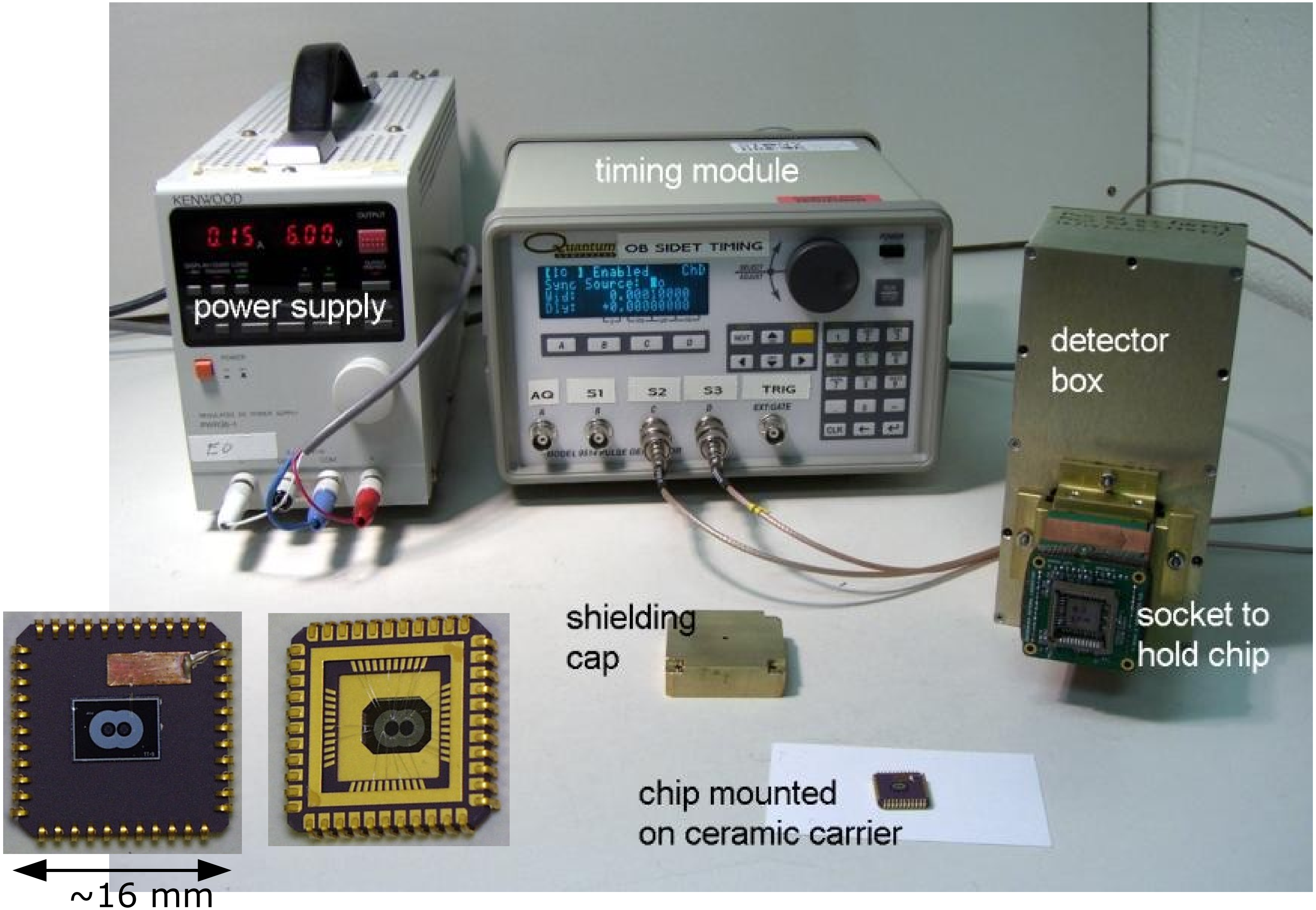
- Two scan modes:
  - Step scan (slow)
  - Fly scan (fast)
- Two signal types
  - Digital (pulse train)
  - Analog (voltage)
    - Voltage to Frequency converter (V2F)
- Operation in fly scan mode:
  - Scan pixels and detector integration in sync
  - Read voltage directly
- Operation in step scan mode:
  - Pixel dwell time >> integration time
  - Use V2F



Detector Calibration

- Measure amplifier output voltage, want photon flux
- Need to know
  - Photon energy (monochromatic illumination!)
  - Charge created per photon: 3.6 eV per e/h pair
  - Calibration constant between input charge and output voltage (amplifier gains, integrating capacitor)
  - Charge integration time (pixel dwell time)
  - Leakage current (measure signal with no x-rays incident for several dwell times)

Detector Components



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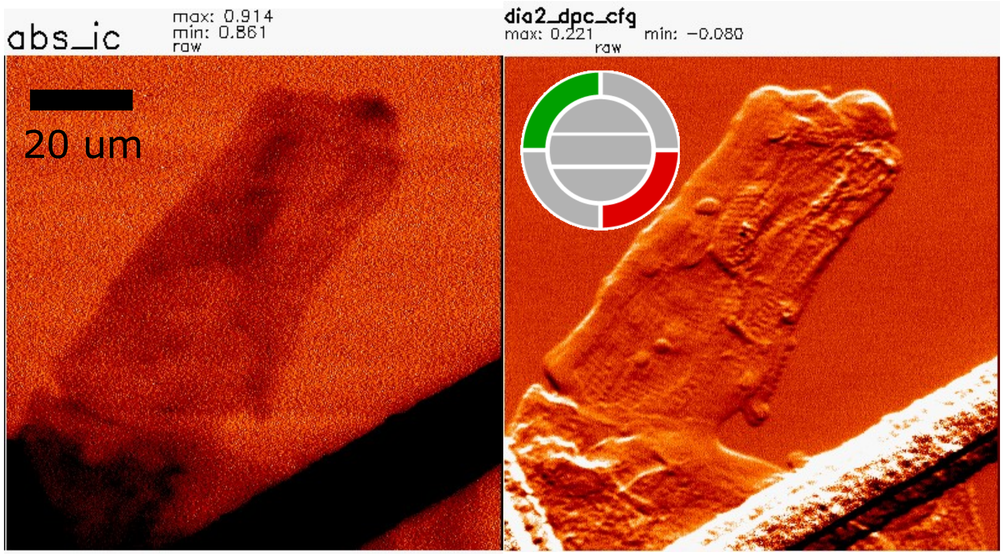
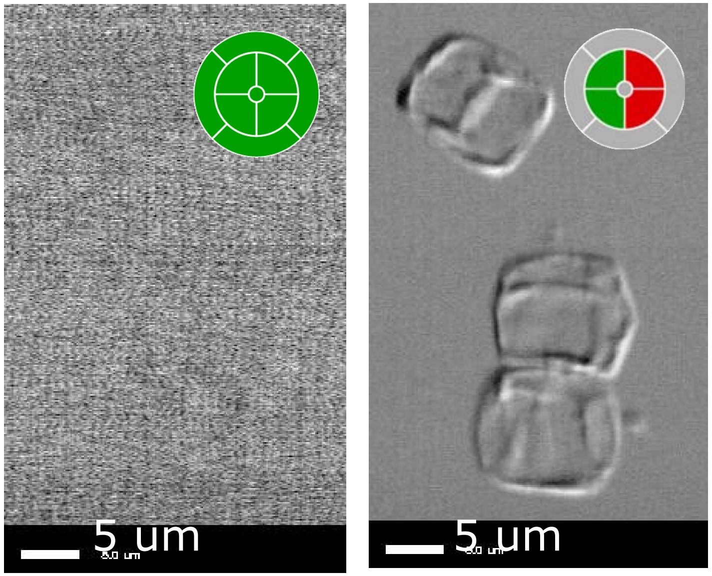
DPC Examples from APS 2-ID-E (8-10 keV)

