

Today's class:

X-ray Crystallography

This lecture follows the materials from the following book

- *Spectroscopy for the Biological Sciences, by GG Hammes, Wiley, 2005*
- *Physical Chemistry for Life Sciences, by PW Atkins and JD Paula, Oxford, 2006*

What is x-ray?



Wilhelm Röntgen, 1895

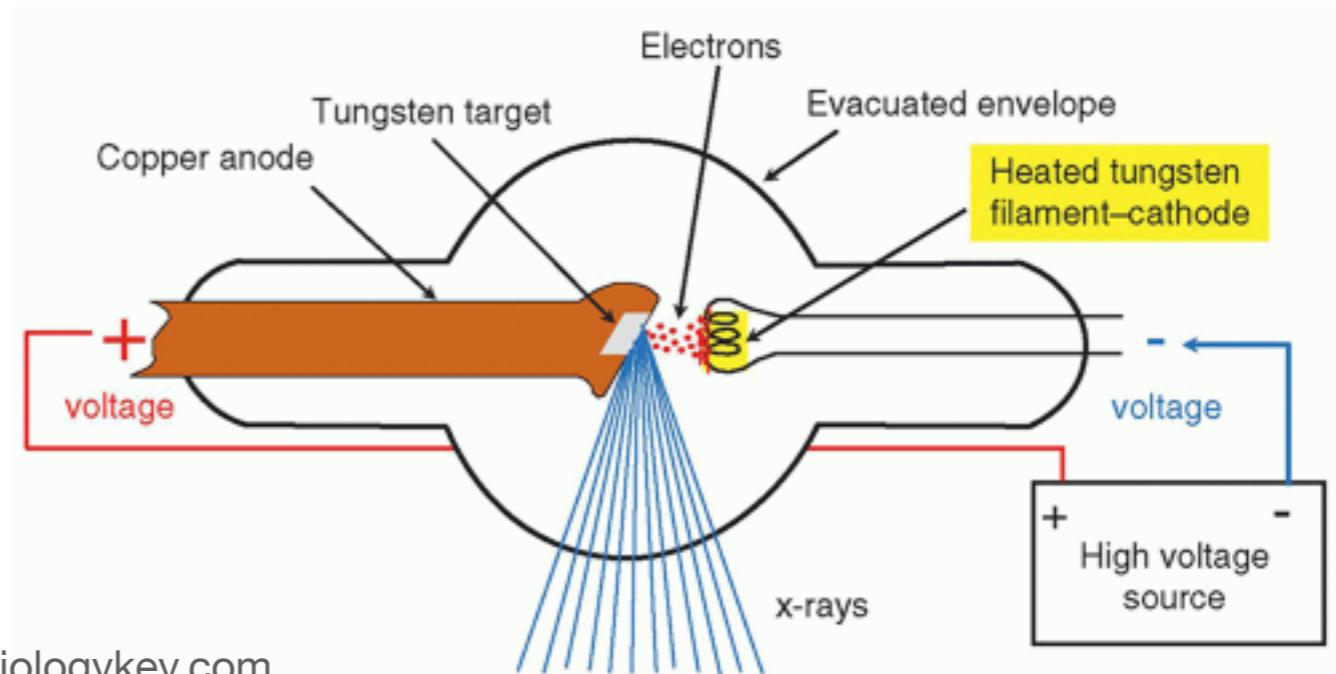


Image courtesy: radiologykey.com

Types of x-ray

Bremsstrahlung x-ray

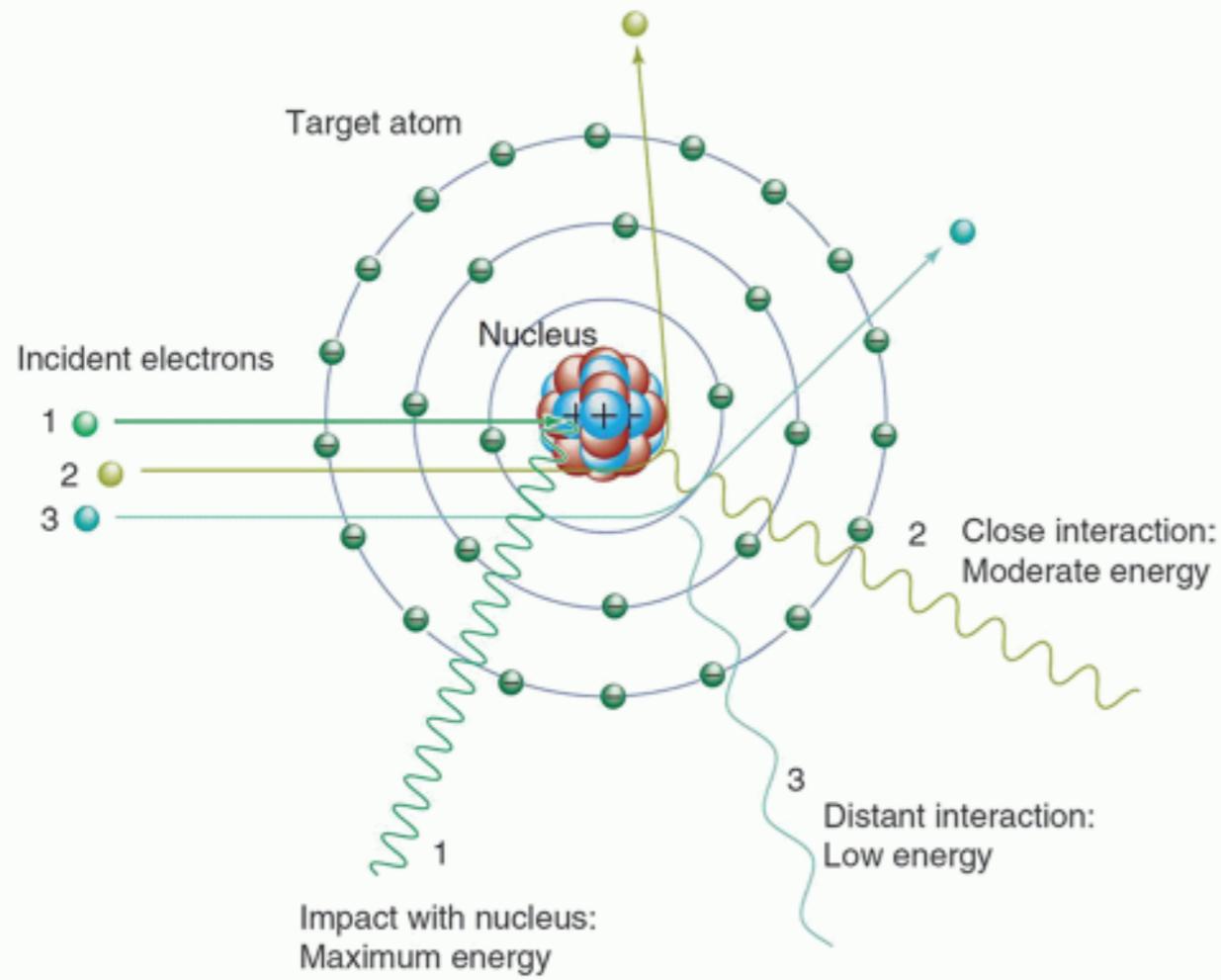
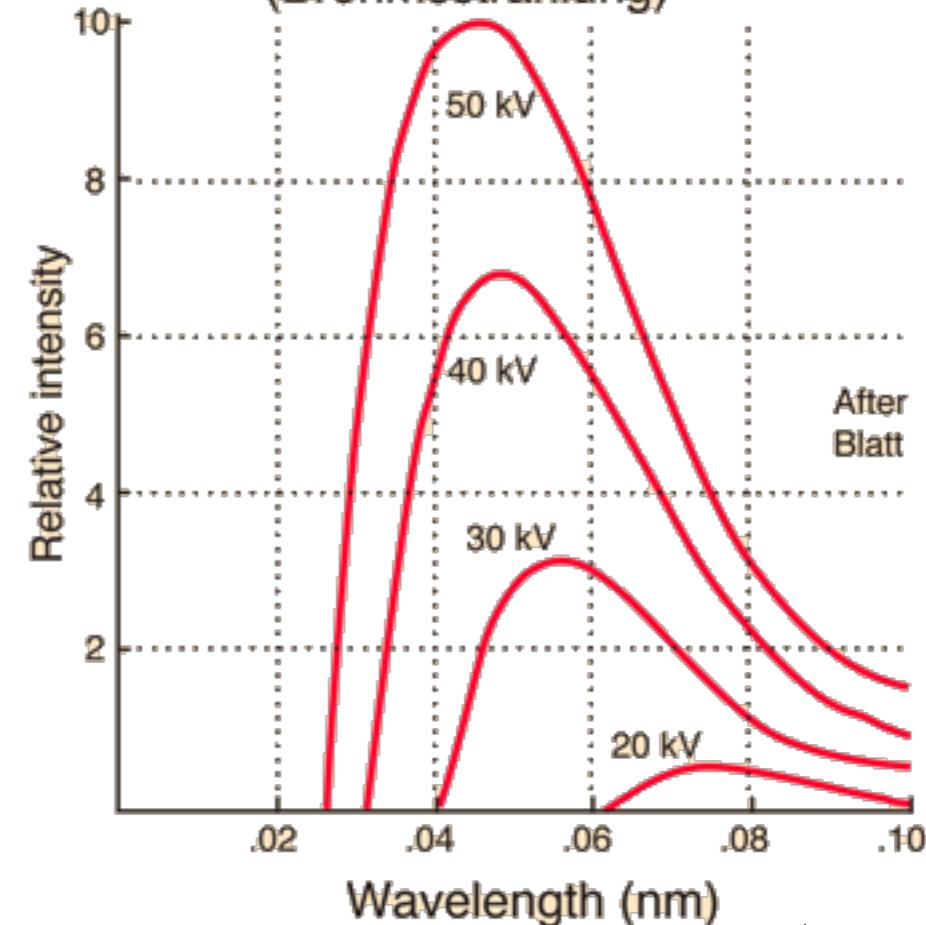


