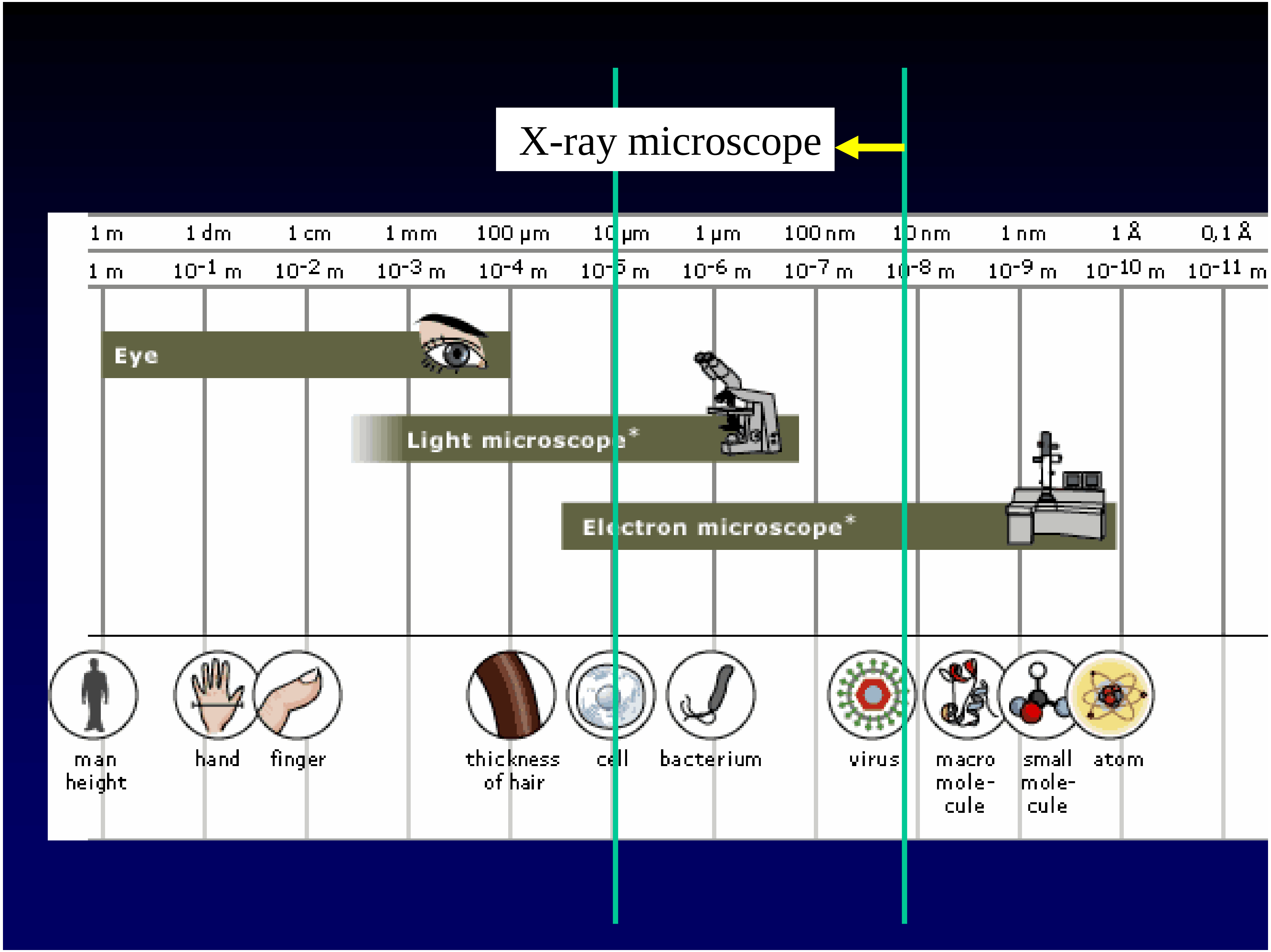
X-ray Microscopy

1. Synchrotron light
2. X-ray optical elements
3. Imaging system of x-ray microscopy
4. Examples
5. Lensless x-ray microscopy

湯茂竹　　　國家同步輻射研究中心

1. “Elements of synchrotron light for biology, chemistry & medical research”, by G. Margaritondo, Oxford, 2001.
2. “同步輻射顯微鏡成像”, 謝行恕&賈成芝, 聯經, 1995
3. D. Wess *et al*., Ultramicroscopy 84 (2000) 185-197.
4. C. Larabell *et al*., Molecular Biology of the Cell, 15 (2004), 957–962. Journal of Microscopy, 201, (2001), 395-403.
5. G. Schneider, Ultramicroscopy 75 (1998) 85.



X-ray microscope
1 m
1 dm
1 cm
1 mm
100 µm
10 µm
1 µm
100 nm
10 nm
1 nm
1 Å
0,1 Å
1 m
10-1 m
10-2 m
10-3 m
10-4 m
10-5 m
10-6 m
10-7 m
10-8 m
10-9 m
10-10 m
10-11 m
Eye
Light microscope*
Electron microscope*
man height
hand
finger
thickness of hair
cell
bacterium
virus
macro mole-cule
small mole-cule
atom

Advantages of Hard X-ray Imaging

- High penetration for nondestructive and in-situ imaging
- Short wavelength for high resolution
- Rich image contrast mechanism: absorption, chemical state, phase, polarization
- Element specific: absorption edges and characteristic fluorescence
- Easier sample preparation, compared to EM.

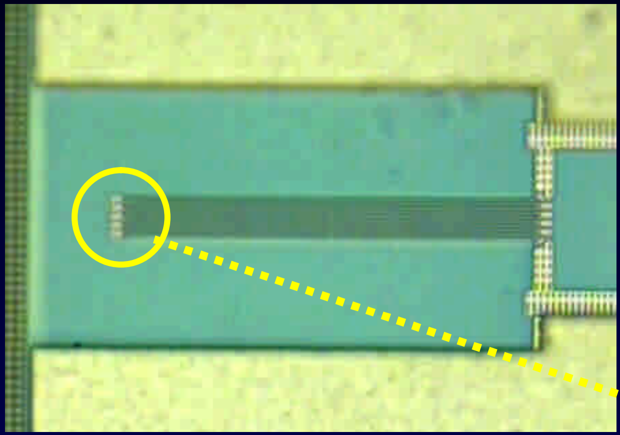
Comparison among microscopes

Feature	X-ray Microscope	Optical	SEM	TEM
Spatial resolution	15 - 25 nm	200-300 nm	1-10 nm typically	0.1 nm
Probing depth	1-50 μm for E<10 keV, 20 mm for E=100 keV	<100 nm for metal, Opaque or transparent materials	< 10 nm typically	<100 nm
Material class	Insulator, semiconductor, and conductor	Insulator, semiconductor, and conductor	Conductive path required	Conductive path required
Vacuum requirement	No	No	Yes	Yes

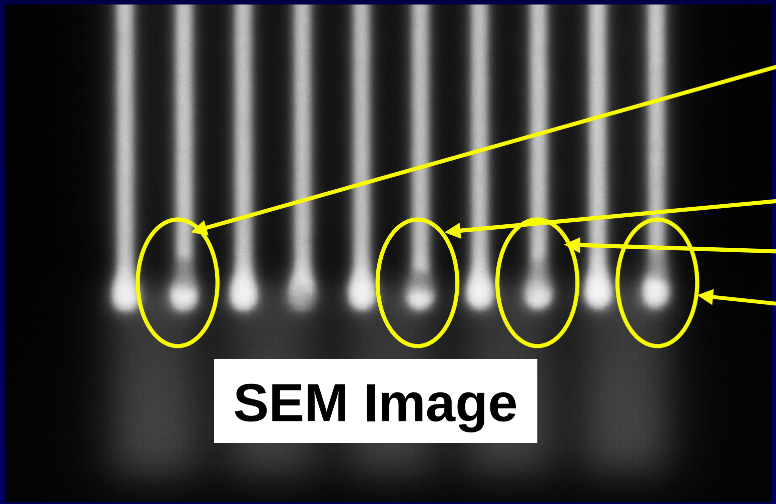
Scientific opportunities

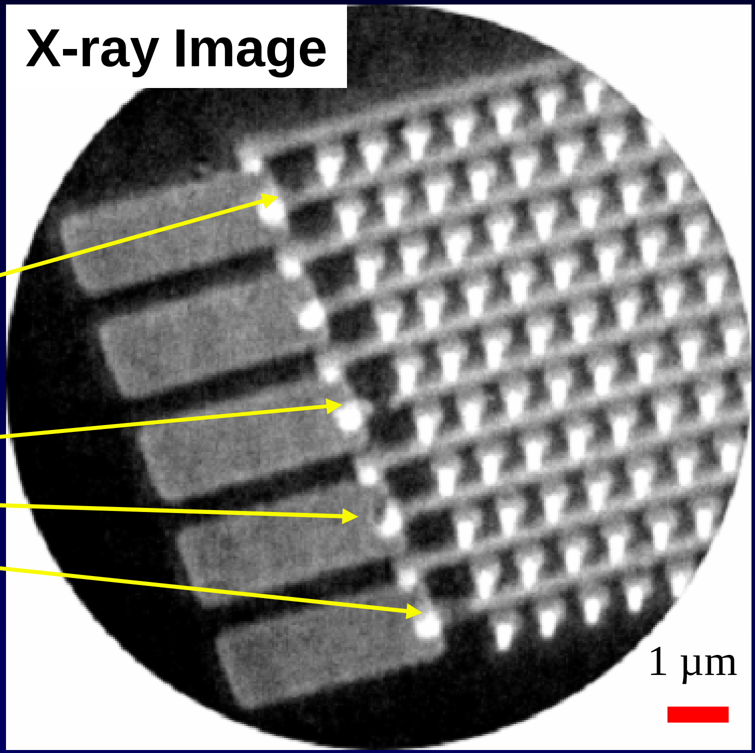
- **Nanotechnology: 3D imaging, structure and function dependence**
- **Semiconductor: electromigration, failure mechanism at small dimension**
- **Materials and engineering: crack initiation and propagation**
- **Geology, agriculture, and environmental science: imaging of soil sand stones in aqueous conditions**
- **Biology: imaging of cells and tissues in their natural state**
- **Biomedical: bones, implants, dental filling, etc.**