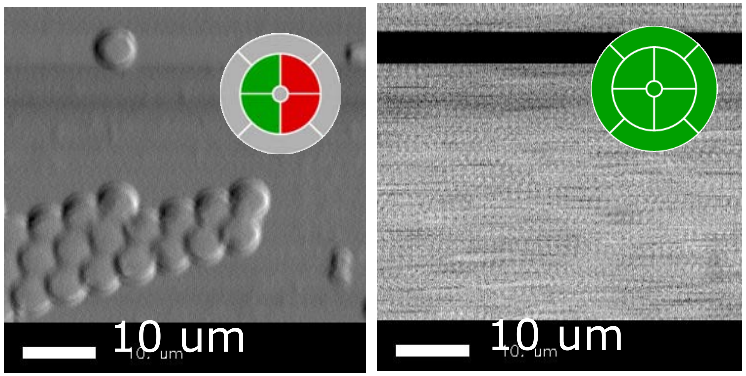


Cardiac myocyte (heart muscle cell) Sample: B. Palmer, U. Vermont. Data: Stefan Vogt (Modified soft x-ray detector)

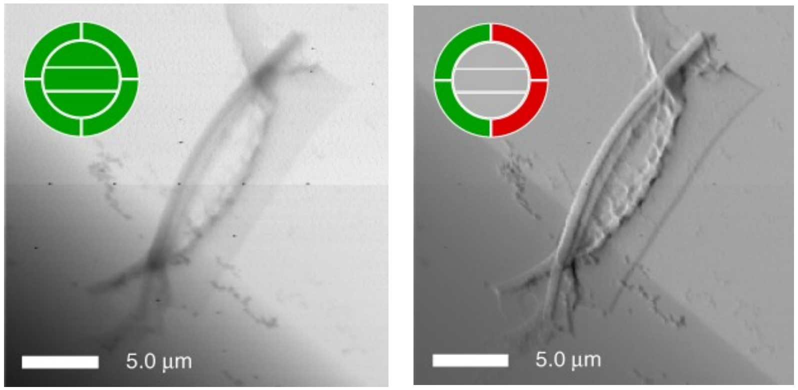


Diatoms (phytoplankton). Sample: Stephen Baines, Stony Brook Marine Sciences.