Image courtesy: radiologykey.com

X-ray Continuum Radiation (Bremsstrahlung)



Data source: <http://hyperphysics.phy-astr.gsu.edu/hbase/quantum/xrayc.html>

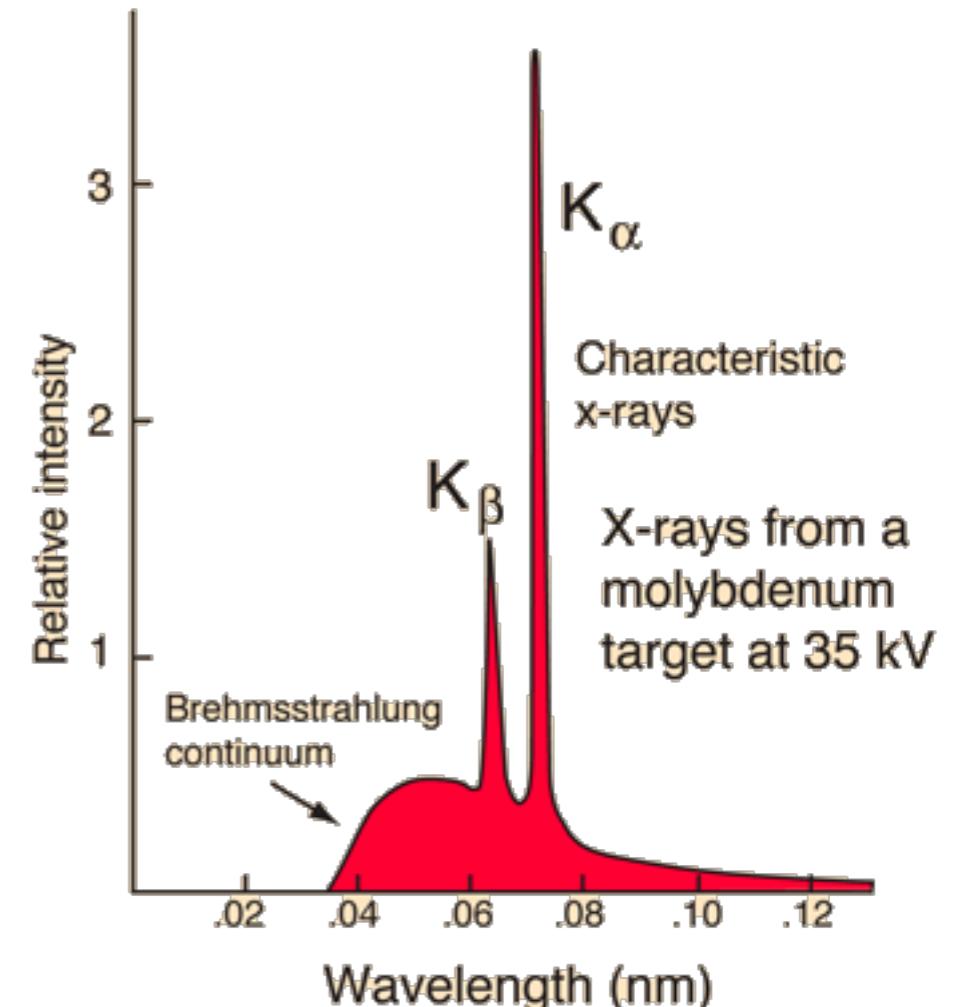
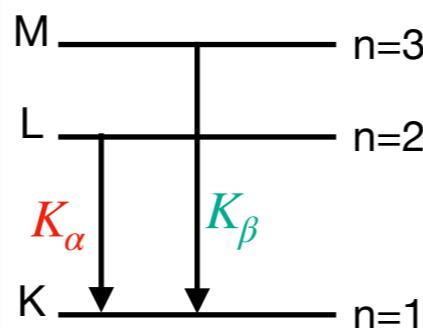
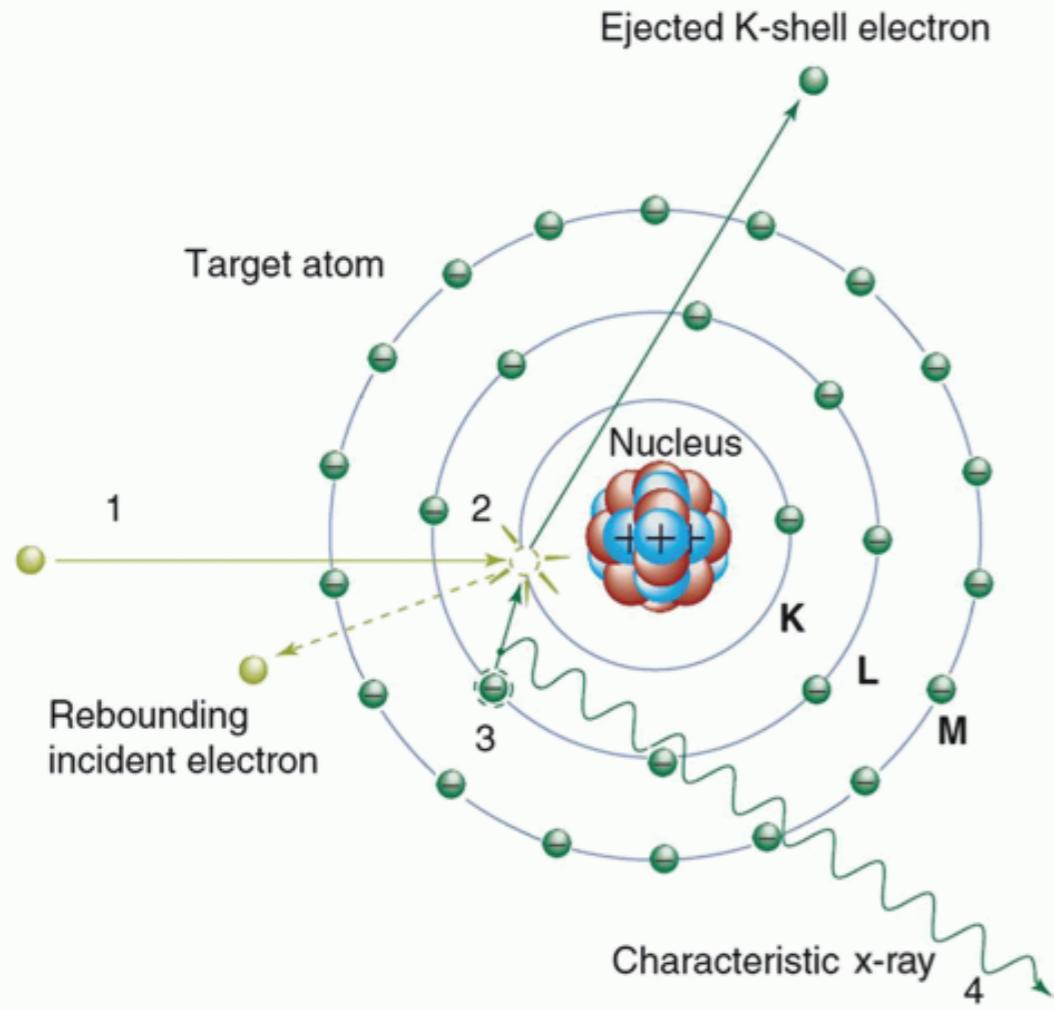
Maximum energy of x-ray photon goes up as potential energy goes up

$$E_{max} = e \times V = \frac{hc}{\lambda_{min}}$$

Min wavelength shifts towards left

Types of x-ray...*contd*

Characteristic x-ray



Data source: <http://hyperphysics.phy-astr.gsu.edu/hbase/quantum/xrayc.html>

Image courtesy: radiologykey.com

How can such radiations help us understand molecular structures?