The X-ray Advantages



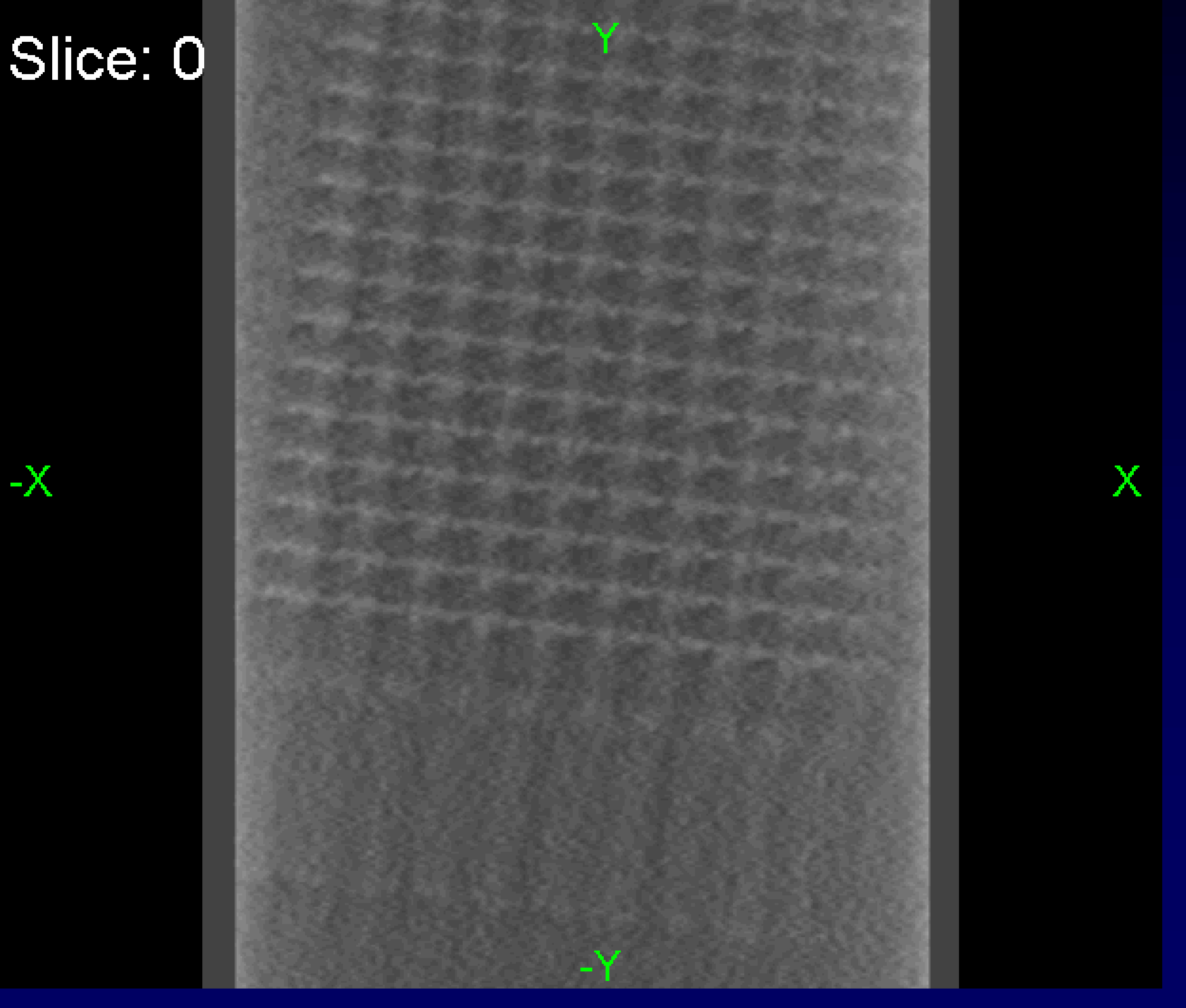
The EM (electromigration) structure consists of a set of 10 metal lines connected in a serpentine by short wide metal straps.



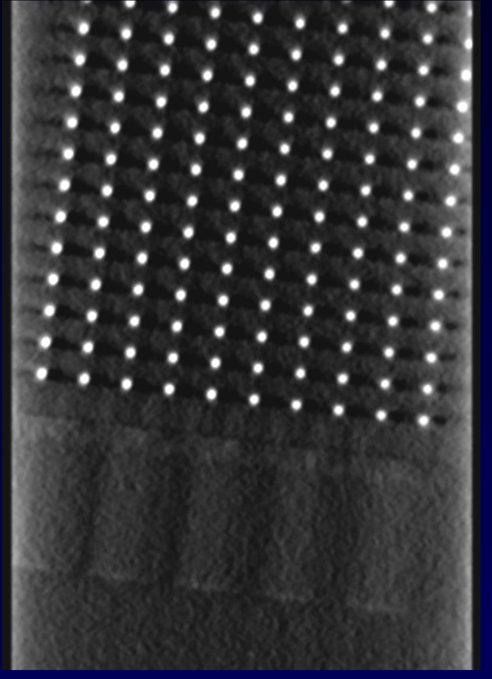


Defects in links 2, 6, 8 and 10 correlate well with the SEM image.
Indication of void at link 4 in x-ray image is not apparent in SEM image

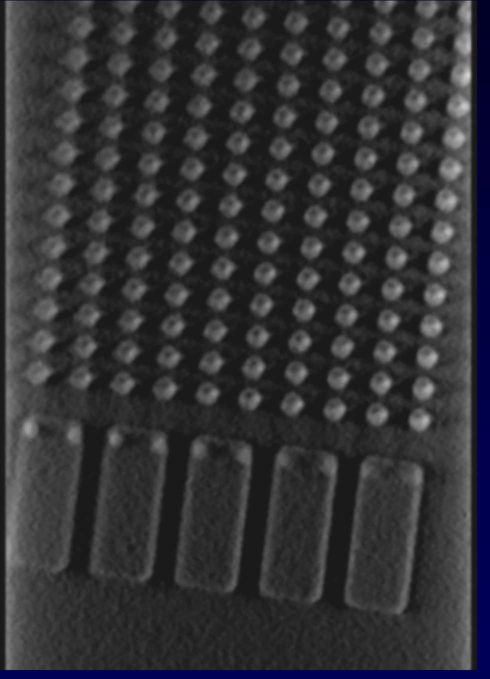
Electromigration Sample Reconstruction



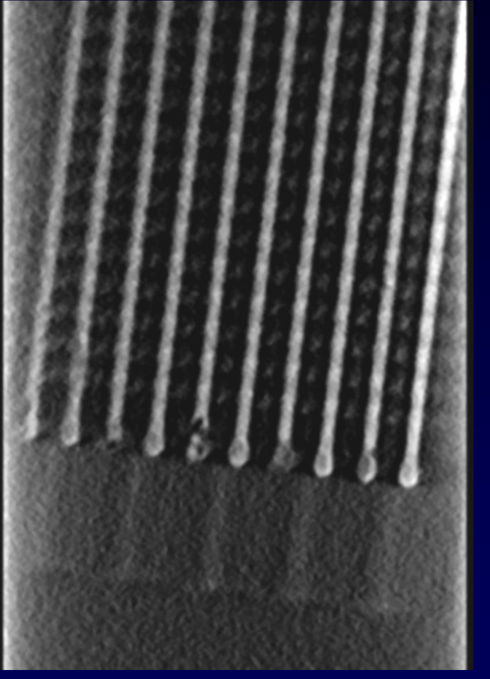
Reconstruction Slices



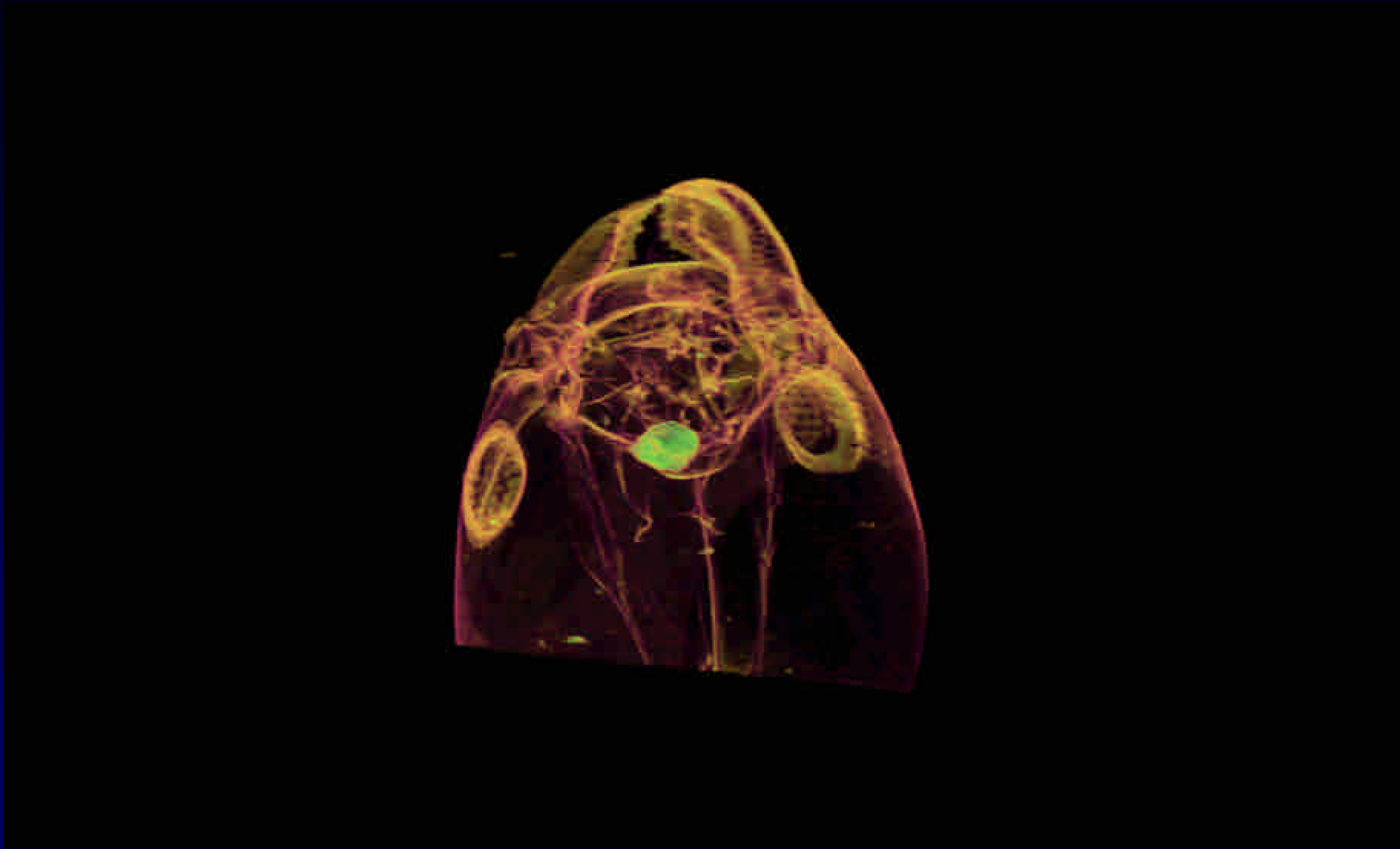
Heat sink layer



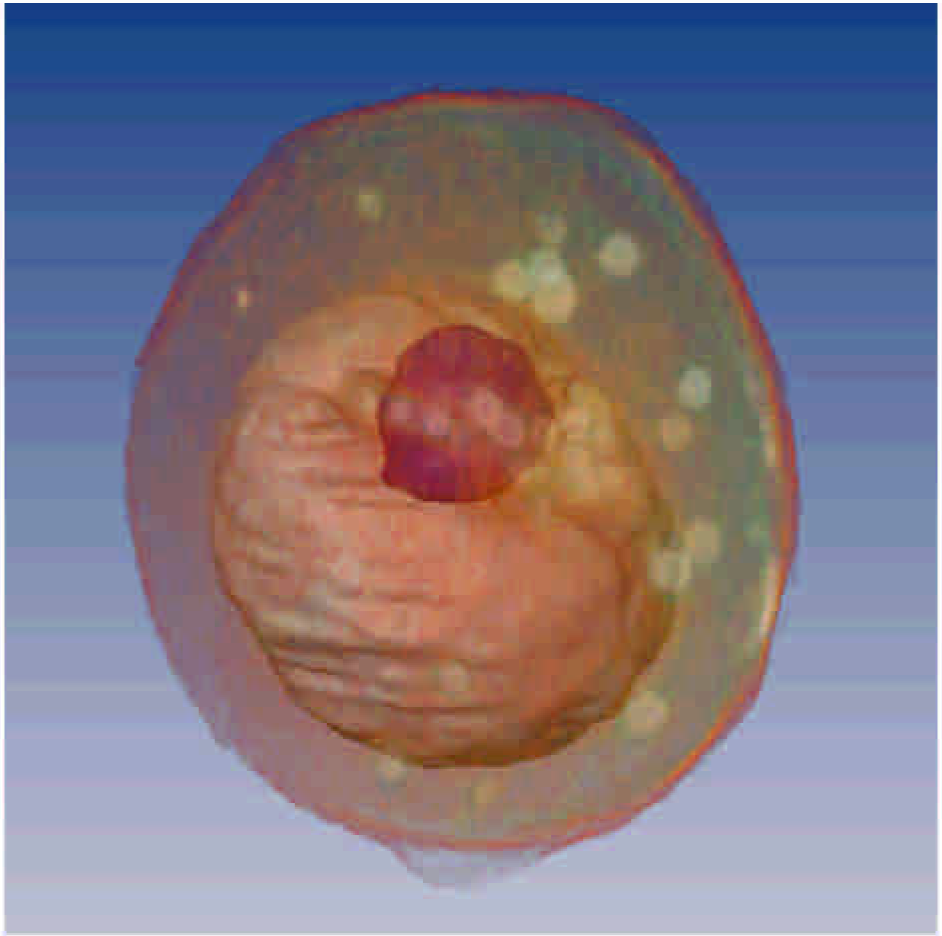
Pad layer



Line layer



Soft X-Ray Nano Tomography of a Yeast Cell



Courtesy of C. Larabell / UCSF & LBNL, and M. LeGros / LBNL

1. Synchrotron light

NSRRC is at the center of golden triangle.