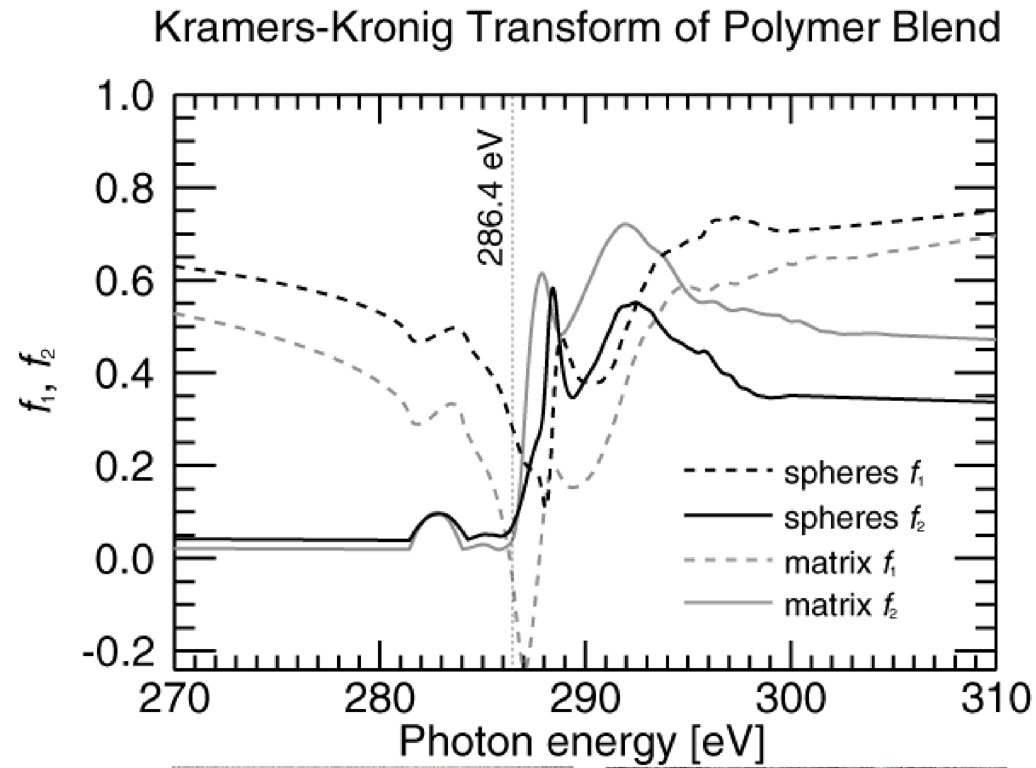
5 µm **Polystyrene spheres**



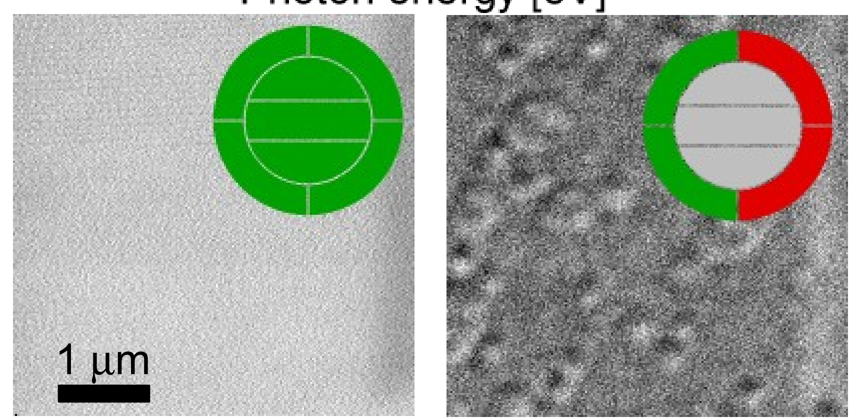
At Lower Energies



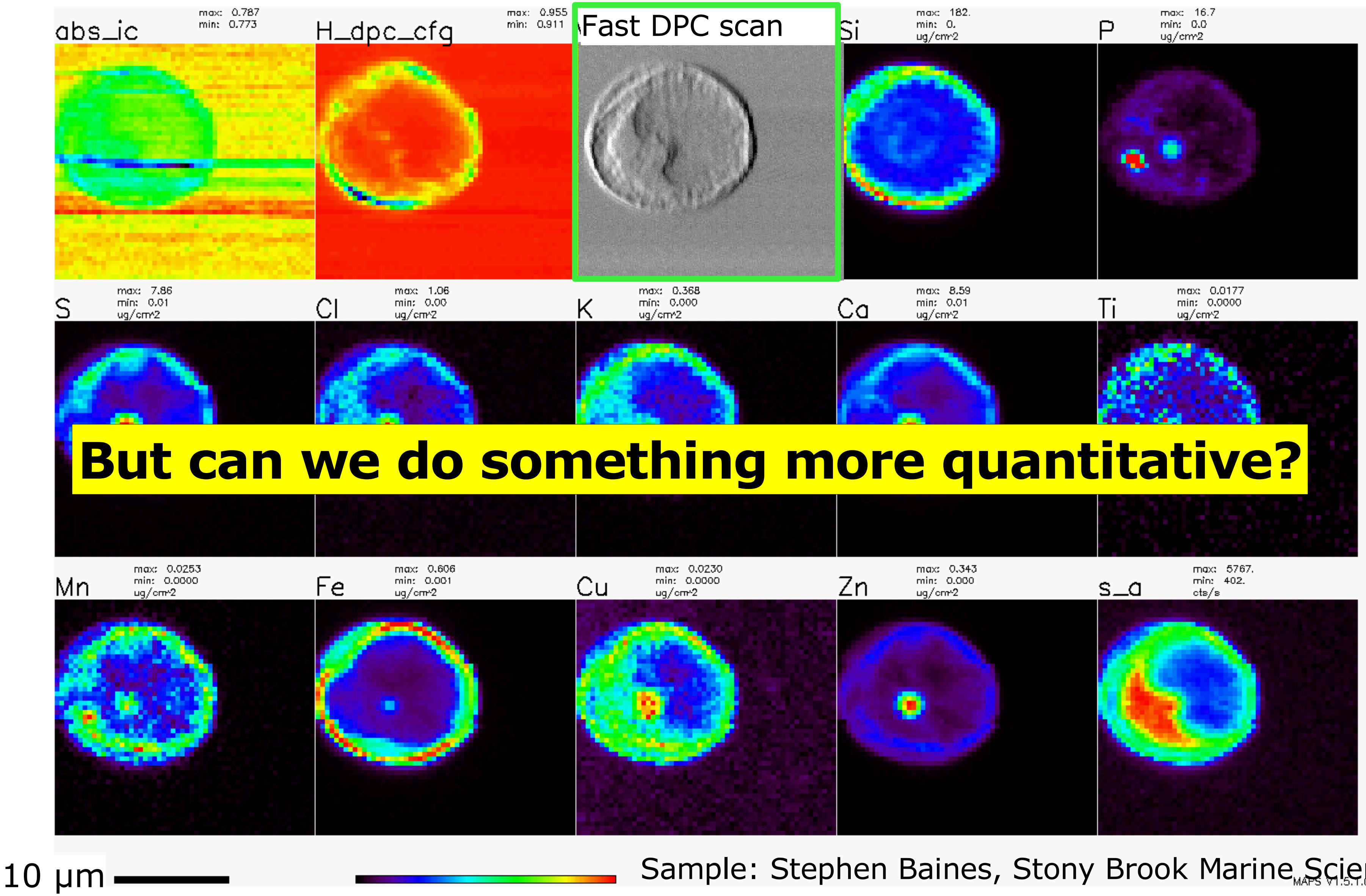
Diatom at 2-ID-B (1.8 keV)



Polymer spheres in polymer matrix @ 286.4 eV (NSLS STXM)
(sample provided by Gary Mitchell, Dow Chemical)

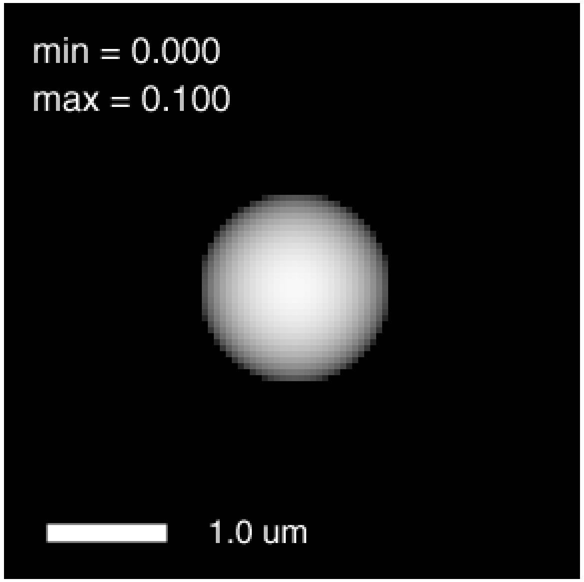


Combination with Fluorescence

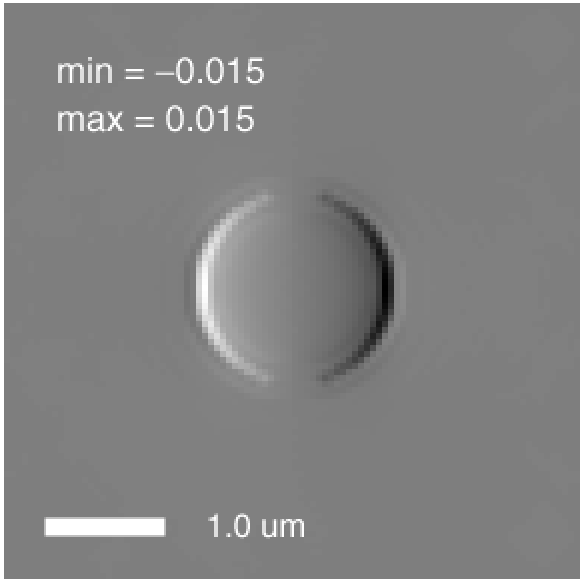


Sample: Stephen Baines, Stony Brook Marine Sciences

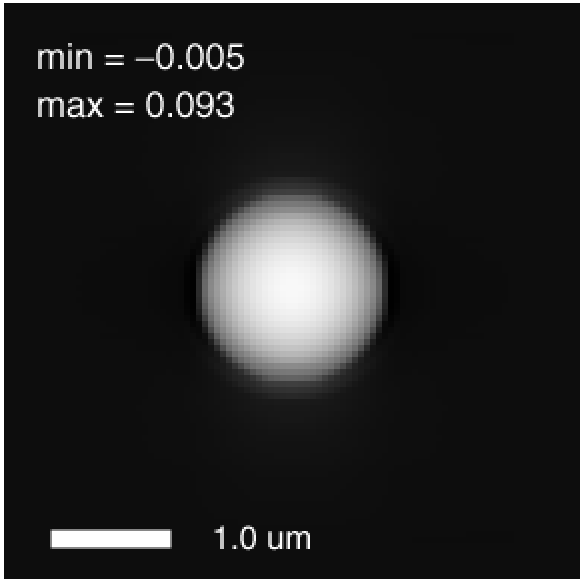
DPC Integration – Noise-Free Simulations