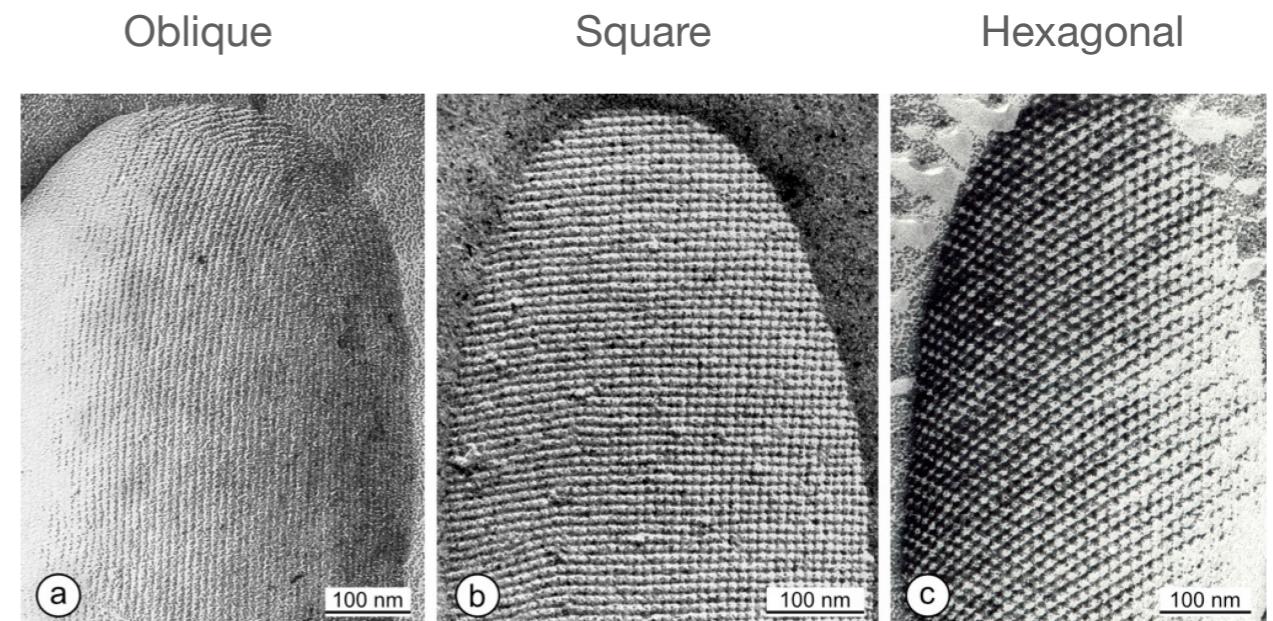
X-ray diffraction

We can use this diffraction property as a probe to determine the atom-level features because atomic size is comparable to the wavelength x-rays

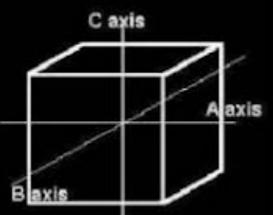
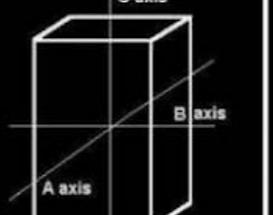
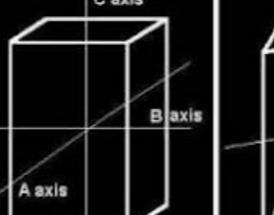
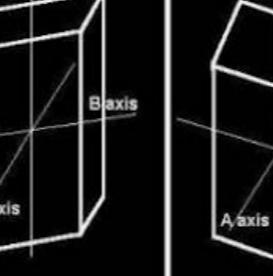
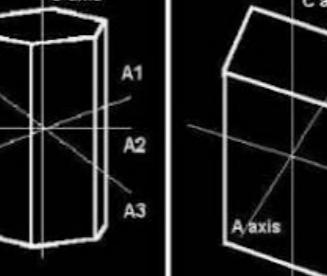
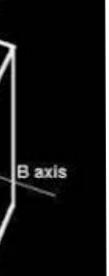
We need crystals of molecules

Diffraction : interference when waves encounter objects comparable in size to their wavelength

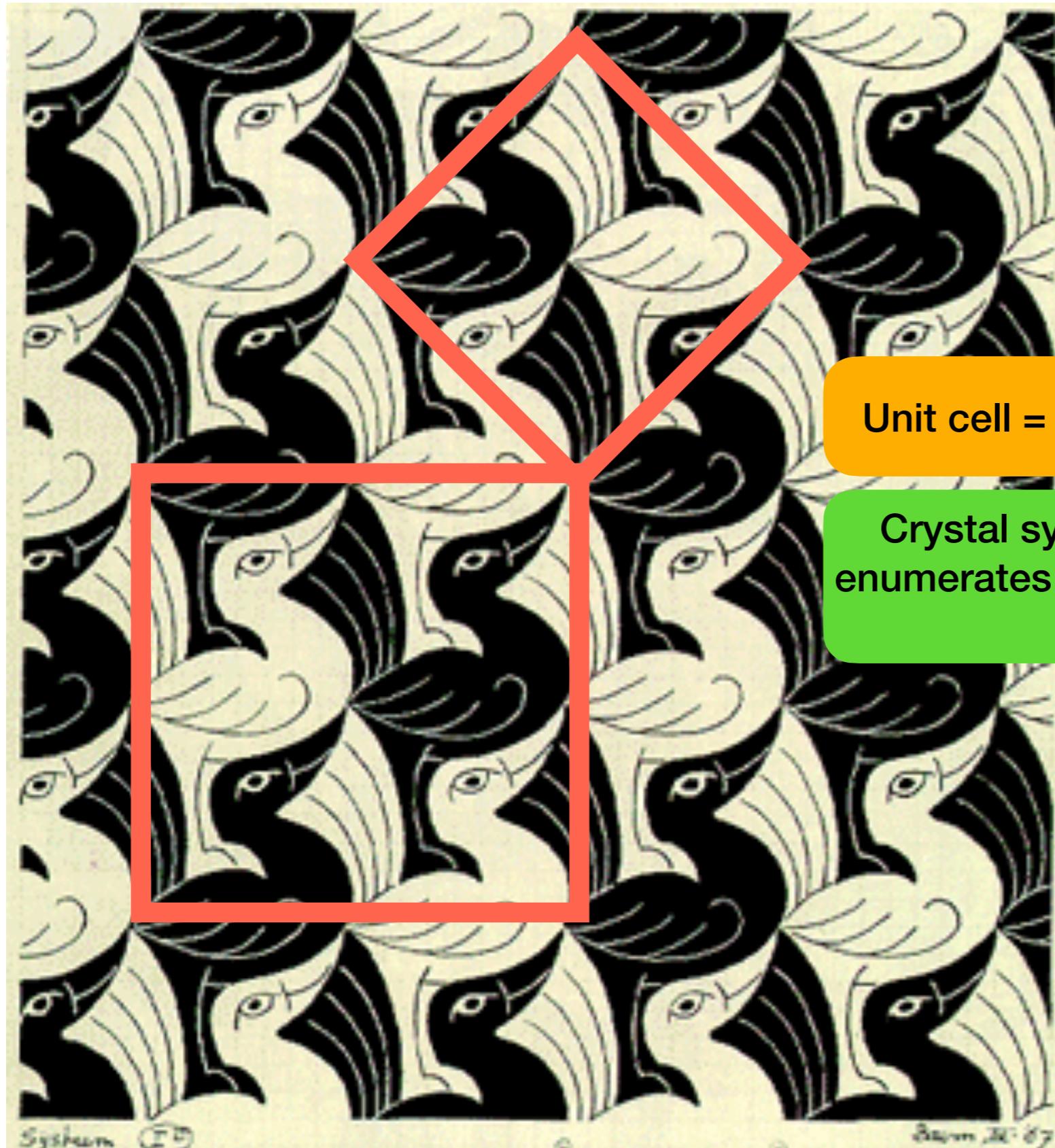
Crystals are based on lattice arrangements common in nature