Facilities

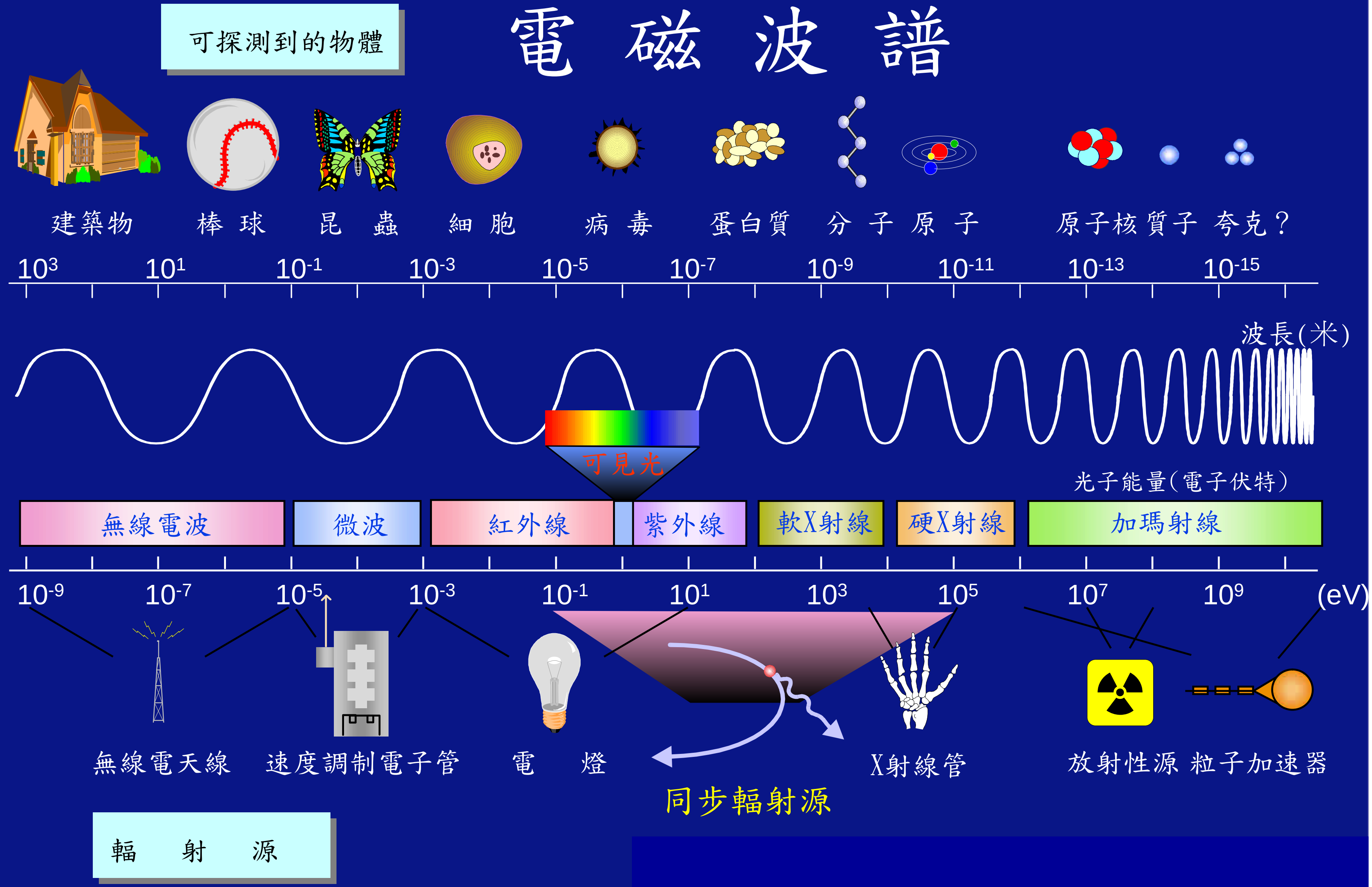
1 Main Gate
2 Administration Building
3 Research Building
4 Instrumentation Building
5 Booster Building
6 Storage Ring Building
7 Utility Building
8 Guest House

Neighboring Institutes

9 Nat'l Center for High Performance Computing
10 Nat'l Chiao Tung University

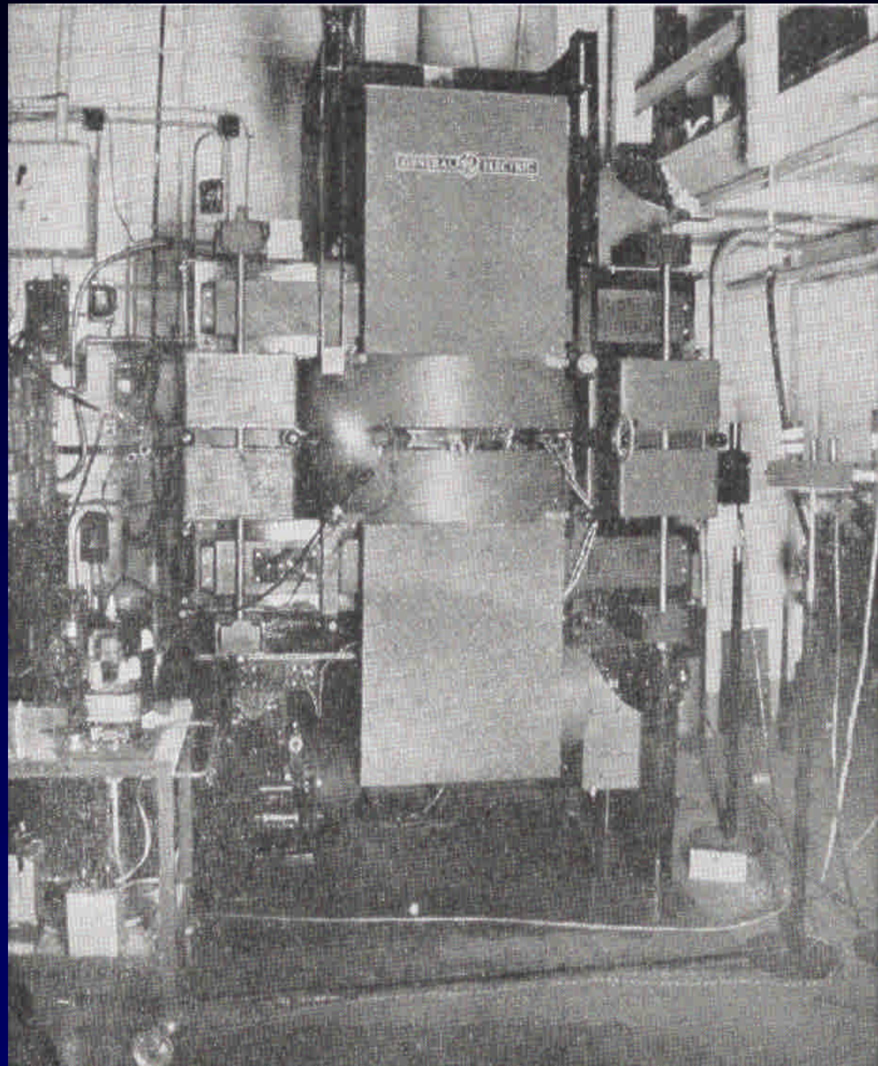
同步輻射是一種「光」。廣義地說，所有電磁波都可以叫做光。同步輻射也是電磁波的一份子，為一連續波段的電磁波，涵蓋紅外線、可見光、紫外線及X光等。

電磁波譜



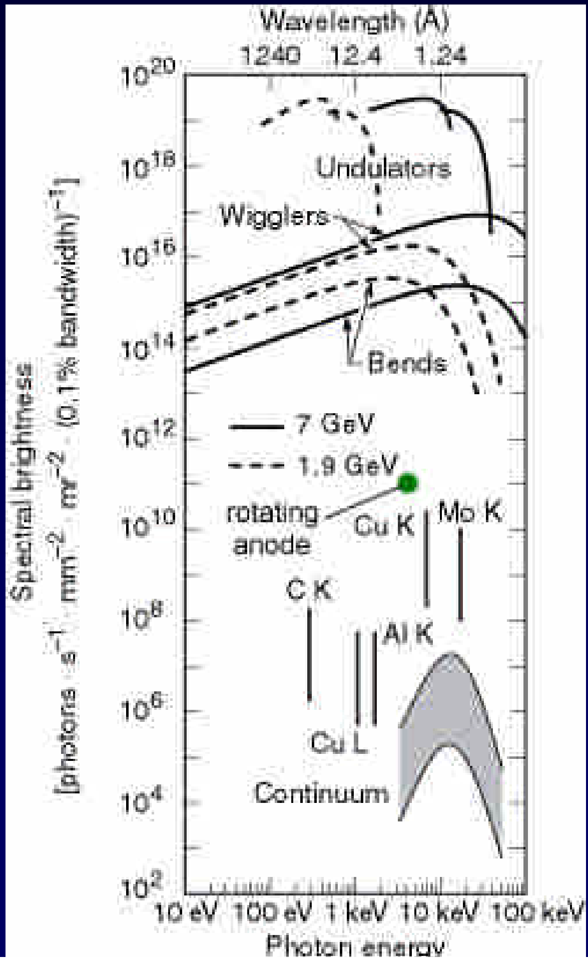
1947年首次在美國奇異公司同步加速器上意外地被發現，因此命名為「同步加速器輻射」，簡稱為「**同步輻射**」或「同步加速器光源」