Simulated sphere



Simulated DPC image

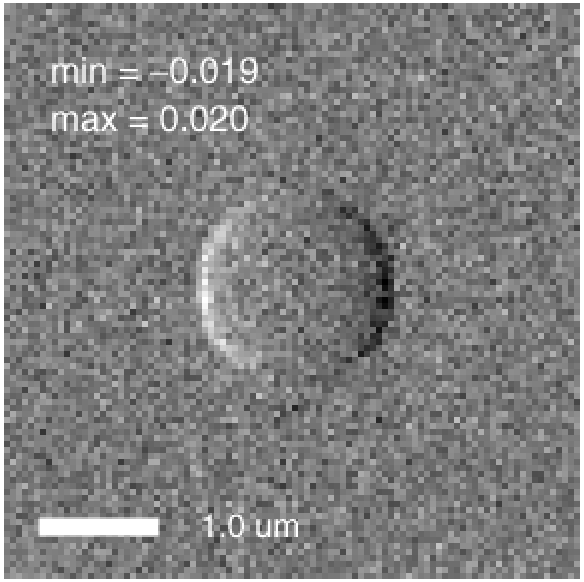


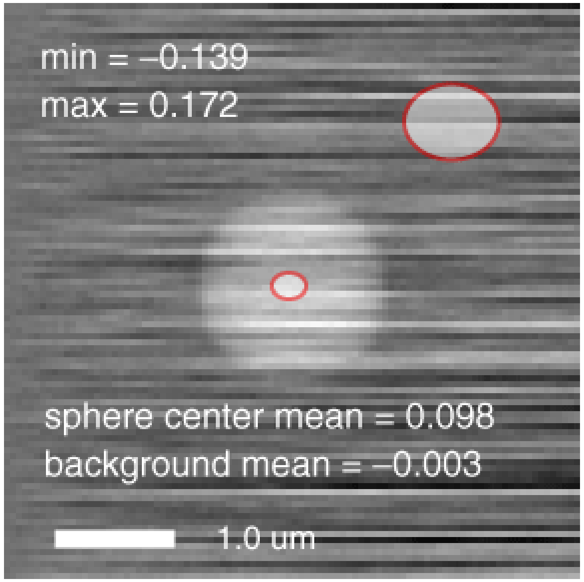
Integrated DPC image

- Sphere: max. phase shift 0.1 rad, no absorption
- Image simulated with “true” wave propagation
- No noise

Simulations with Noisy Data

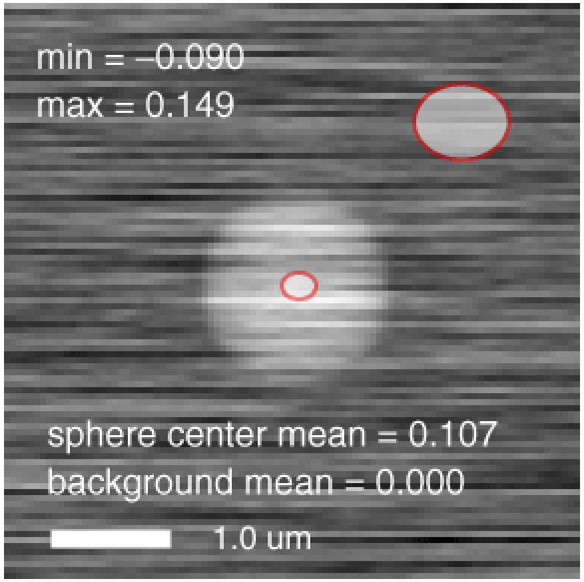
DPC image

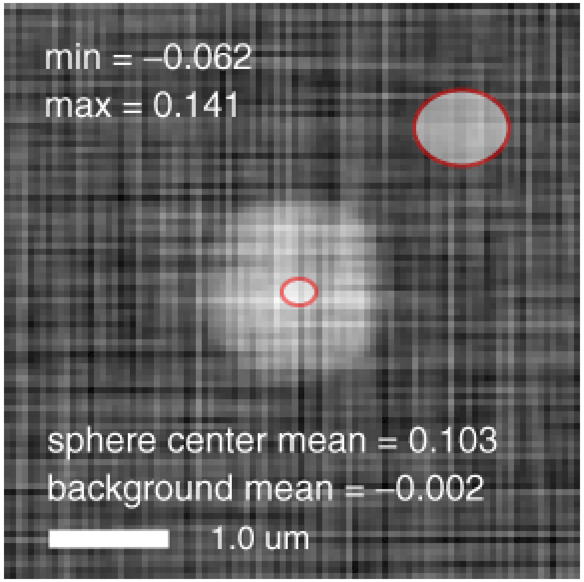




one-directional integration

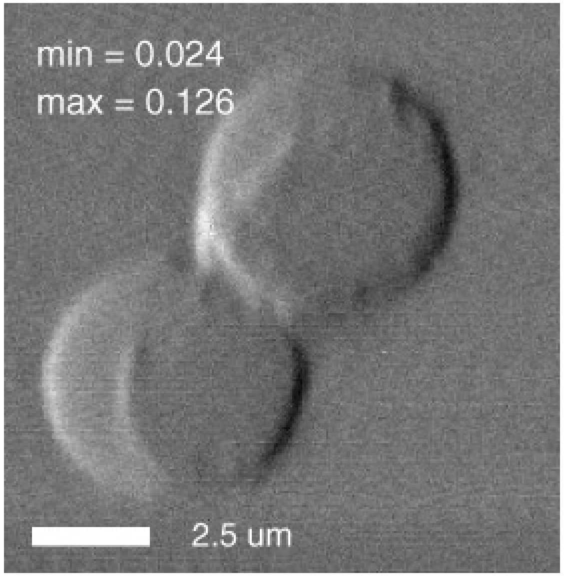
bi-directional integration



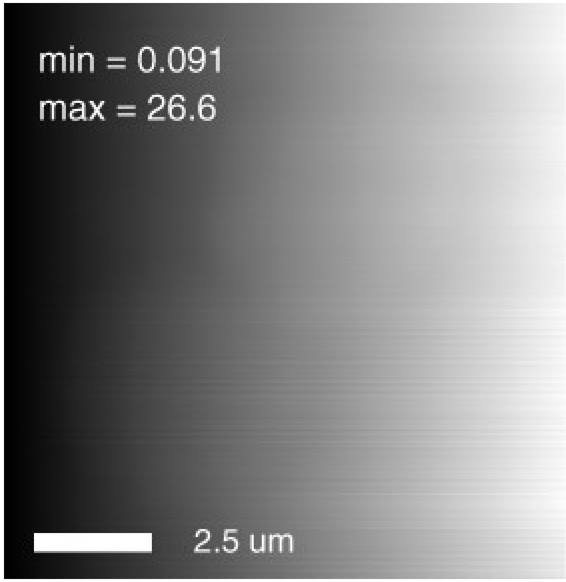


two orthogonal bi-directional integrations

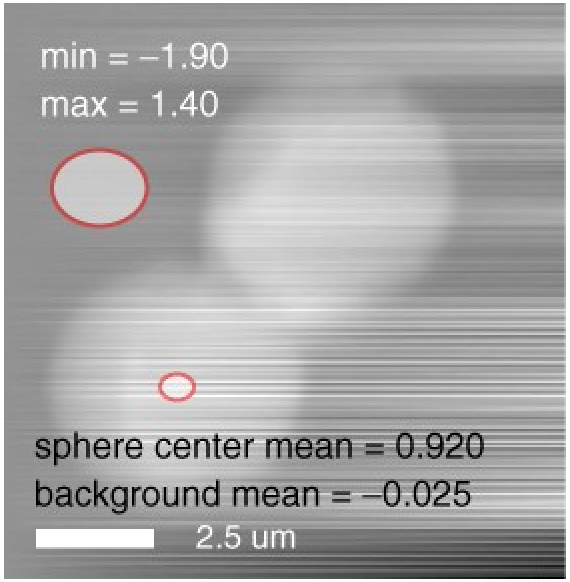
Integration of DPC Data



DPC image



Simple integration



Background norm.