Pum et al, Crystals 2021, 11(8), 869

Isometric	Tetragonal	Orthorhombic	Monoclinic	Triclinic	Hexagonal	Trigonal
						
						
Fluorite	Wulfenite	Tanzanite	Azurite	Amazonite	Emerald	Rhodochrosite

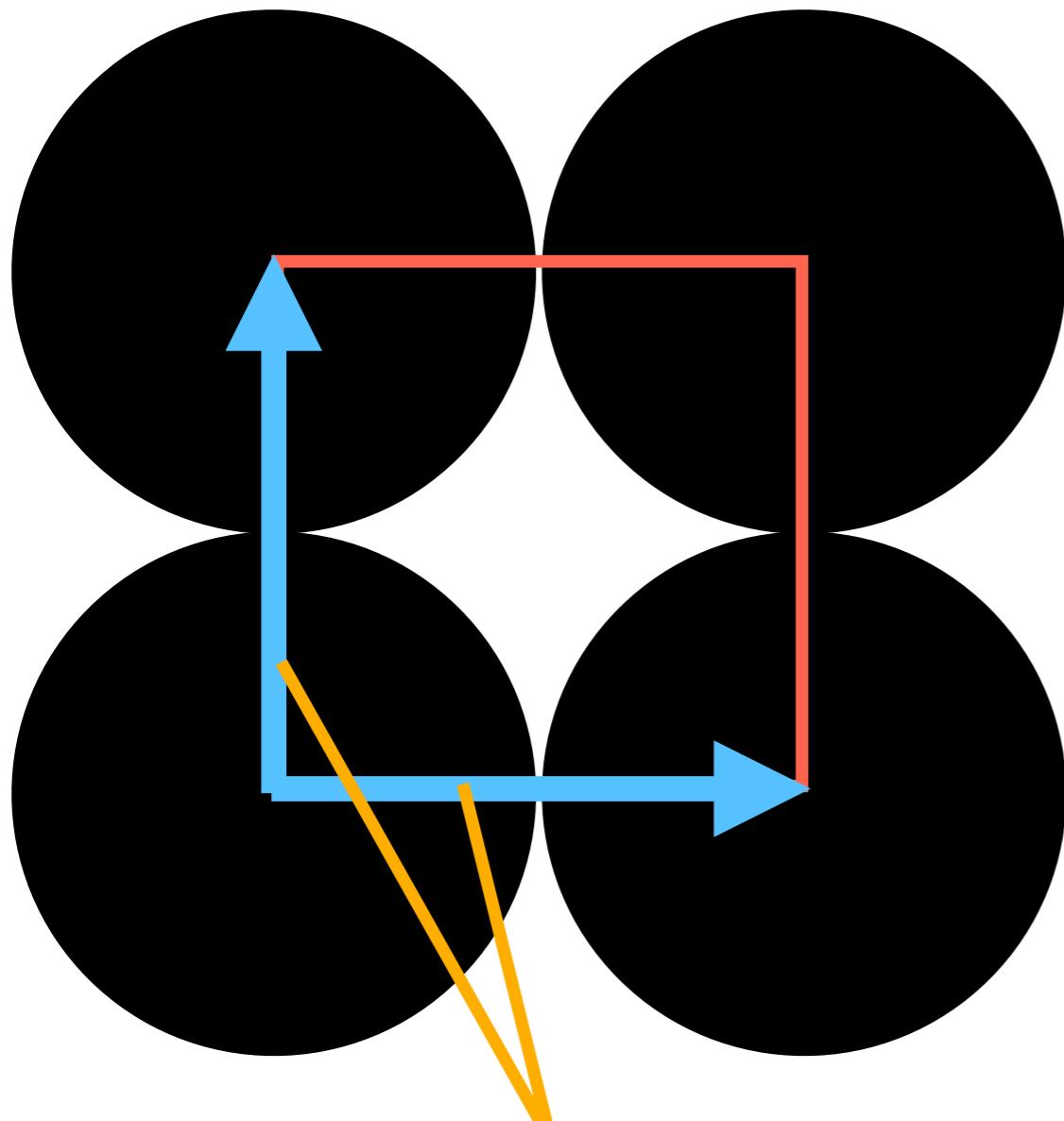
A crystal is just a repetition



Unit cell = repeating unit in a crystal

Crystal system or lattice system =
enumerates ways a space can be filled
with no voids

Lattice vectors



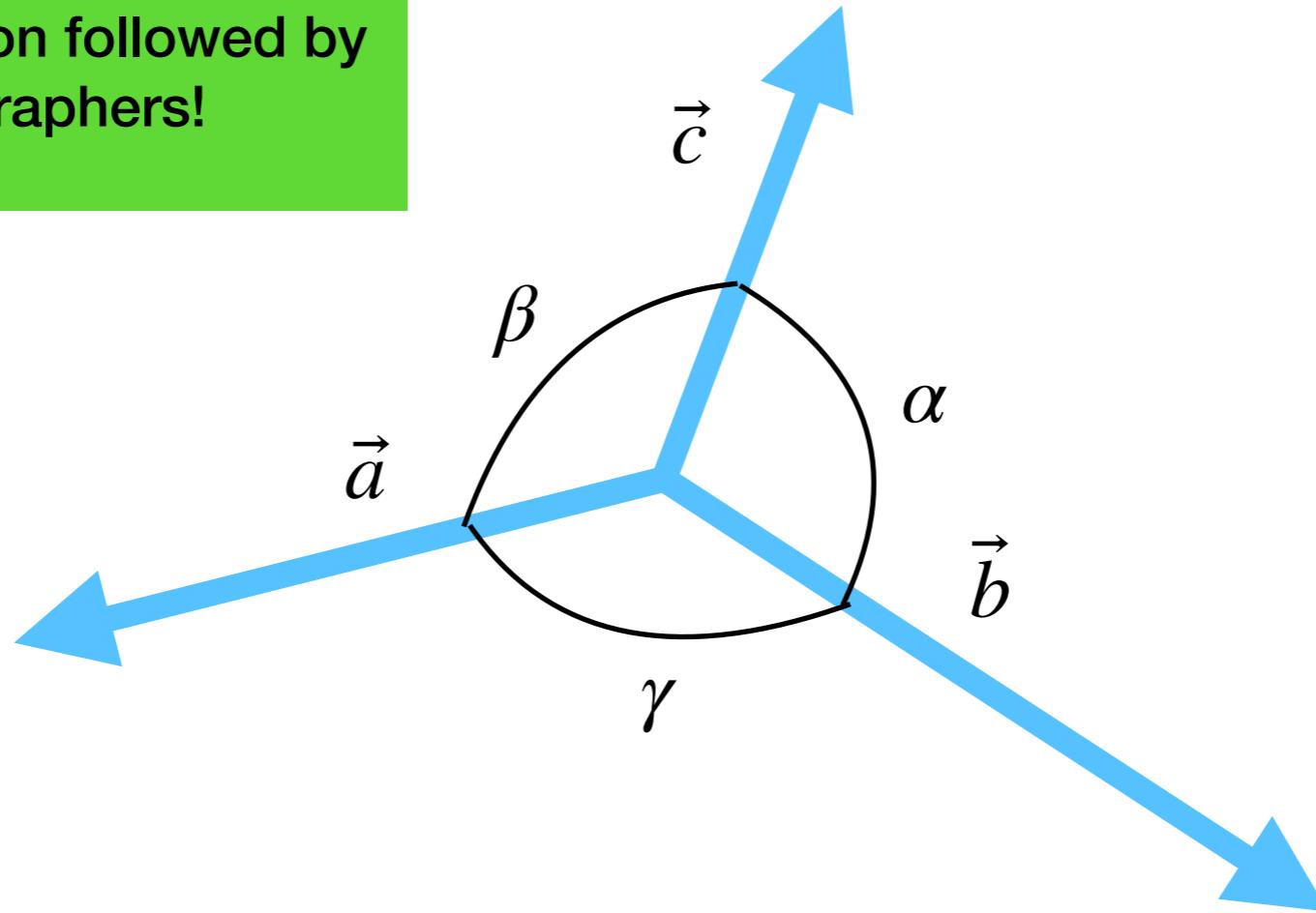
Lattice vectors connect any two lattice points

Lattice points are those separated by a vector sum of an integral number of lattice vectors

A vector sum of non-integral number of lattice vectors give the atomic positions in the crystal

Lattice angles

This is the convention followed by the crystallographers!



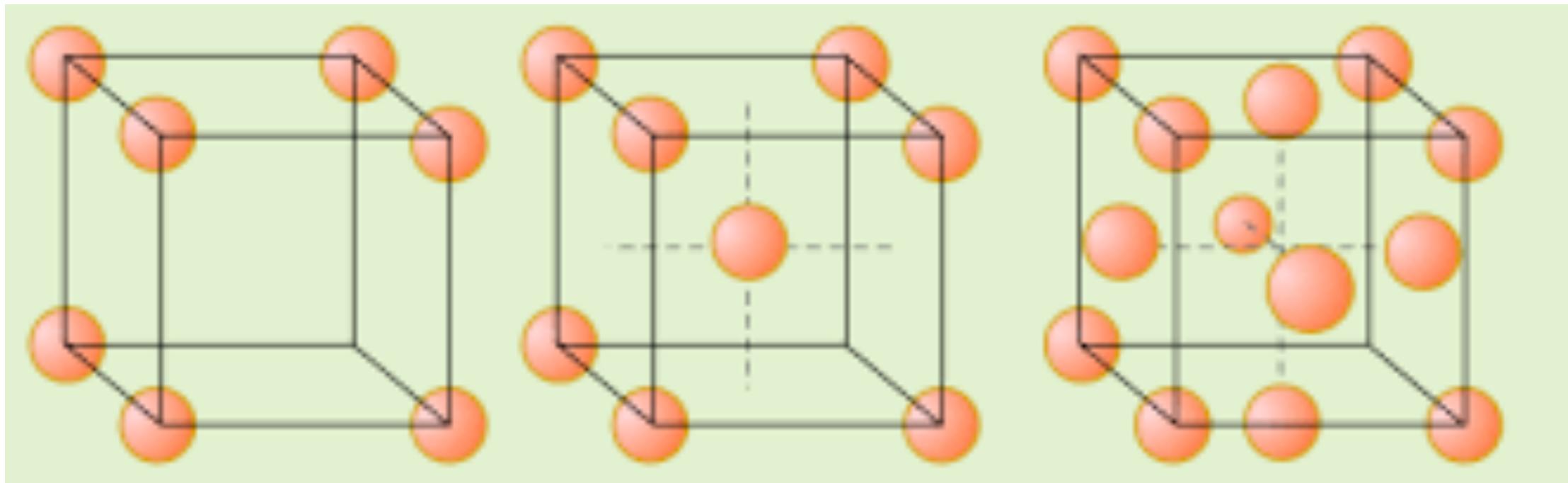
Together a, b, c and α, β, γ define the lattice parameters

Then there were 7!