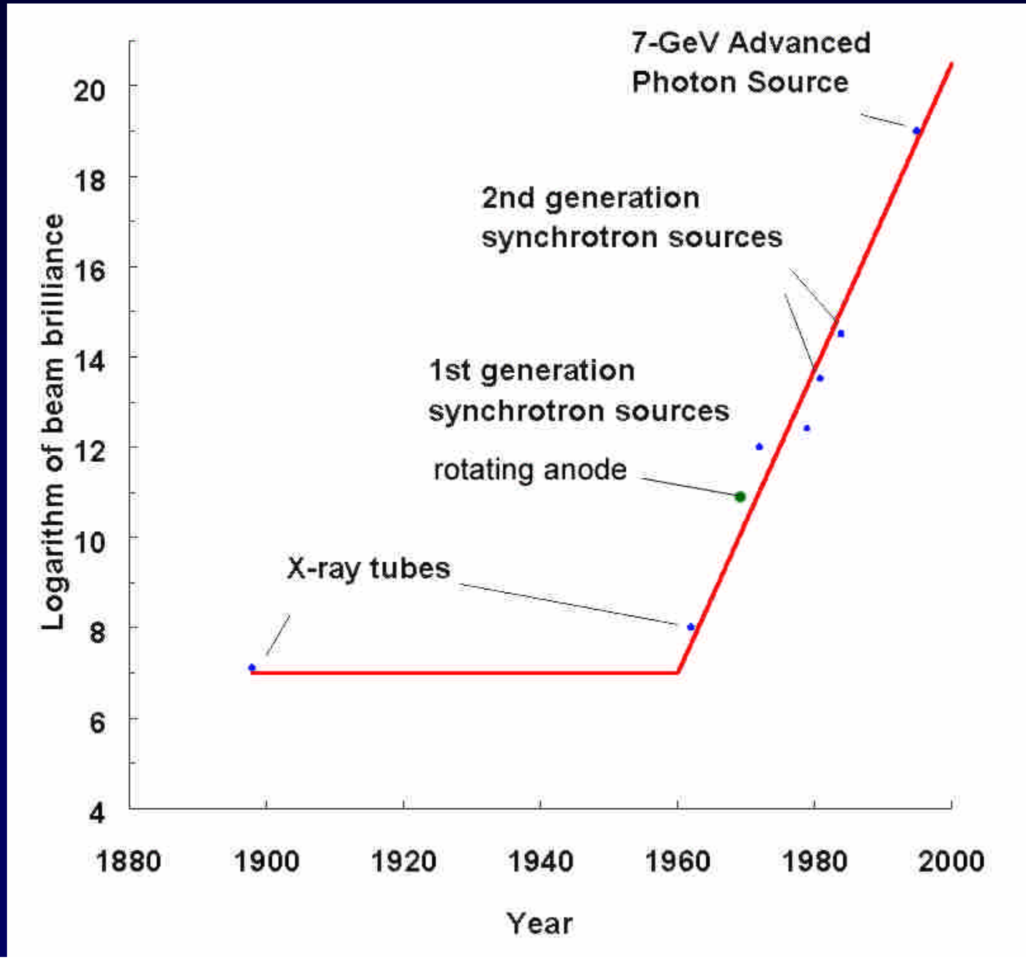
倫琴，1901年諾貝爾物理獎

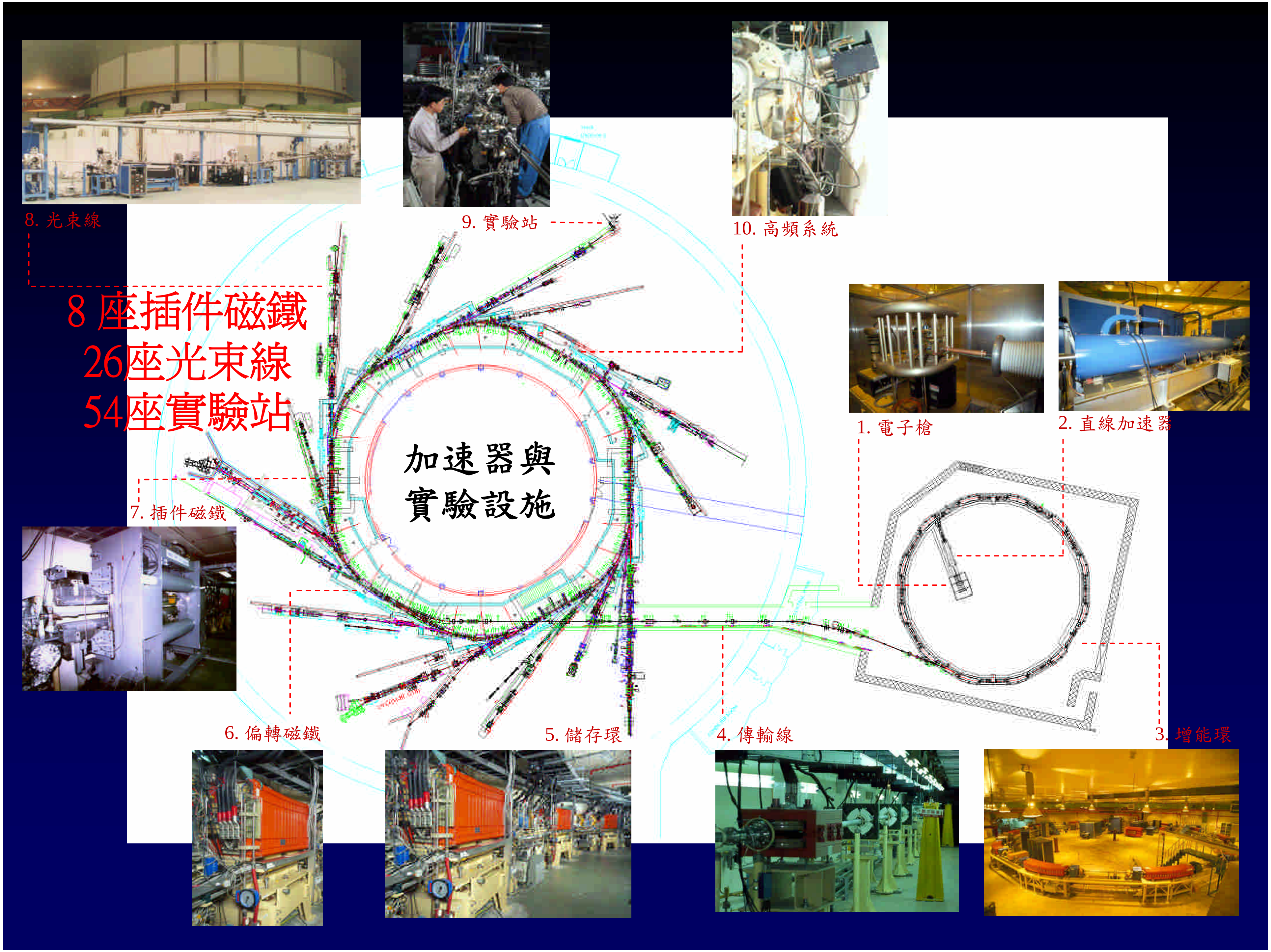


同步輻射的特色

- 高亮度
- 能量涵蓋廣，波長連續
- 準直性佳
- 光束截面積小
- 具有時間脈波性與偏振性

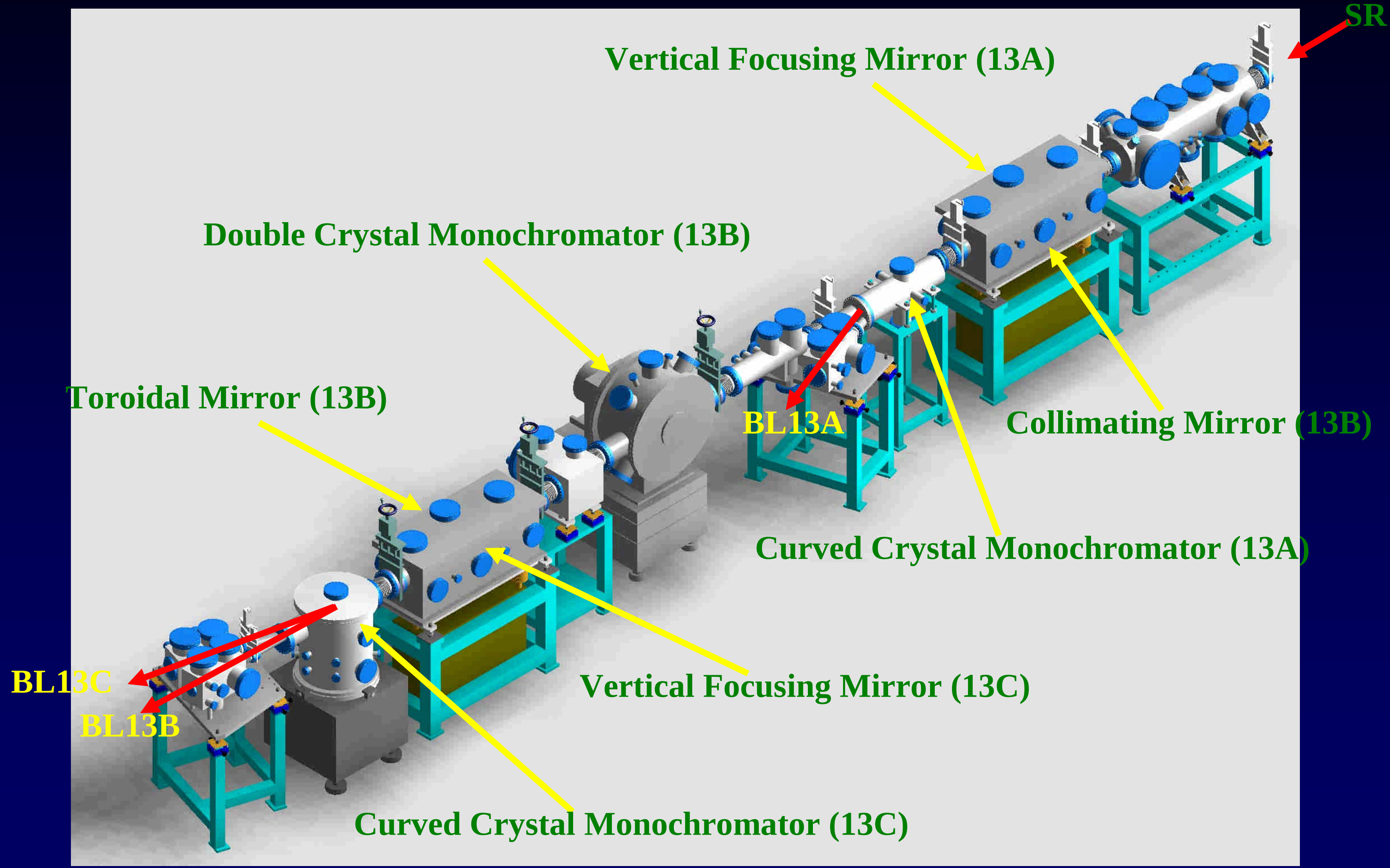






8 座插件磁鐵
26座光束線
54座實驗站
加速器與
實驗設施
1. 電子槍
2. 直線加速器
3. 增能環
4. 傳輸線
5. 儲存環
6. 偏轉磁鐵
7. 插件磁鐵
8. 光束線
9. 實驗站
10. 高頻系統

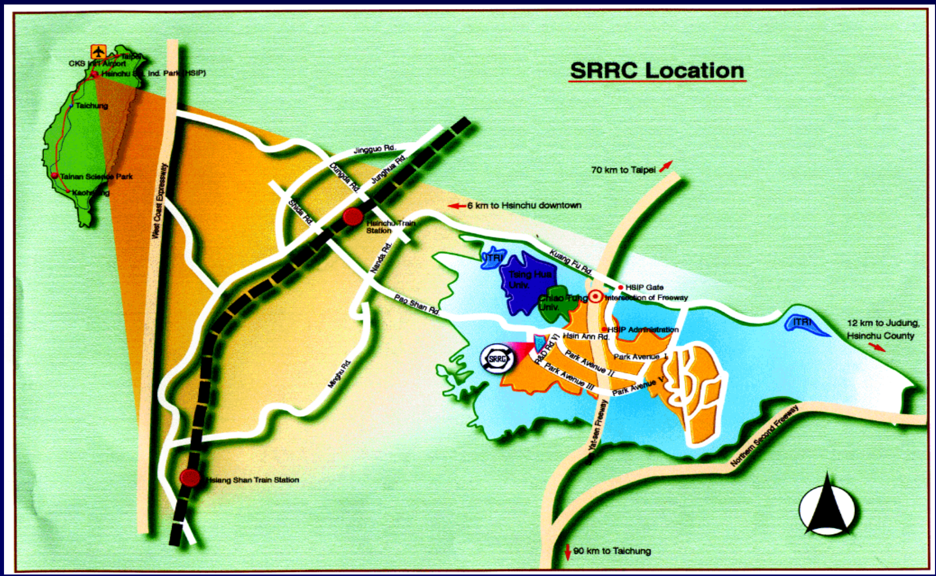
高效能生物結晶學專用光束線



Taiwan Beamlines @ SPring-8

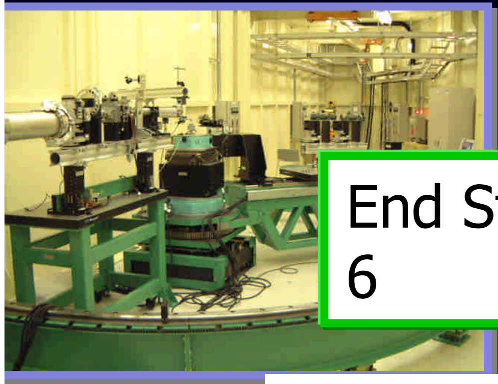
早晨，從中正機場出發，坐飛機，
路上不耽擱，傍晚前可以到達 SPring-8

距離:1600公里
時差:1小時
語言:日語或英語
文化:人民和善，親切，守法，有禮貌。
駕駛:左邊
工作時間:16小時/日 ，週休2日(科學家除外)