- 5 μm diameter polystyrene spheres
- E = 10 keV
- expected $\delta kt = 0.60$

DPC – Conclusions

- Vastly improved contrast for weakly absorbing specimens at multi-keV energies
- Easily available with segmented detector (real-time)
  - Quick orientation images (finder scans)
  - High-resolution images of sample morphology

- Hard to interpret
  - Differential signal
  - Directional dependence
  - Hard to quantify
  - Simple integration doesn't work well

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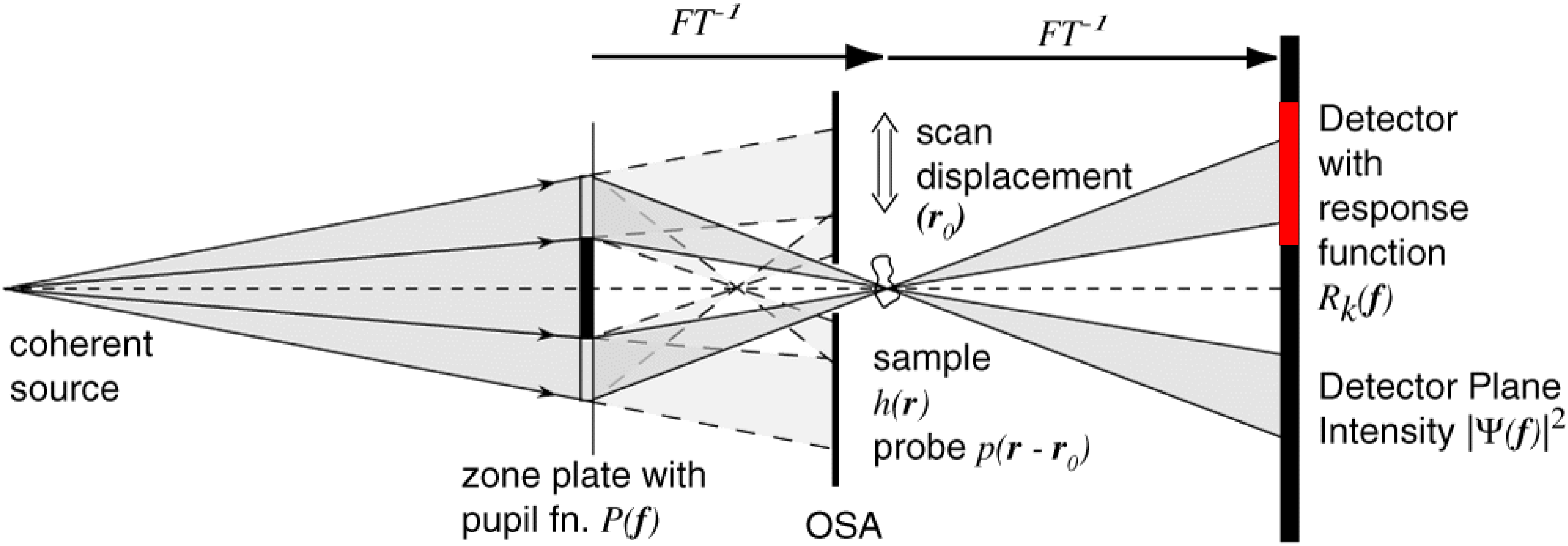
Image Formation in a Scanning Instrument



- **Wave propagation** from source to detector plane
- Segmented detector

Contrast Transfer Functions

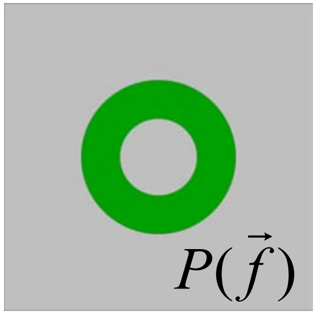
- Complex specimen function: $\Psi_{\text{out}}(r) = h(r - r_0) \cdot \Psi_{\text{in}}(r)$
- Weak specimen approximation: $h(r) = 1 + h_r(r) + i h_i(r)$

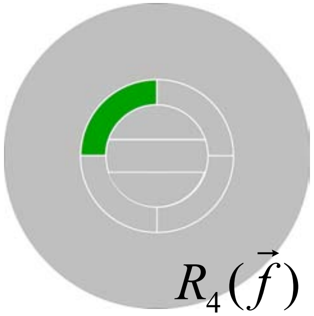
$$h_r(r) = -\beta(r)\, k\, dz \qquad h_i(r) = \delta(r)\, k\, dz$$

- Image recorded by detector segment *k* (Fourier space)

$$S_k(f) = c_k \cdot \delta(f) + H_r(f) \cdot T_r^k(f) + i H_i(f) \cdot T_i^k(f) + N_k(f)$$

- Contrast Transfer Functions depend on
  - P: Zone plate pupil function (assume coherent illumination)
  - R: Detector response function