There are only 7 lattice systems in 3D

System	Axes	Angles
Cubic	$a = b = c$	$\alpha = \beta = \gamma = 90^\circ$
Rhombohedral	$a = b = c$	$\alpha = \beta = \gamma$
Tetragonal	$a = b; c$	$\alpha = \beta = \gamma = 90^\circ$
Hexagonal	$a = b; c$	$\alpha = \beta = 90^\circ; \gamma = 120^\circ$
Orthorhombic	$a; b; c$	$\alpha = \beta = \gamma = 90^\circ$
Monoclinic	$a; b; c$	$\alpha = \gamma = 90^\circ; \beta$
Triclinic	$a; b; c$	$\alpha; \beta; \gamma$

Cubic symmetry have only three possible lattices



Simple cubic (SC)

Body-centered cubic (BCC)

Face-centered cubic (FCC)

This way total of 14 types of lattice are possible

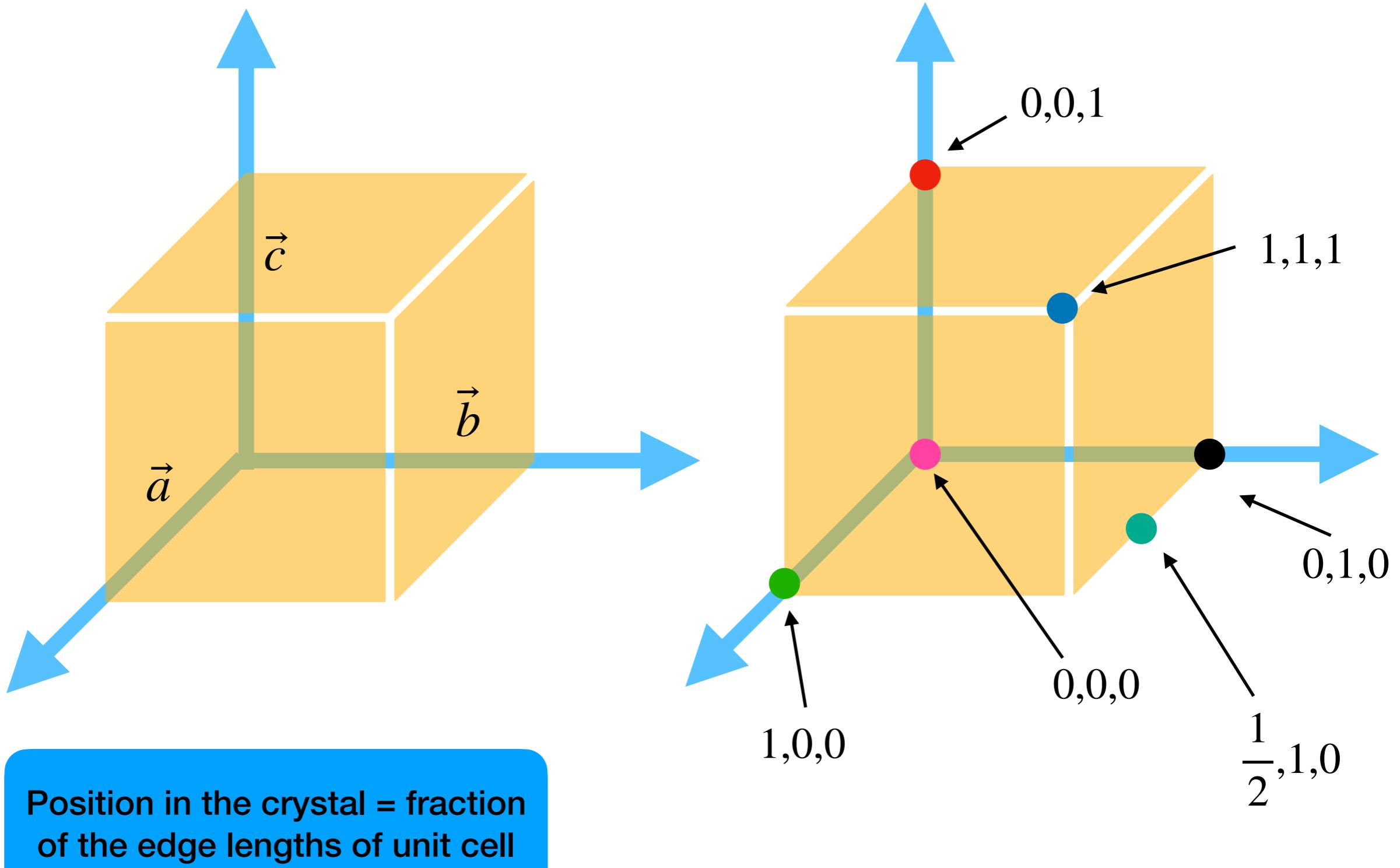
Bravais Lattice

Enumerates the ways to pack within a unit cell



Auguste Bravais (c. 1850)