Proposal

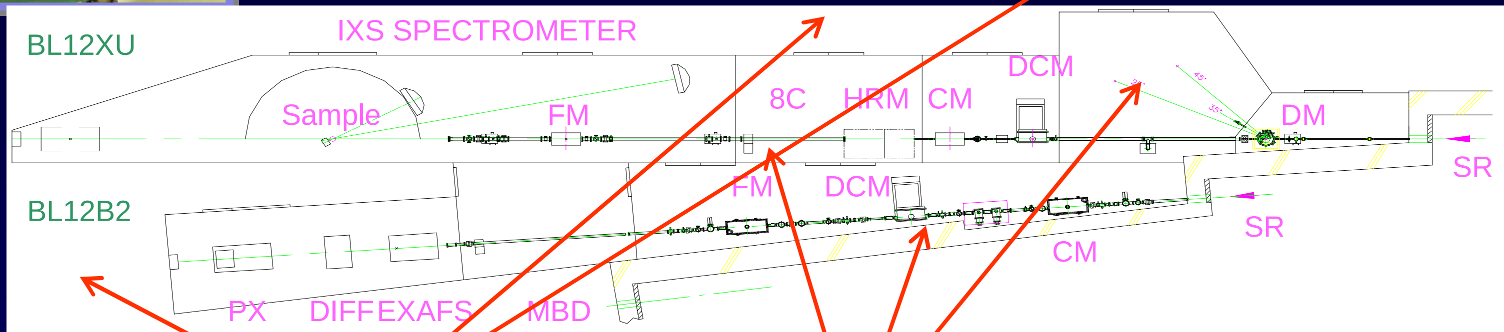
- Bending magnet beamline (Biostructure & Materials Research BL12B2)
- Insertion device beamline (Inelastic X-ray Scattering BL12XU)



End Stations x 6

Radiation Enclosures

Insertion Device x1

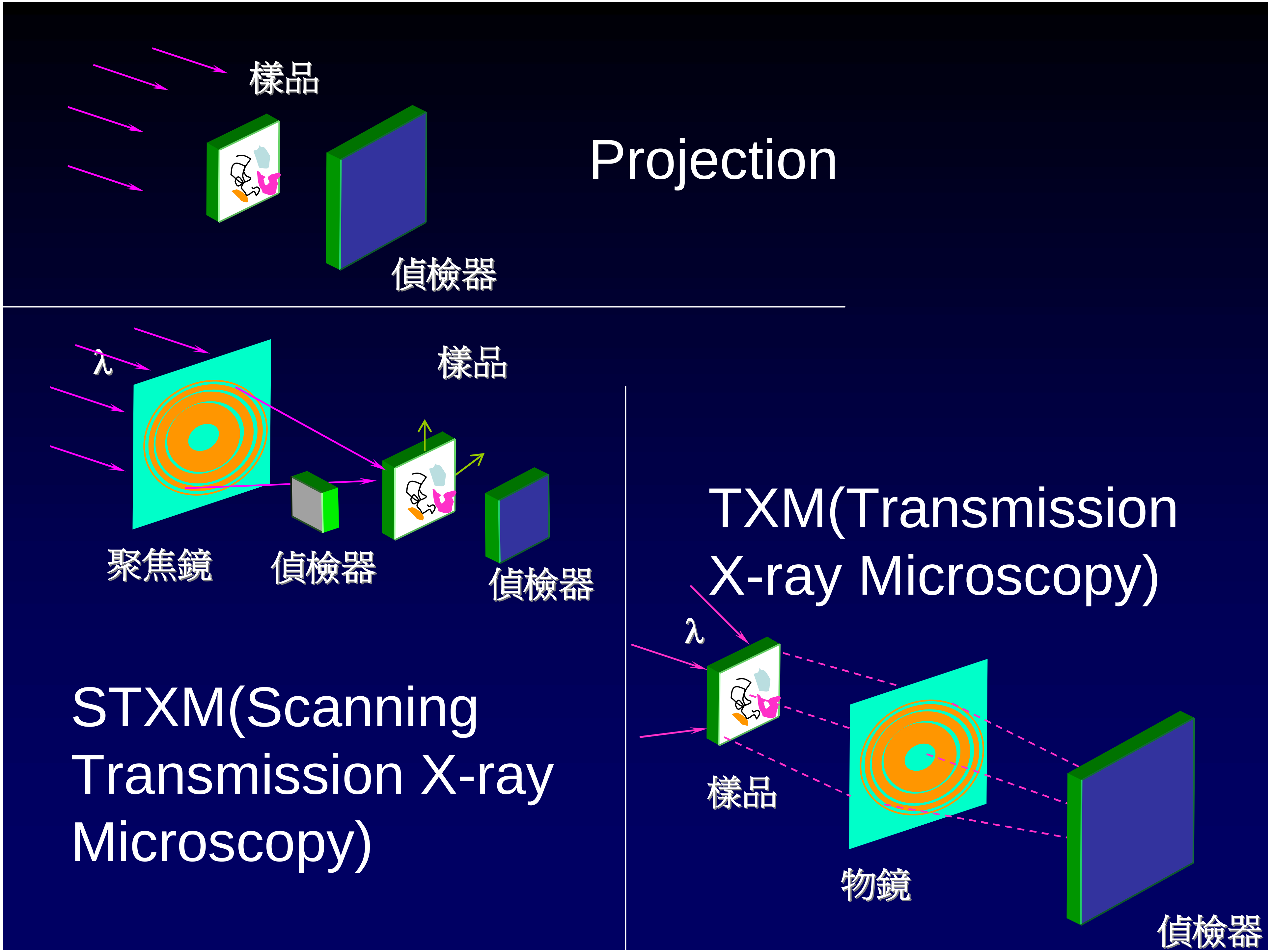


Front End x2

Experimental Areas x3

Beamline x2.5

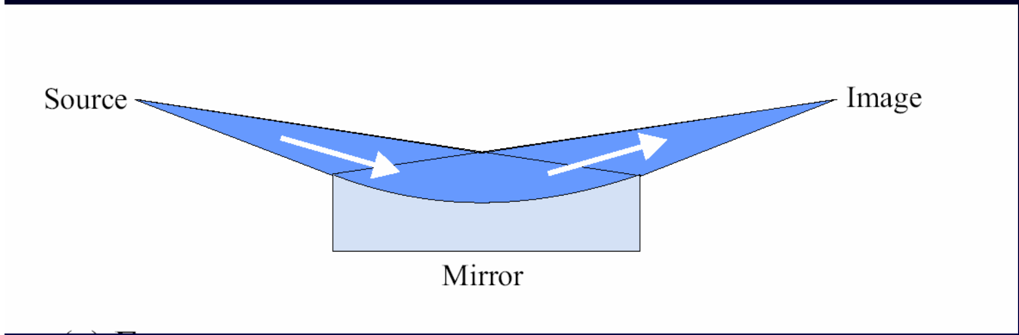
2. X-ray optical elements



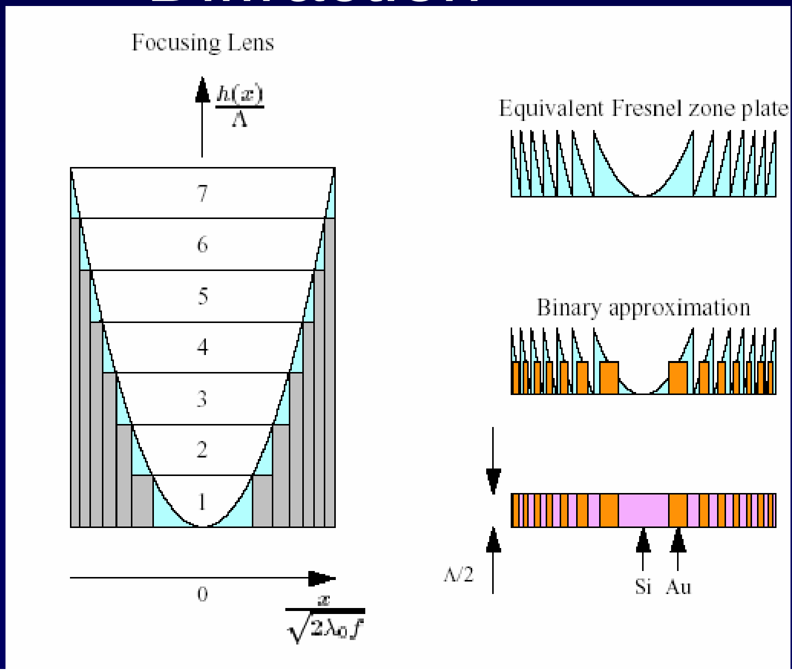
樣品
Projection
偵檢器
λ
樣品
聚焦鏡
偵檢器
偵檢器
STXM(Scanning Transmission X-ray Microscopy)
TXM(Transmission X-ray Microscopy)
λ
樣品
物鏡
偵檢器