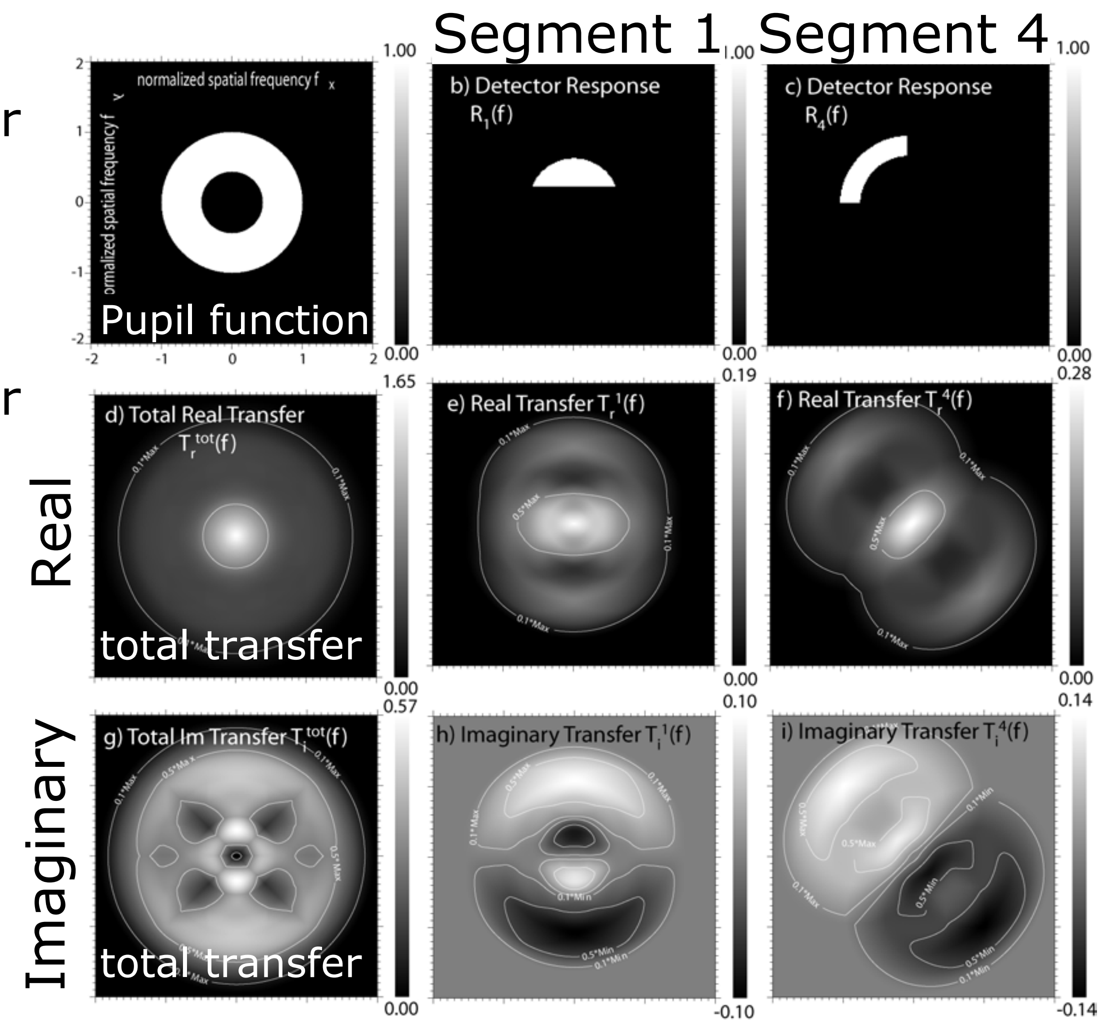


Calculated Contrast Transfer Functions

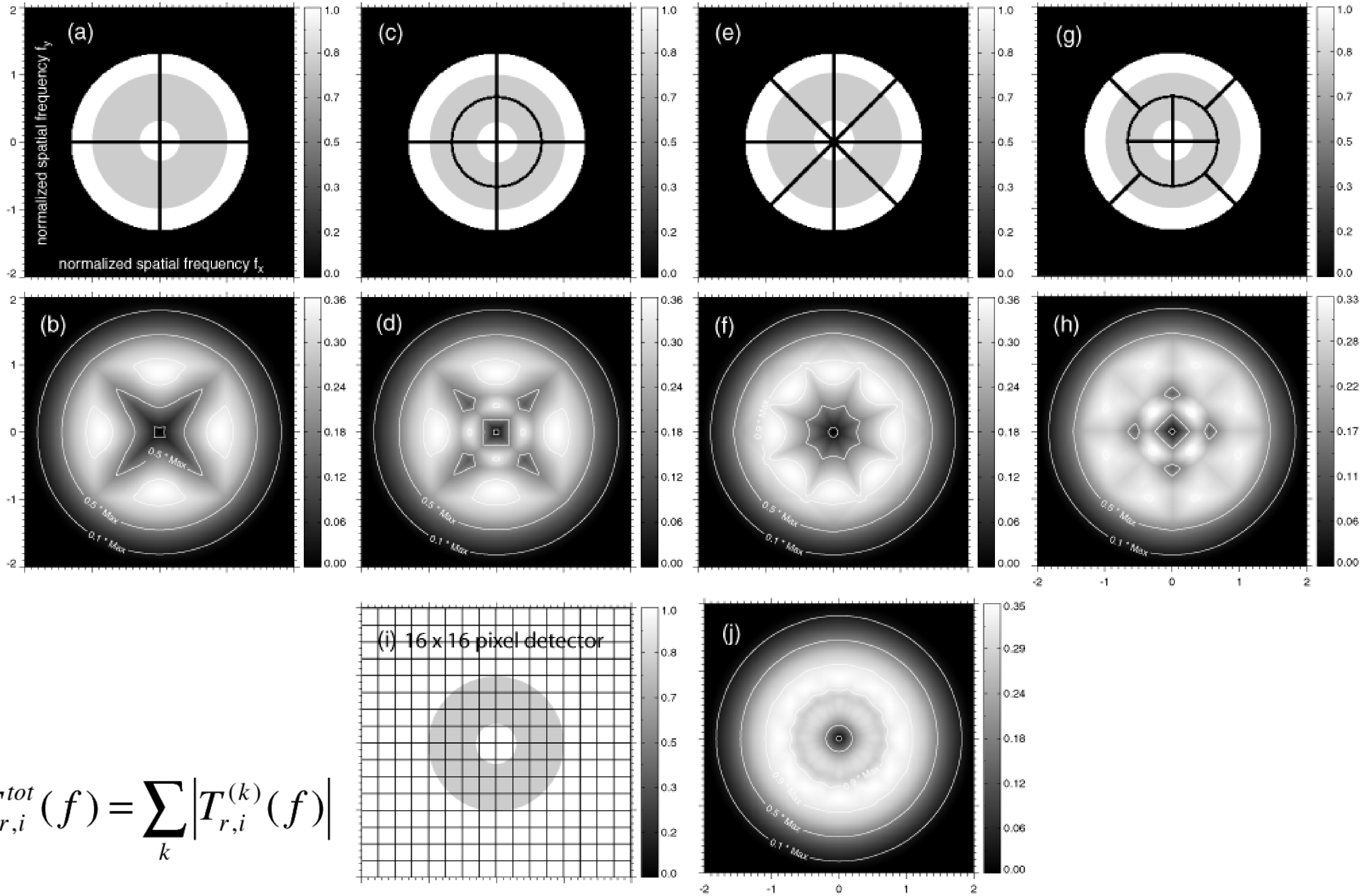
- Real part CTFs:
  - even symmetry
  - CTFs for opposing detector segments are identical
- Imaginary part CTFs:
  - odd symmetry
  - CTFs for opposing detector segments are opposite in sign

→ **Sum** of opposing segments shows only ***absorption contrast***

→ **Difference** of opposing segments shows ***differential phase contrast***



Comparison of Detector Geometries



$$T_{r,i}^{tot}(f) = \sum_k \left| T_{r,i}^{(k)}(f) \right|$$

Amplitude and Phase Reconstruction

- Reconstruction of the complex specimen function by **Fourier filtering** detector images
- Proposed for scanning transmission electron microscopy (McCallum *et al.*, Optik 101(2) 1995)
- Similar to Wiener Filter
- **Best estimate** of complex specimen function:

$$\breve{H}(f) = \sum_k W_k(f) S_k(f)$$

Images from det. segments

Filter functions

- Calculate filter functions by minimizing reconstruction error
- Weak specimen approximation
- Account for noise