Crystallographers' notations

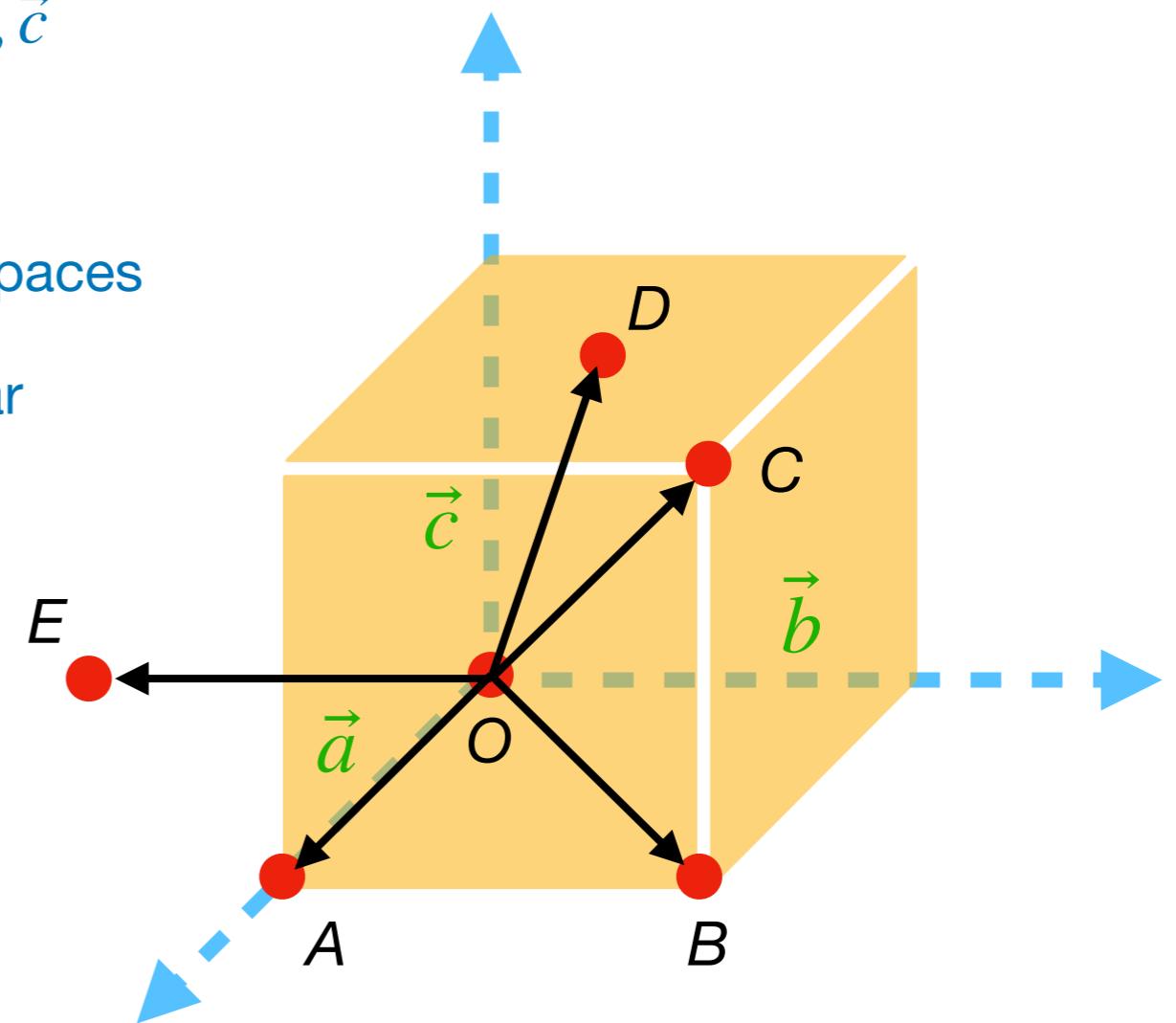


Crystallographers' notations for moving in a crystal

- Reposition the vector to pass through origin
- Take projections in terms of lattice vectors $\vec{a}, \vec{b}, \vec{c}$
- Adjust the results to smallest integer values
- Put them in square brackets, no commas, no spaces
- A negative direction is indicated with an overbar

These notations are called Miller indices

Vector	X	Y	Z	Miller indices
OA	1	0	0	[100]
OB	1	1	0	[110]
OC	1	1	1	[111]
OD	1/2	1/2	1	$\left[\frac{1}{2} \frac{1}{2} 1\right]$ [112]
OE	0	-1	0	$[0 \cancel{-1} 0]$ [0 $\bar{1}$ 0]



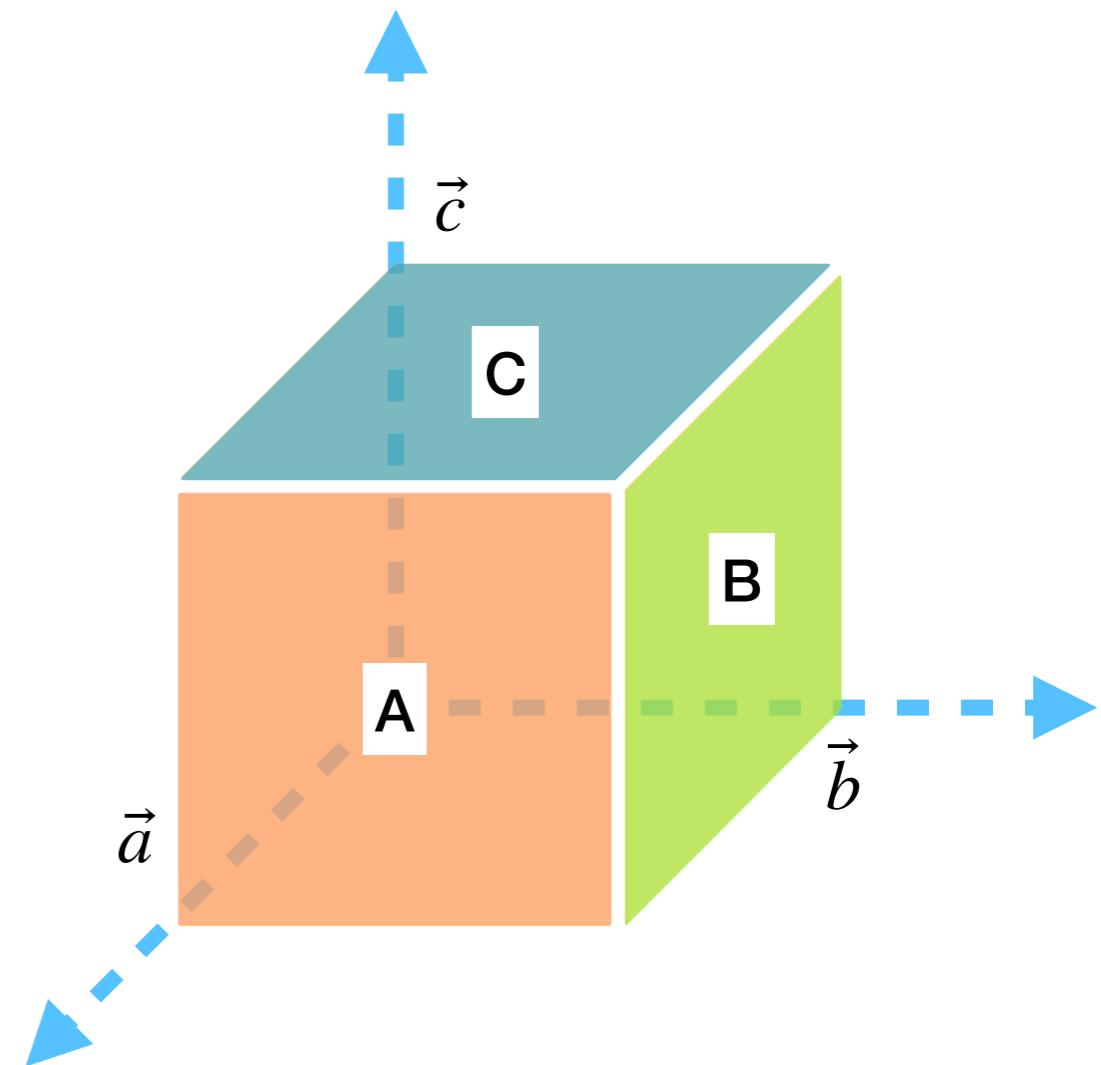
Crystallographers' notations for planes in a crystal

Miller indices can be used to describe crystallographic planes

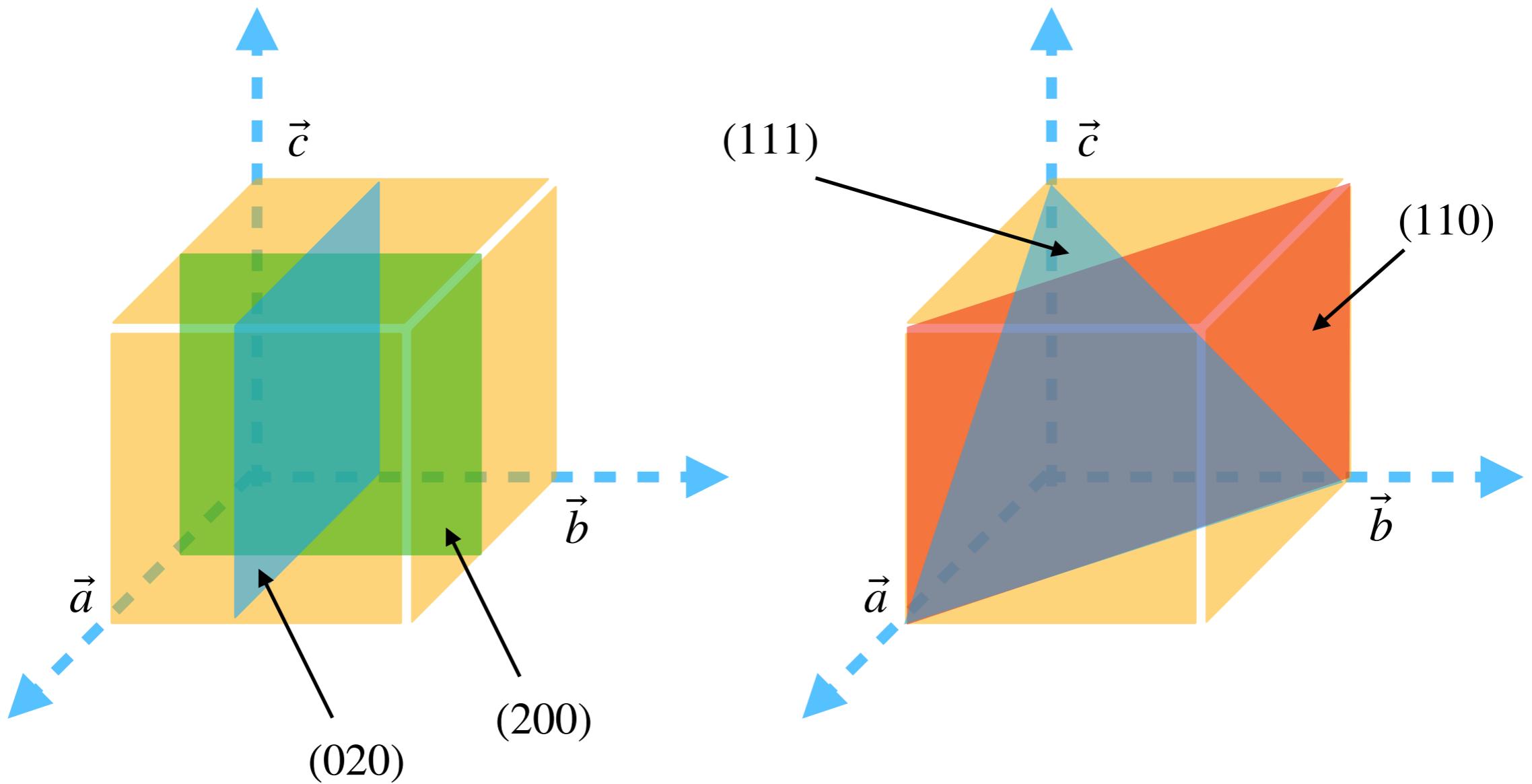
- Identify the points where the plane intercepts the axes in terms of fractions of \vec{a} , \vec{b} , \vec{c}
- Take reciprocals of the intercepts
- Divide by the greatest common factor to yield integer values
- Enclose in parentheses, with no commas (hkl)