Three types of X-ray focusing elements

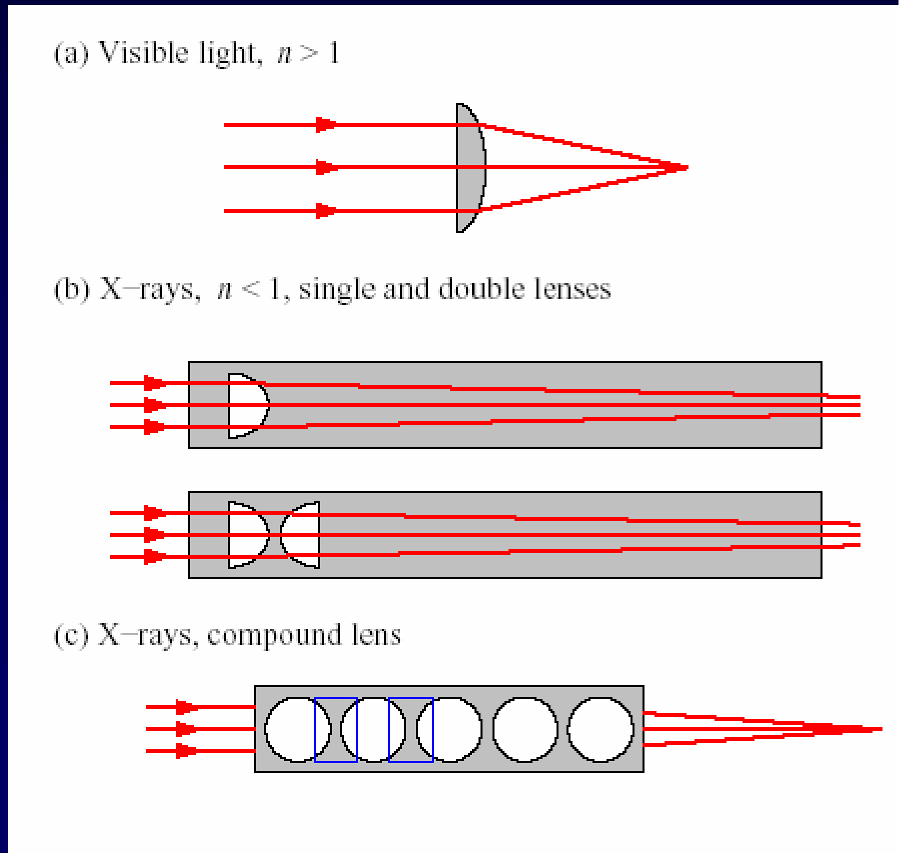
Reflection



Diffraction



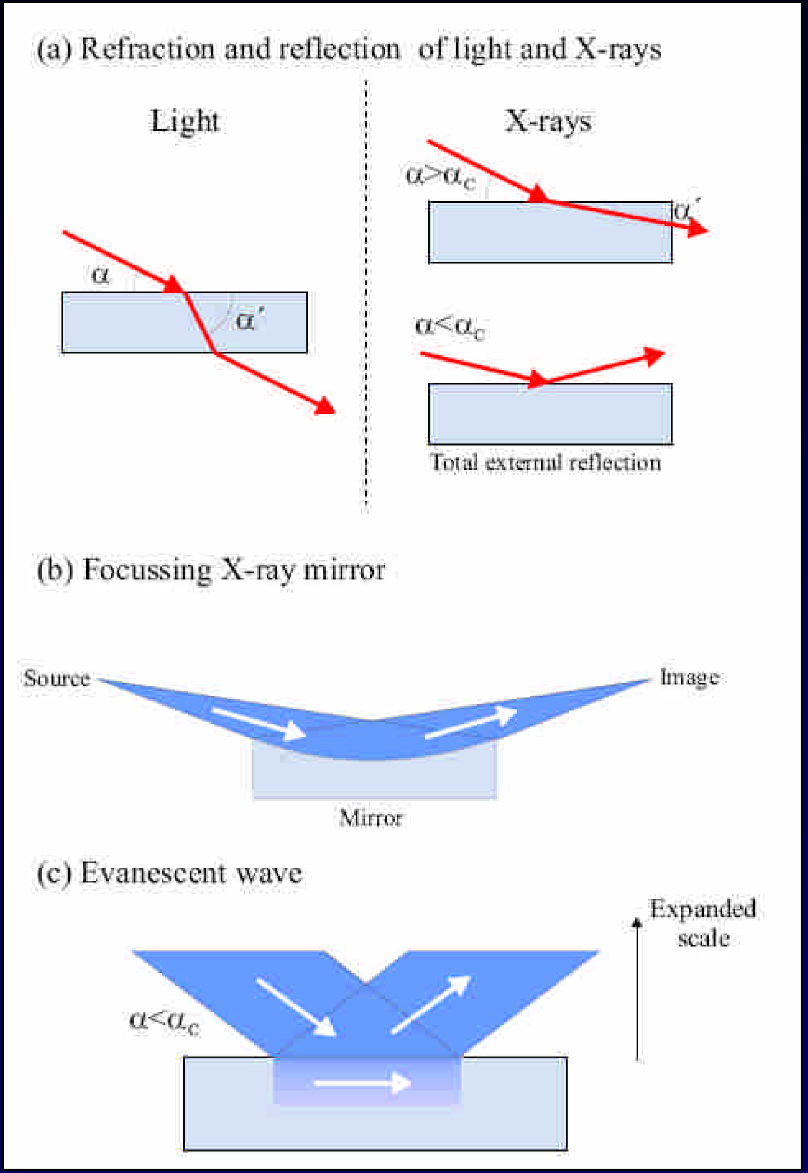
Refraction



Total reflection of x-rays

Refractive index of X – ray

$n = 1 - \delta + i\beta < 1$



Compound Refractive Lens (CRL)

$$\tilde{n} = 1 - \frac{N_a r_e}{2\pi} \lambda^2 (f_1 + i f_2) = 1 - \delta - i\beta < 1$$

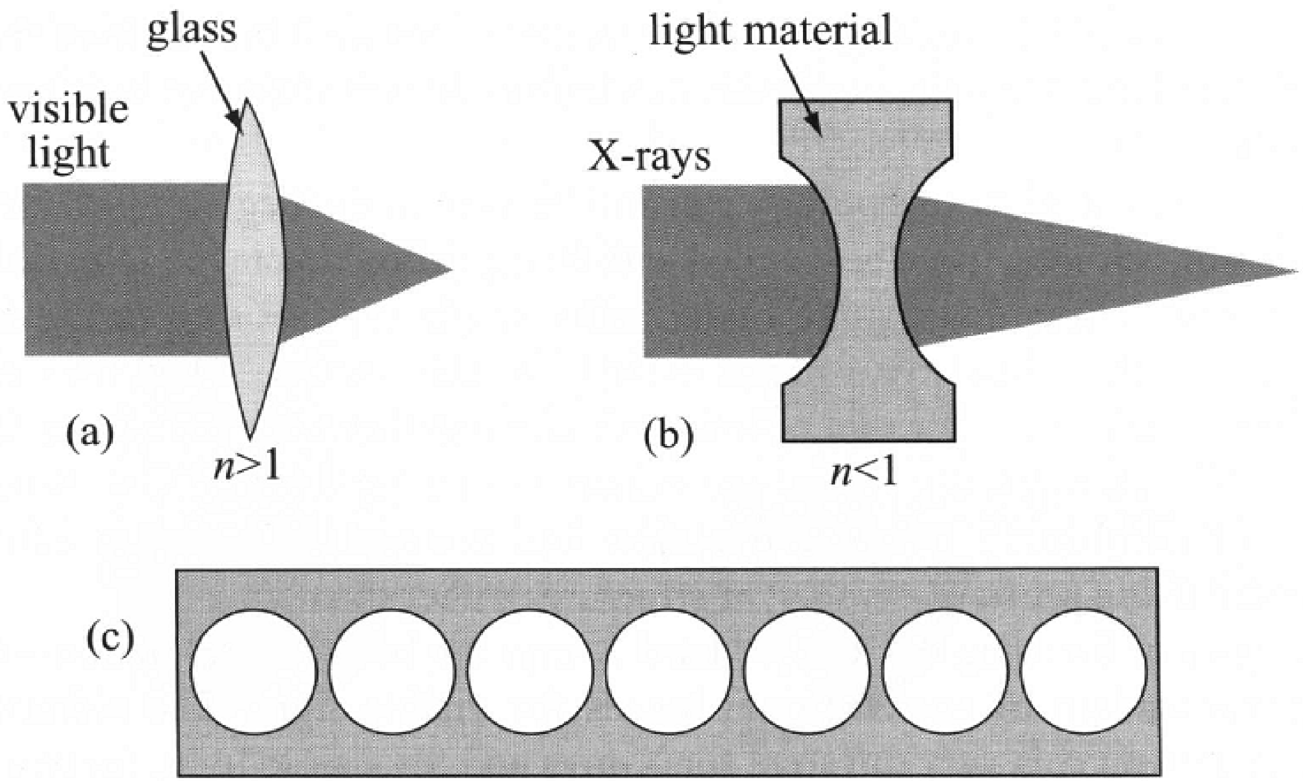
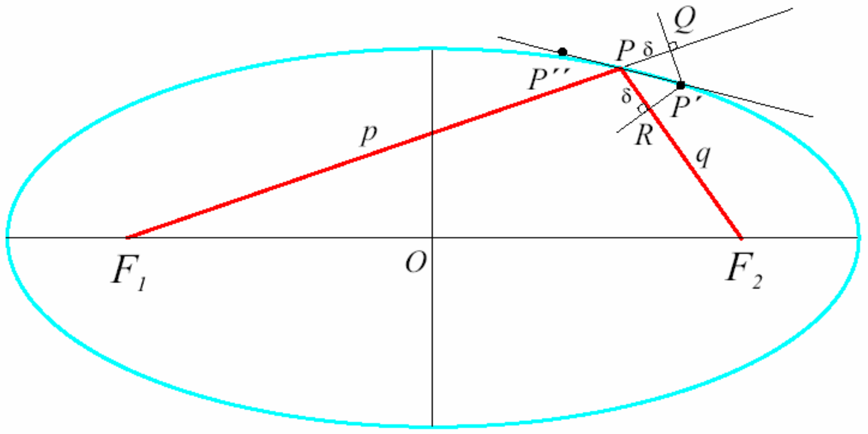


Fig. 3.74 (a) A typical biconvex glass (refractive) lens to focus visible light; (b) scheme of an idealized biconcave refractive lens for X-rays; (c) scheme of a more practical 'compound refractive lens' (CRL) for X-rays.



$$\frac{1}{p} + \frac{1}{q} = \frac{1}{f}$$

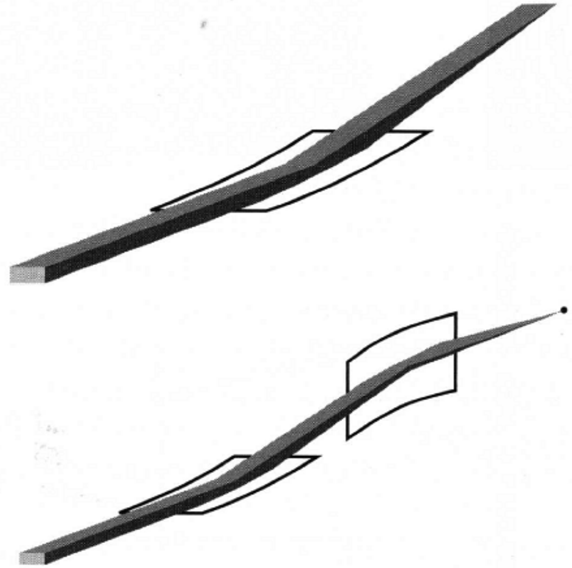
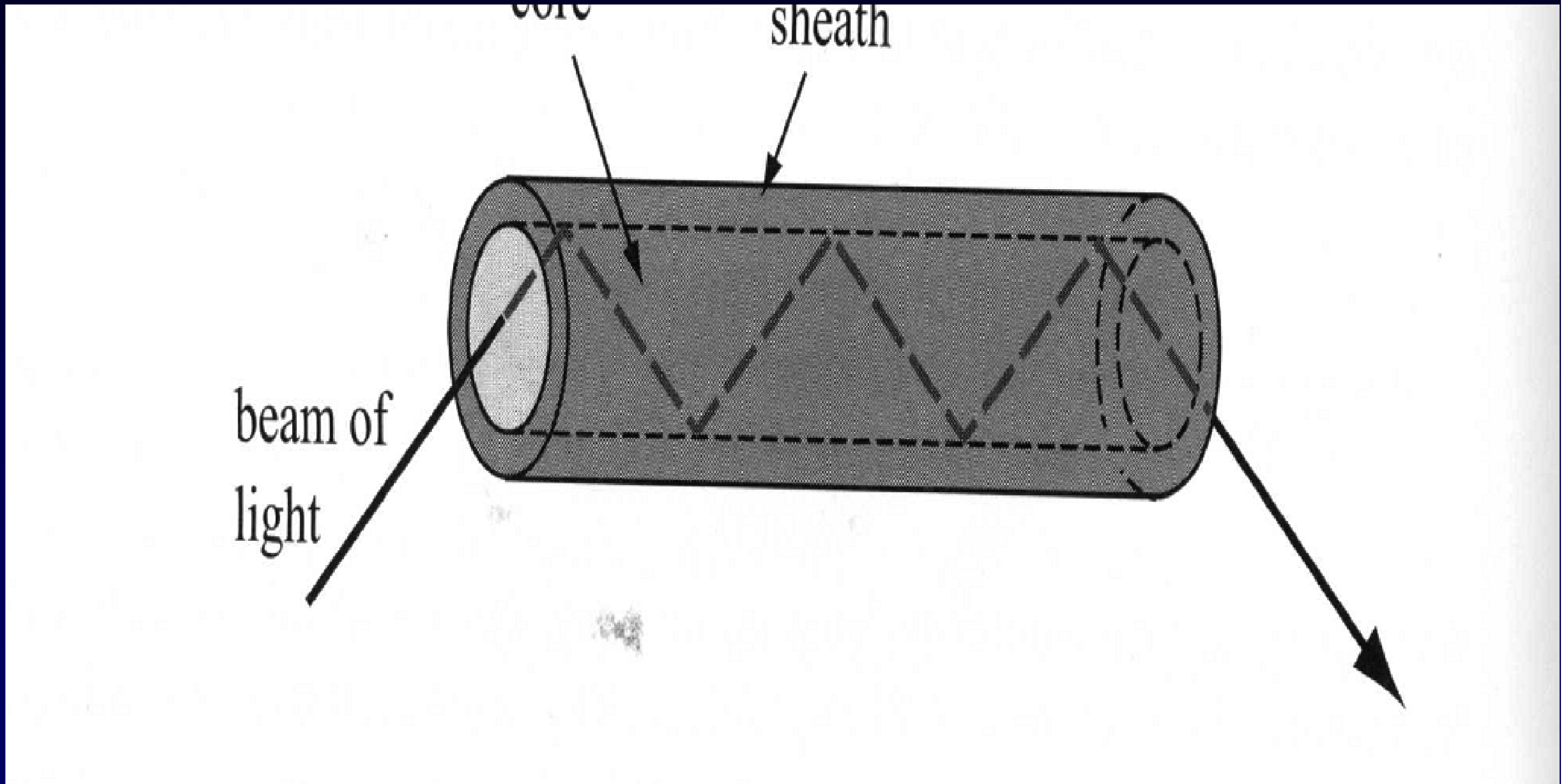


Fig. 3.69 Top: a cylindrical mirror (working at a grazing incidence) can focus a beam of X-rays only in one direction. Bottom: the combination of two perpendicular cylindrical mirrors pr focusing in two perpendicular directions. This is the device called the 'Kirkpatrick–Baez