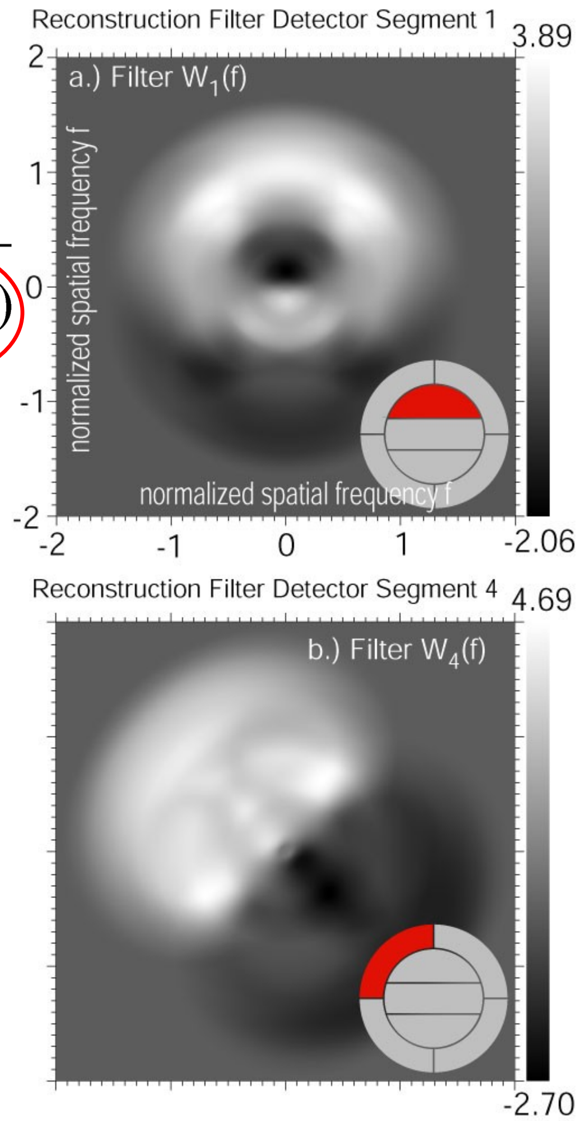
Reconstruction Filters

- Result for filter functions

$$W_k(f) = \frac{T_r^{k*}(f)}{\sum_l \left|T_r^l(f)\right|^2 + \beta_r^k(f)} + \frac{T_i^{k*}(f)}{\sum_l \left|T_i^l(f)\right|^2 + \beta_i^k(f)}$$

- Noise parameter

$$\beta_{r,i}^k(f) = \frac{\left\langle \left|N_k(f)\right|^2 \right\rangle}{\left|H_{r,i}(f)\right|^2} = \frac{\text{Spectral noise of segment } k}{\text{Specimen power spectrum}}$$

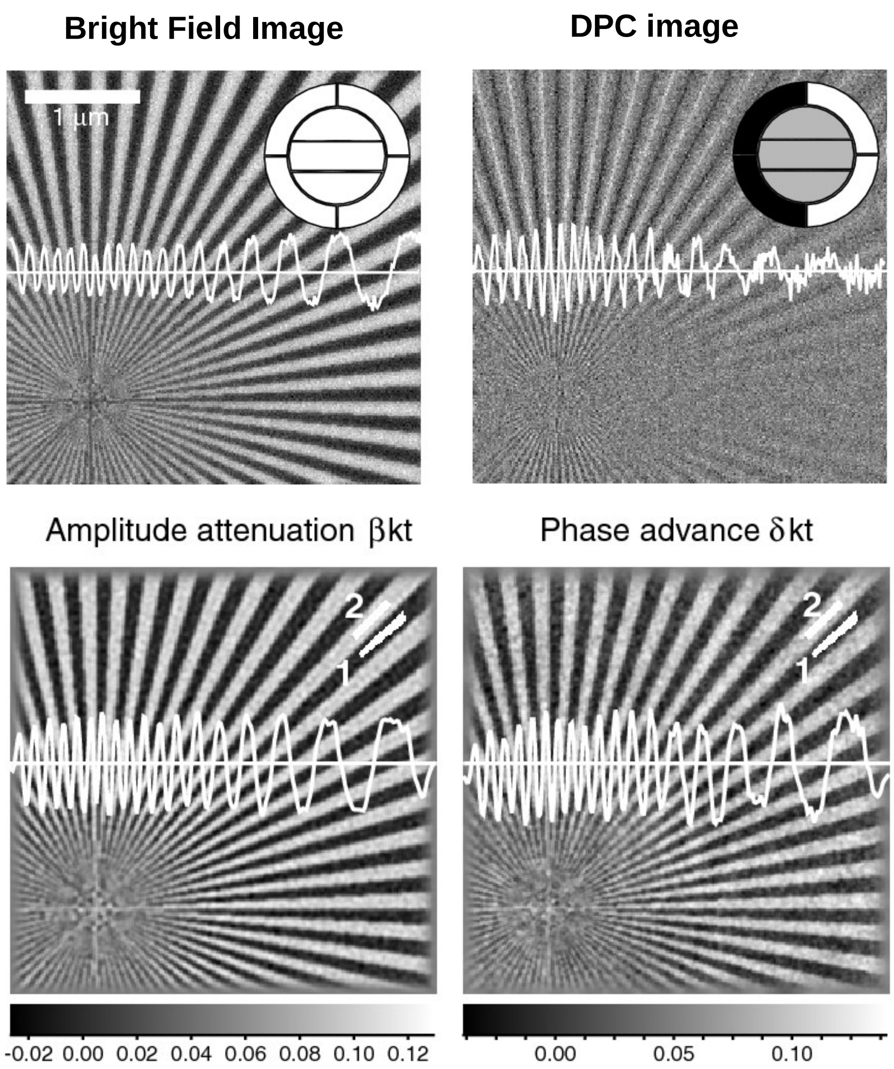


Soft X-ray Simulations of a Test Pattern

- Simulated weak and strong test pattern
- Conditions as in experiment (next slide)

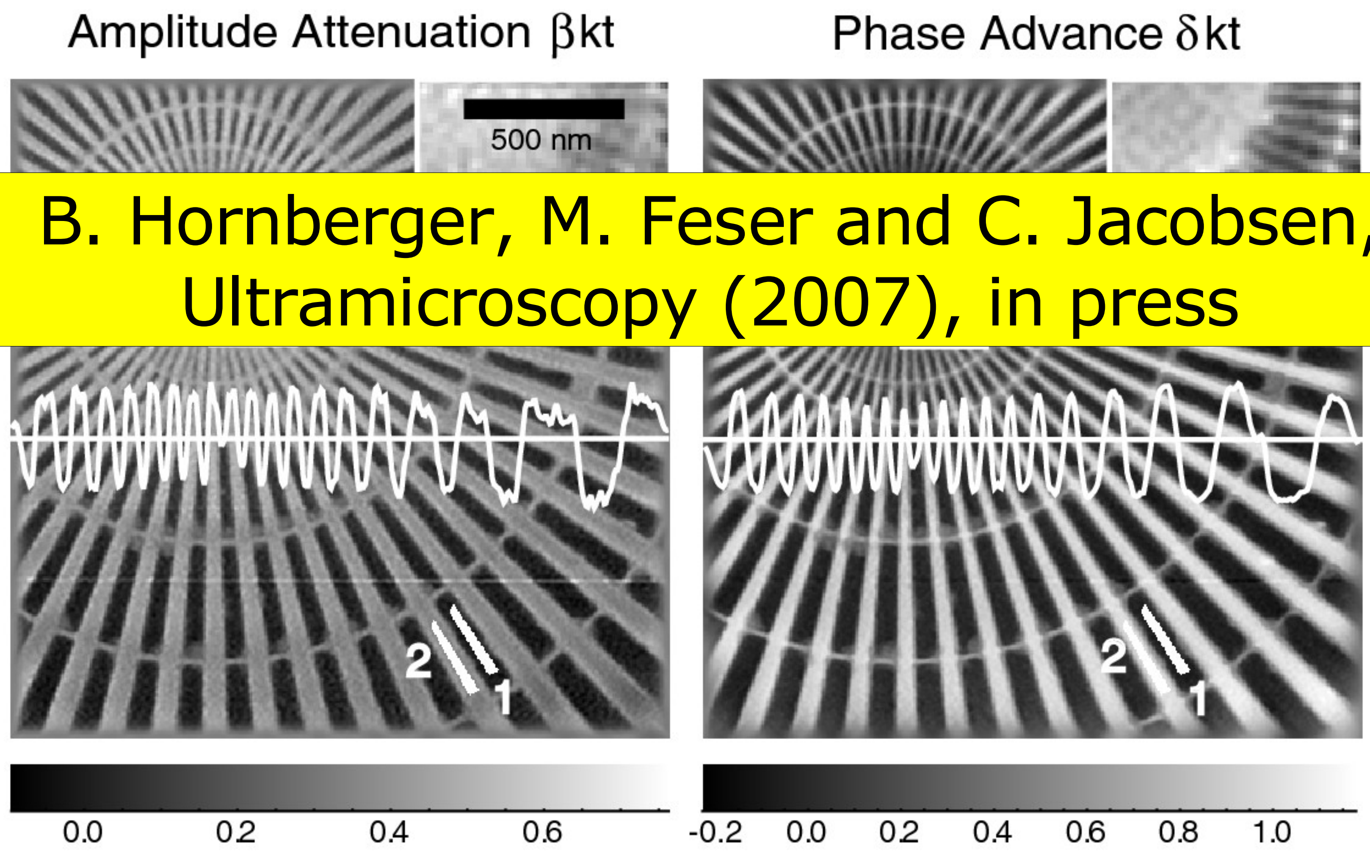
	weak specimen		strong specimen	
	simul.	recon.	simul.	recon.
βkt	0.100	0.098	0.410	0.349
δkt	0.100	0.103	1.140	0.896