	Axis intercepts			
Plane	X	Y	Z	Notation
A	1	∞	∞	(100)
B	∞	1	∞	(010)
C	∞	∞	1	(001)

These planes are called Miller Planes

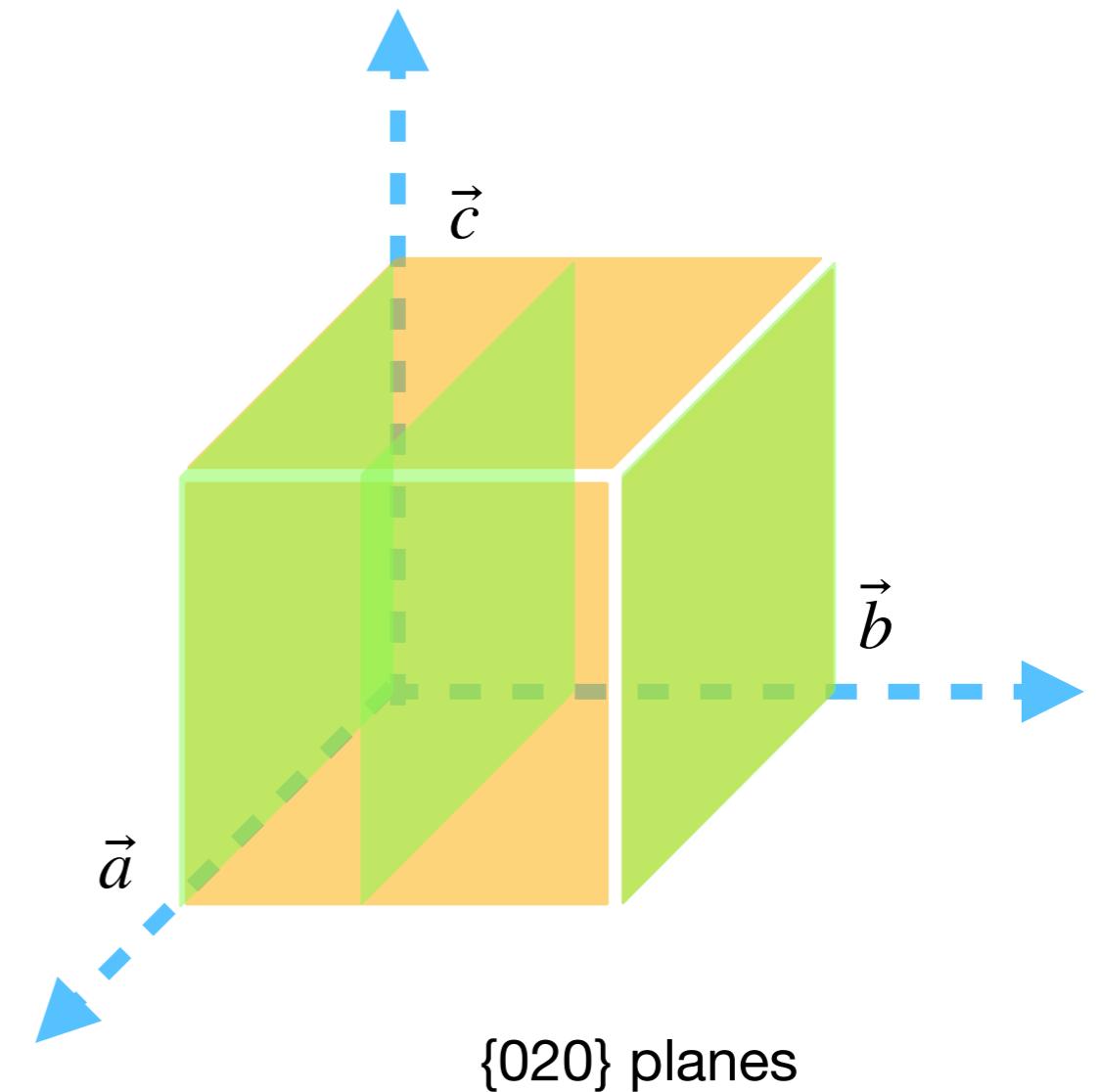
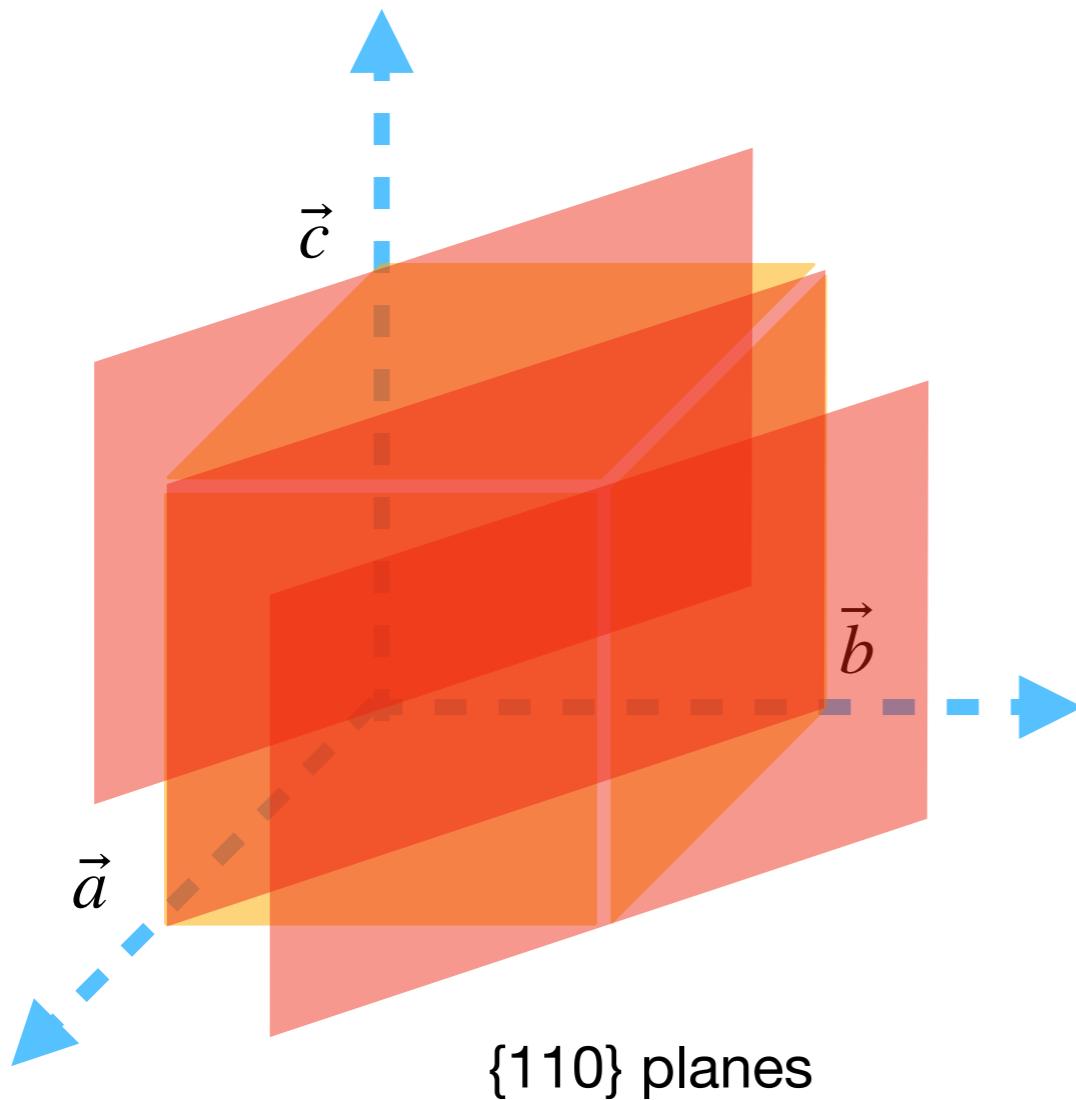