X-ray mirrors

Taped capillary



Diffraction of x-rays by zone-plate

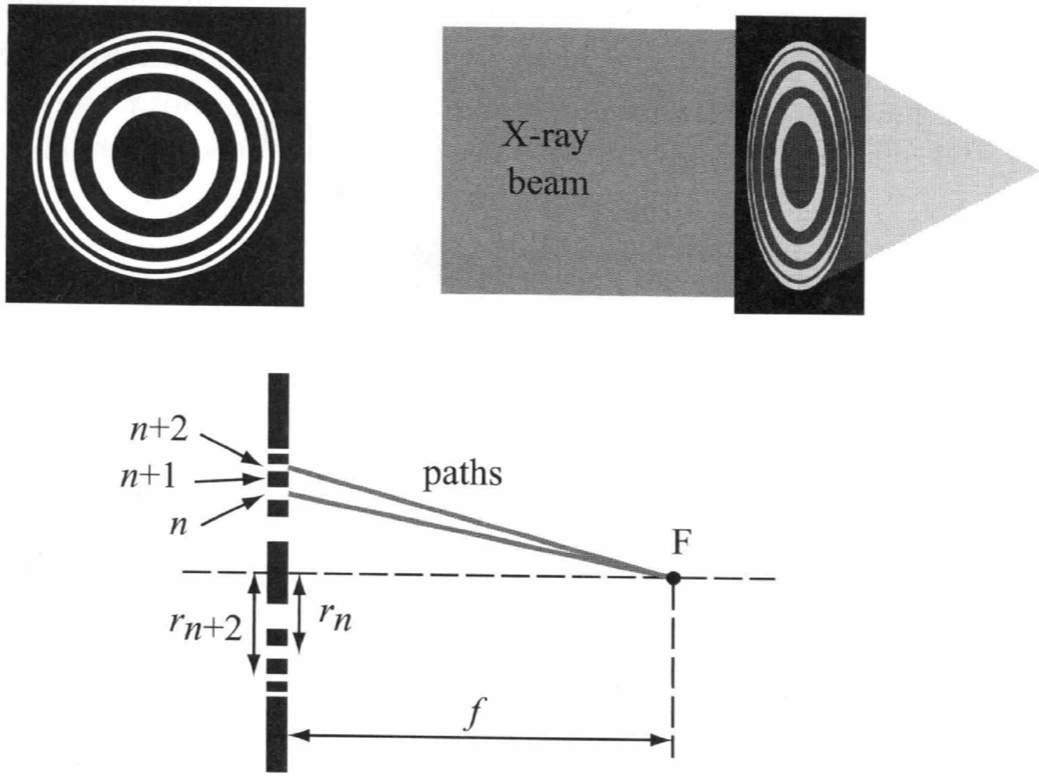


Fig. 3.70 Top left: schematic diagram of a zone plate (with only a very small number of zones shown). Top right: the zone plate acts as a focusing lens for X-rays. Bottom: analysis of the focal conditions (see text).

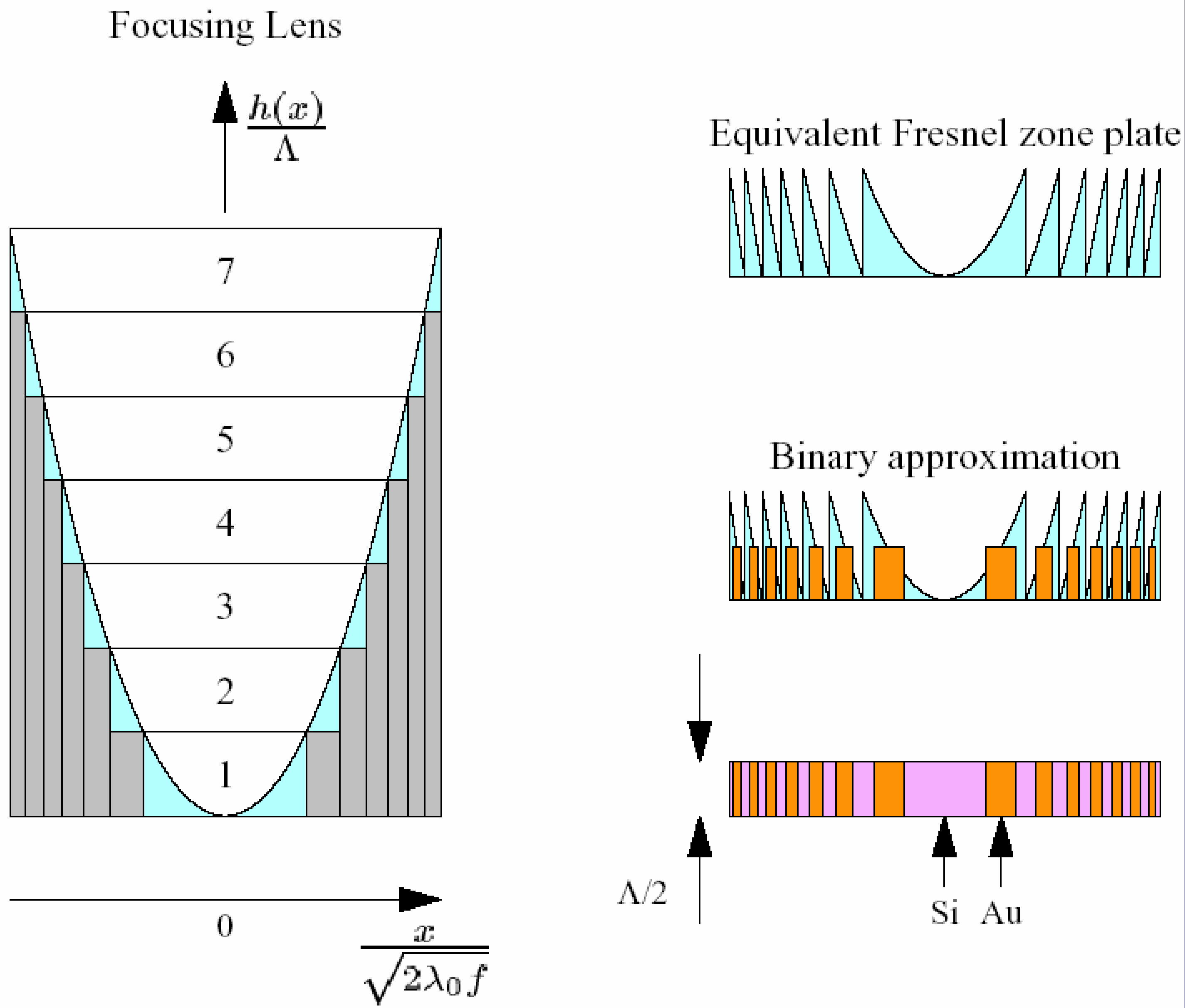

Focusing Lens
h(x)/Λ
7
6
5
4
3
2
1
0
x/√(2λ0f)
Equivalent Fresnel zone plate
Binary approximation
Λ/2
Si
Au

Image contrast

Absorption contrast

Elemental contrast
(water window)

Phase contrast

…...

Absorption of X-rays

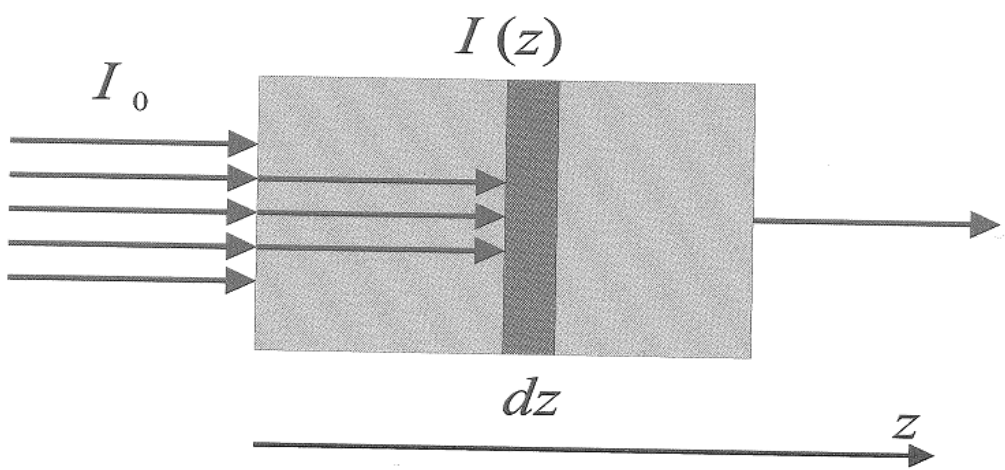
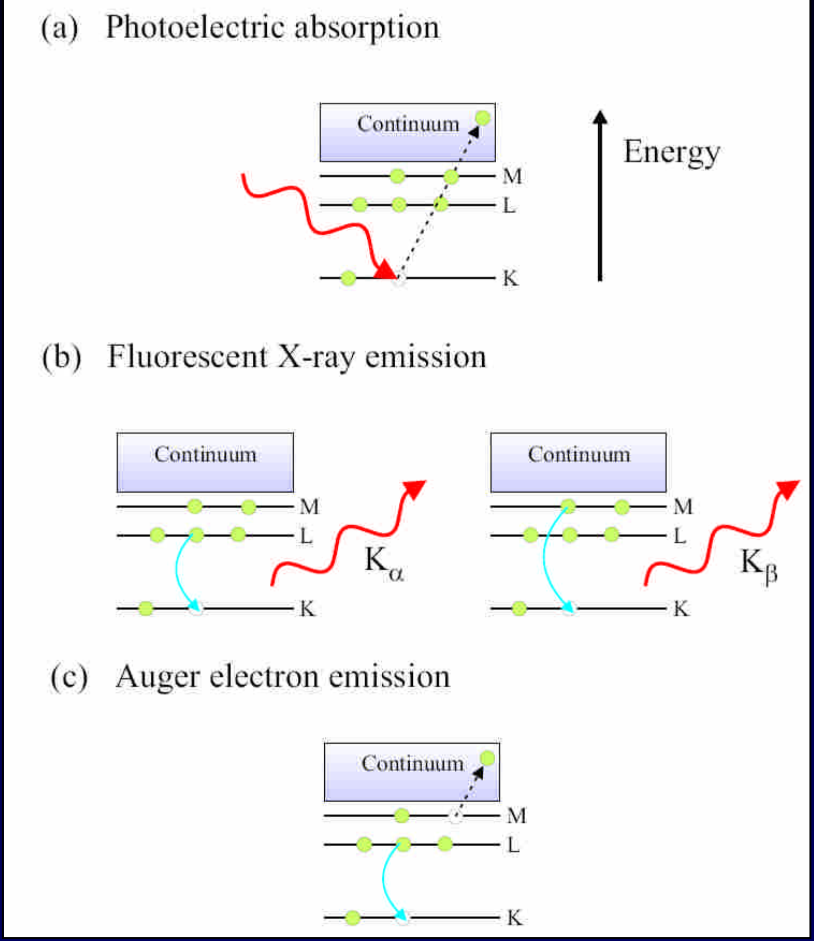
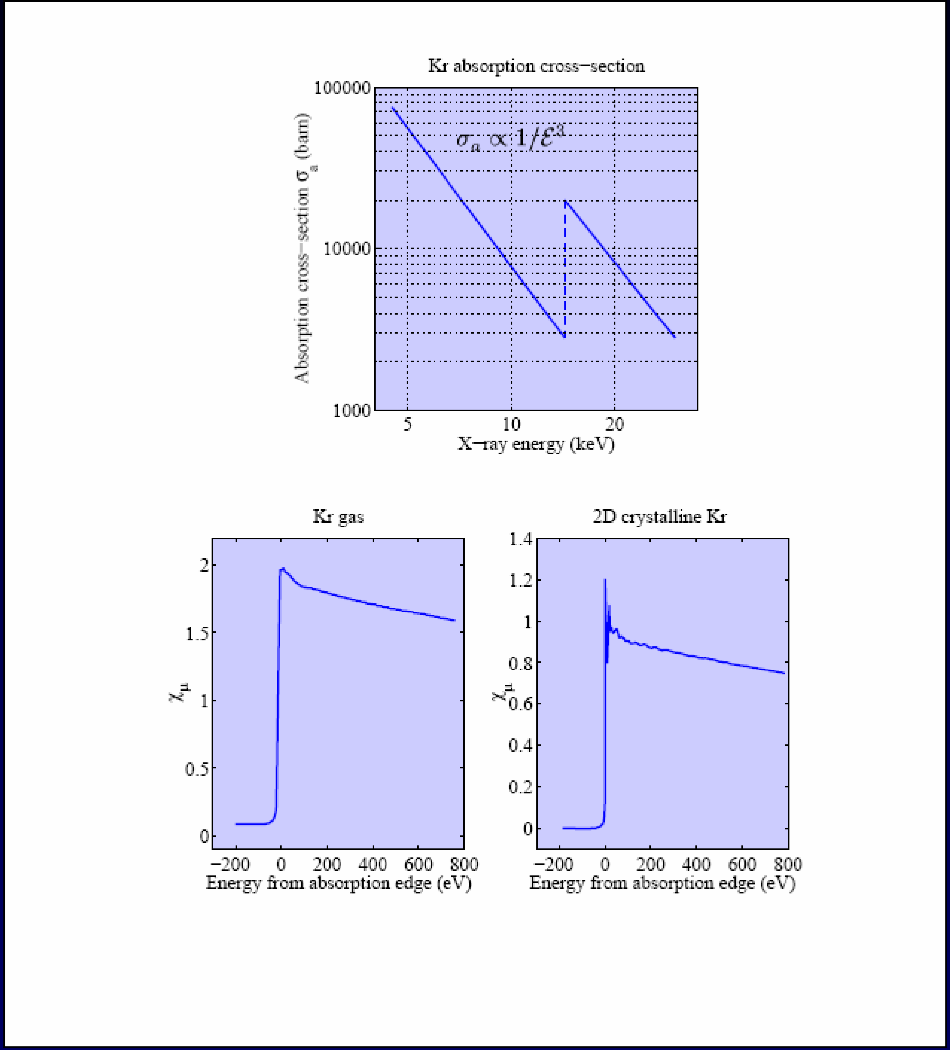