Weak specimen simulation



Reconstruction of a Germanium Test Pattern

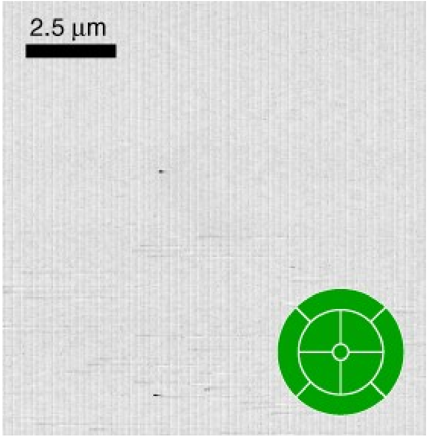
- Data acquired by Michael Feser @ 525 eV
- Recovered βkz ≈ 0.35, δkz ≈ 0.99 in good agreement with expected values
- More details in the phase image

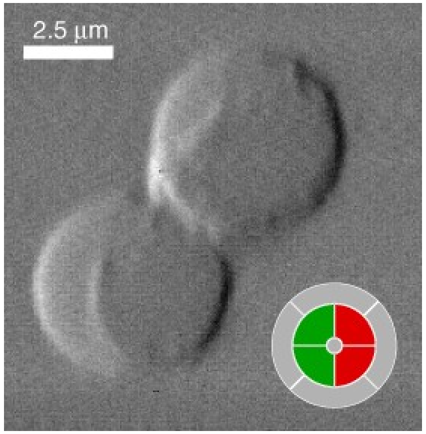


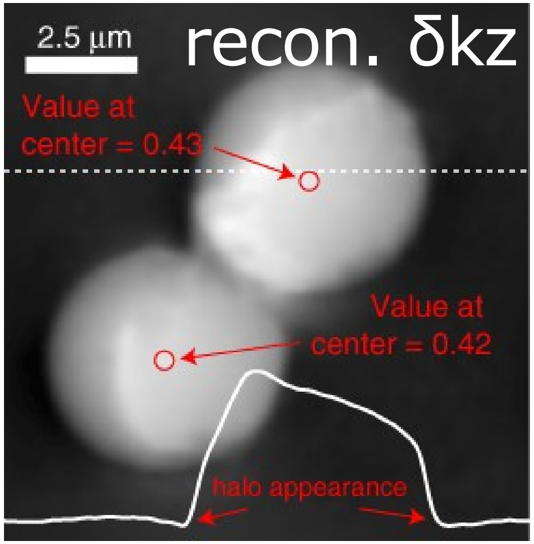
B. Hornberger, M. Feser and C. Jacobsen, Ultramicroscopy (2007), in press

Polystyrene Spheres at APS 2-ID-E

- 5 µm Polystyrene spheres
- 10 keV photon energy
- invisible in amplitude contrast
- expected δkz ≈ 0.6
- reconstructed δkz ≈ 0.43
- Uneven zone plate illumination?
- Limited knowledge about zone plate?
- Independent verification of expected value?







Outline

- Introduction
  - X-ray Microscopy 101
  - Phase Contrast 101
- A Segmented Detector for Hard X-ray Microprobes
  - Segmented Silicon Chip
  - Charge Integrating Electronics
- Differential Phase Contrast (DPC)
  - Comparison with Amplitude Contrast and DPC Examples
  - Integration of the DPC Signal
- Quantitative Amplitude and Phase Reconstruction
  - Reconstruction Scheme
  - Simulations and Experiments with Soft and Hard X-rays
- Summary and Outlook

Summary

- Phase contrast is useful!
  - reduce radiation dose at lower energies
  - superior transmission contrast at higher energies
    - combination with fluorescence
    - high resolution images of specimen ultrastructure
    - fast finder scans
- Segmented detector for hard x-ray microprobes
  - simultaneous amplitude and phase contrast
  - installation in parallel with fluorescence detector
  - segmented silicon chip
  - 10 channel charge integrating electronics
  - adjustable dynamic range
  - wide range of pixel dwell times
  - absolute calibration

Summary (2)

- Differential phase contrast
  - vastly superior contrast at higher energies
  - easily available
  - not so good for quantitative interpretation
  - simple integration doesn't give good results
- Quantitative amplitude and phase reconstruction by Fourier filtering
  - quantitative phase contrast can give specimen mass / thickness
  - “invert” image formation process
  - includes noise filter
  - works great in simulations for weak specimens
  - good experimental results
  - more careful measurements and consideration of experimental conditions

Future Work

- Detector installation at more beamlines
  - APS 2-ID-B, 2-ID-D, Nanoprobe
  - Australian Synchrotron
  - Better incorporation in data acquisition system
- More investigations about the Fourier filtering algorithm
- Go beyond test specimens to “real” applications
- Future hardware improvements?
  - fast readout pixel detectors
  - germanium detectors for higher energies

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- Hard x-ray experiments: David Paterson, Stefan Vogt, Daniel Legnini, Martin de Jonge, Ian McNulty (APS)
- Detector chips: L. Strüder, P. Holl *et al.* (Max Planck Institute)
- Electronics layout / assembly: R. Ryan, J. Triolo, D. Pinelli (BNL Instrumentation)
- Samples: B. Palmer (U. Vermont), M. Kissel (CEMS / Stony Brook), S. Baines *et al.* (Stony Brook Marine Sciences)