Figure 1.10: The attenuation of an X-ray beam through a sample due to absorption. attenuation follows an exponential decay with a characteristic linear attenuation length where μ is the absorption coefficient.

$$I(z) = I_o \exp(-\mu z)$$

Elemental Absorption





Water window for soft X-ray microscopy

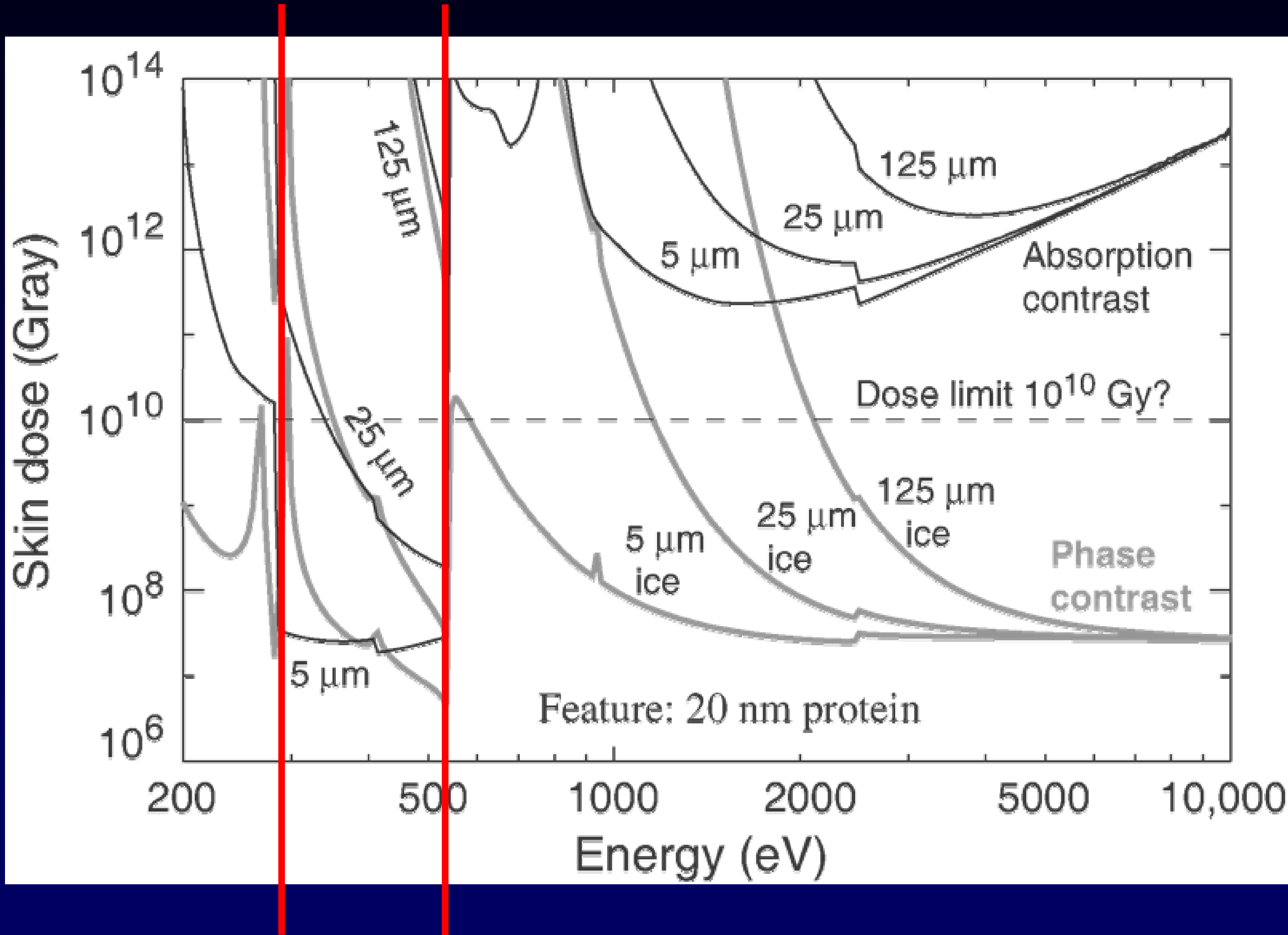


表 3.1　一些生物上重要物質的X射線吸收係數 *

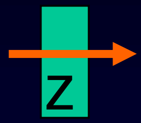
波長 (nm)	水	碳水化合物	蛋白質	類脂化合物	核酸	空氣
0.13	0.0006	0.0007	0.0006	0.0003	0.0014	0.0005
0.2	0.002	0.003	0.002	0.001	0.005	0.002
0.4	0.018	0.022	0.017	0.008	0.037	0.015
0.8	0.126	0.158	0.113	0.065	0.169	0.108
1.4	0.560	0.719	0.524	0.310	0.768	0.492
2.3	1.93	2.54	1.88	1.13	2.71	1.74
O　吸收邊						
2.4	0.112	1.02	1.38	1.00	1.36	1.46
2.7	0.151	1.36	1.84	1.35	1.82	1.95
3.0	0.198	1.77	2.38	1.74	2.34	2.50
N　吸收邊						
3.1	0.215	1.92	1.90	1.89	1.74	0.172
3.7	0.336	2.92	2.88	2.87	2.63	0.277
4.2	0.458	3.91	3.85	3.84	3.50	0.369
C　吸收邊						
4.4	0.521	0.624	0.522	0.245	1.19	0.415
5.2	0.782	0.941	0.769	0.372	1.69	0.622
7.4	1.84	2.20	1.69	0.872	3.26	1.45
[illegible]	2.80	3.45	2.38	1.36	4.76	2.28

Phase Contrast

Refraction index : $n = 1-\delta-i\beta$

$E(z)=E_0 e^{-i2\pi knz} = E_0 e^{i2\pi\delta z/\lambda \, -2\pi\beta z/\lambda}$

$I(z) \sim |E(z)|^2 \sim I_0 e^{4\pi\beta z/\lambda}$

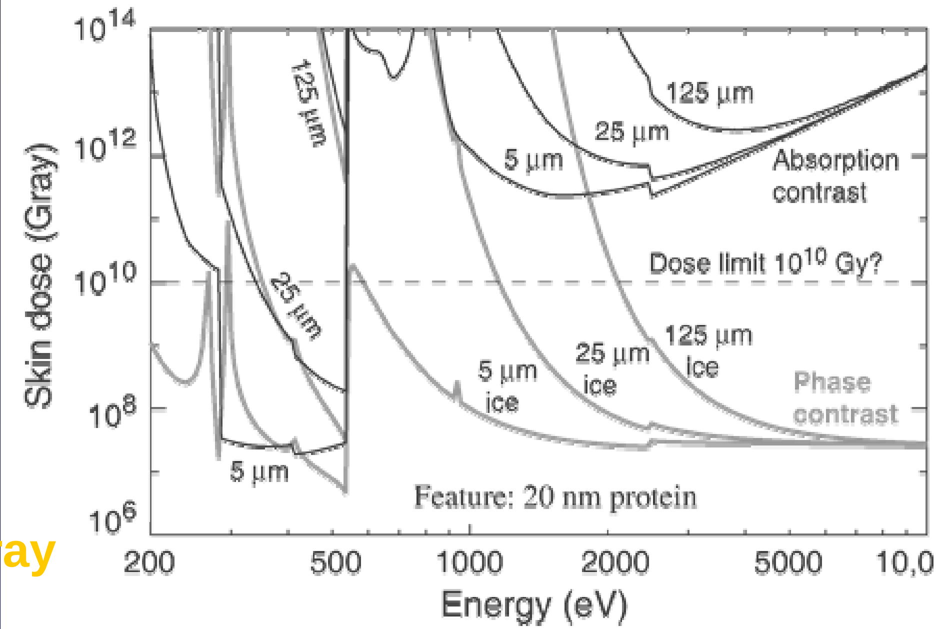


Absorption contrast

$\mu z = 4\pi\beta z/\lambda \sim \lambda^3$

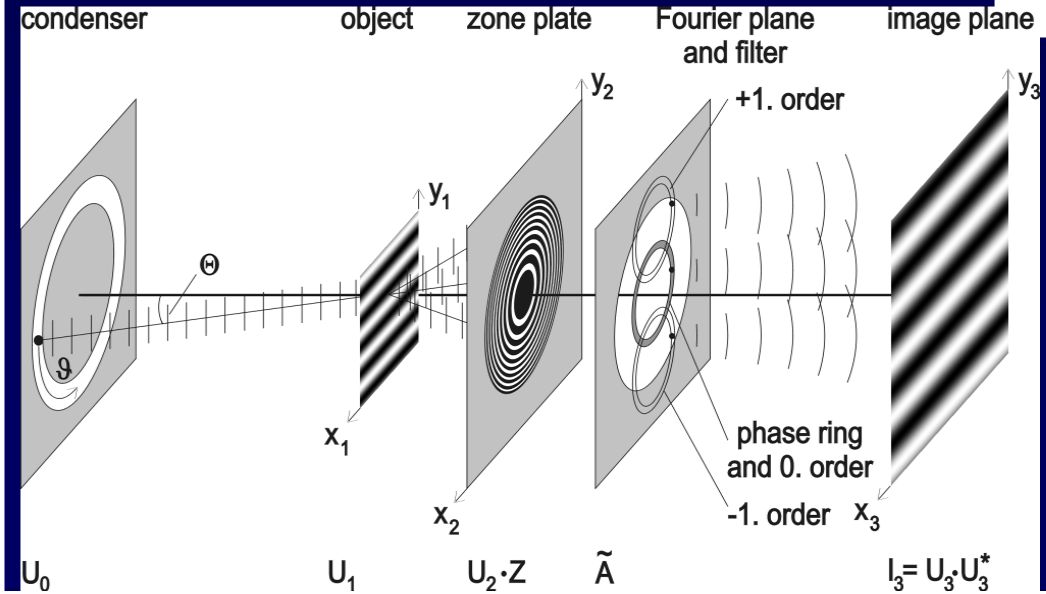
Phase contrast

$\phi(z) = 2\pi\delta z/\lambda \sim \lambda$



Schematic of Zernike phase contrast in X-ray

G. Schneider (1998)



Courtesy of C. Jacobsen