More Miller planes



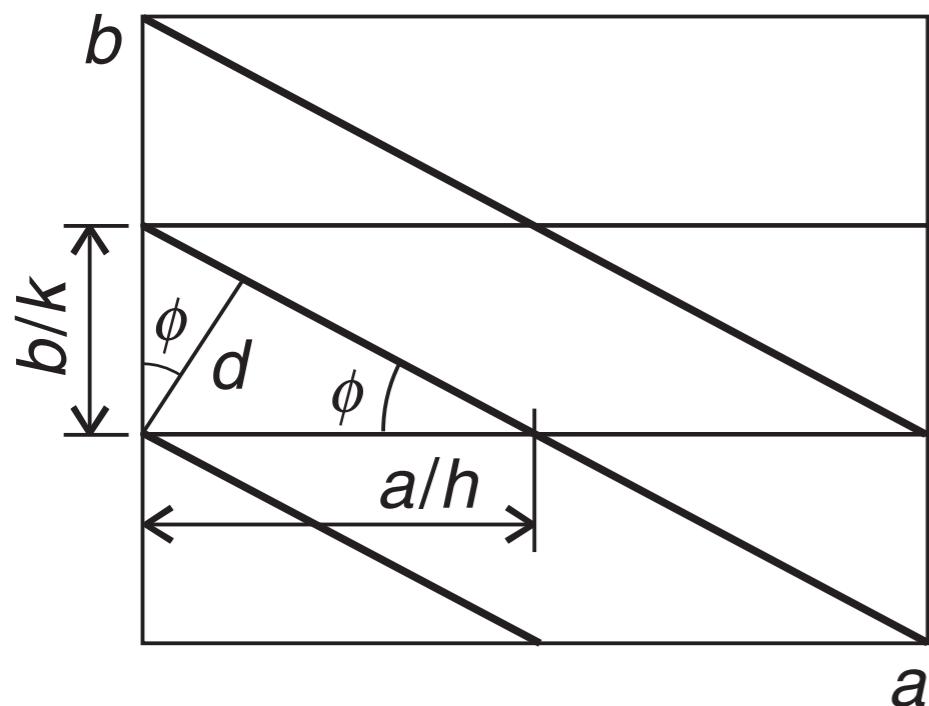
Miller planes are not single

A family of planes in the cubic system contains all of the planes with the same set of Miller Indices but in any order.
Families of planes are indicated by curly brackets, $\{hkl\}$.



Distance between planes in a crystal

Illustrating in 2D



$$\cos\phi = \frac{d}{b/k} \quad \sin\phi = \frac{d}{a/h}$$

$$\sin^2\phi + \cos^2\phi = 1$$

$$\Rightarrow \frac{h^2 d^2}{a^2} + \frac{k^2 d^2}{b^2} = 1$$

$$\Rightarrow \frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{b^2}$$

Distance between two consecutive planes in family {hkl}



$$\frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{b^2} + \frac{l^2}{c^2}$$

Distance between planes in a crystal...*contd*

Problems

Derive the distance formula for simple cubic

$$\frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{b^2} + \frac{l^2}{c^2}$$

For simple cubic: $a = b = c \implies \frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{a^2} + \frac{l^2}{a^2}$

$$\implies d = \frac{a}{\sqrt{h^2 + k^2 + l^2}}$$

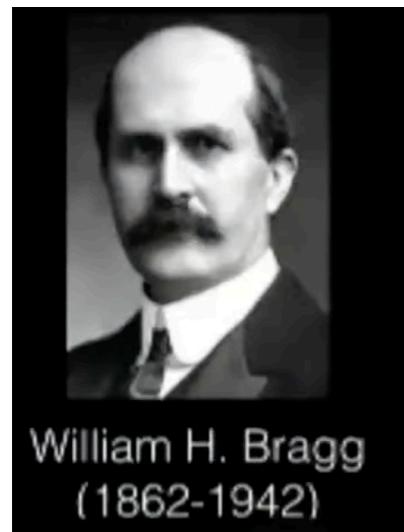
Find out the separation between {123} & {246} planes in an orthorhombic cell with
 $a = 0.82$ nm, $b = 0.94$ nm, $c = 0.75$ nm

For {123}: $d \approx 0.21$ nm

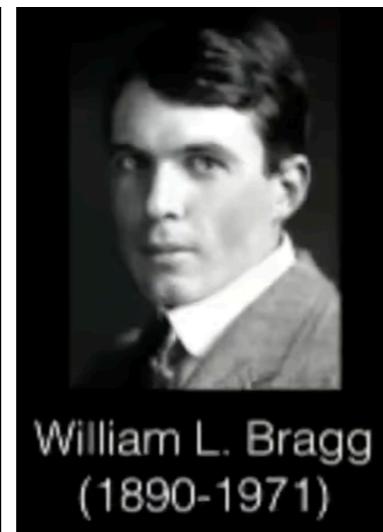
For {246}: $d \approx 0.11$ nm

In general, increasing the indices uniformly by a factor n decreases the separation of the planes by n .

Principle of x-ray diffraction by crystals



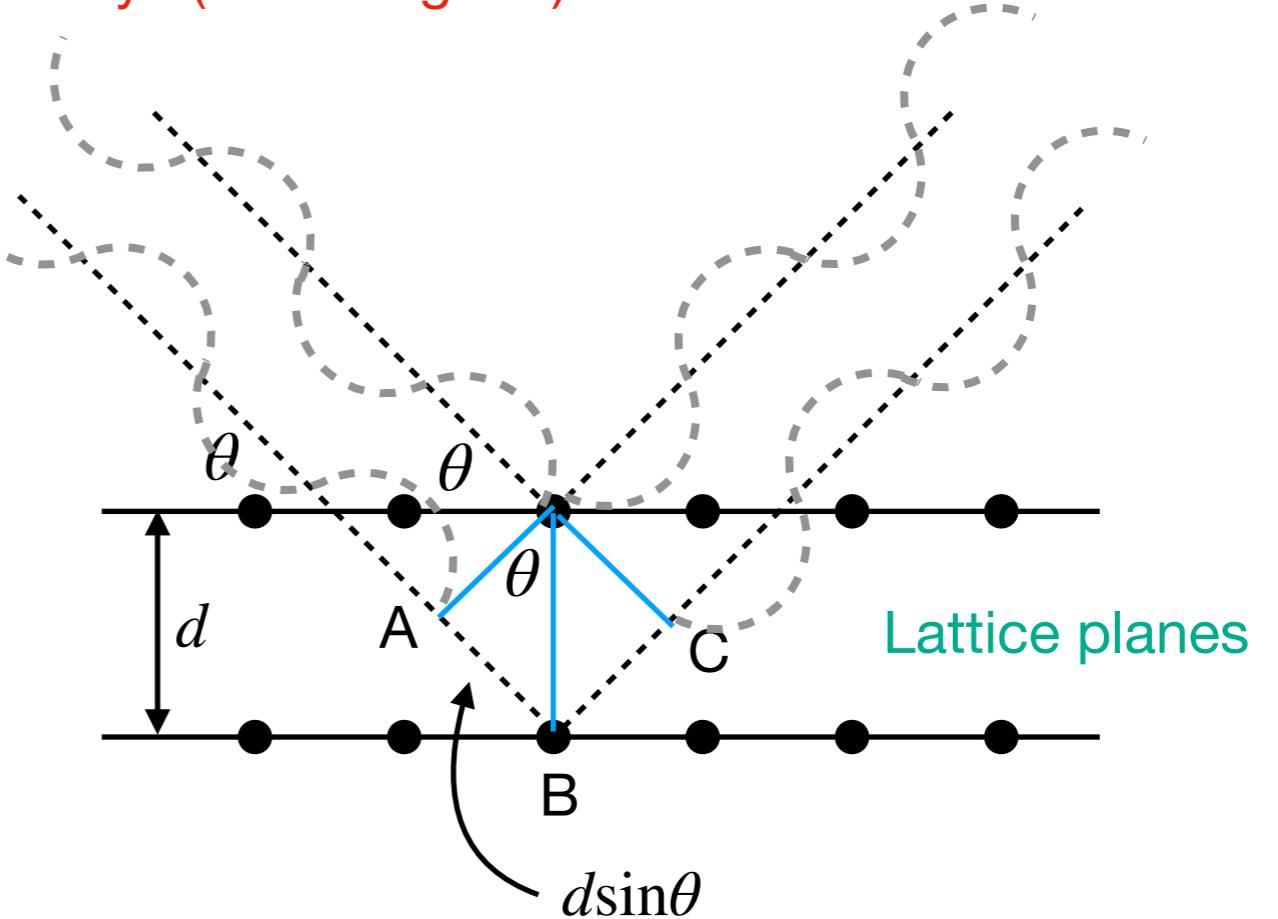
William H. Bragg
(1862-1942)



William L. Bragg
(1890-1971)

Incoming x-rays (wavelength λ)

Reflected x-rays



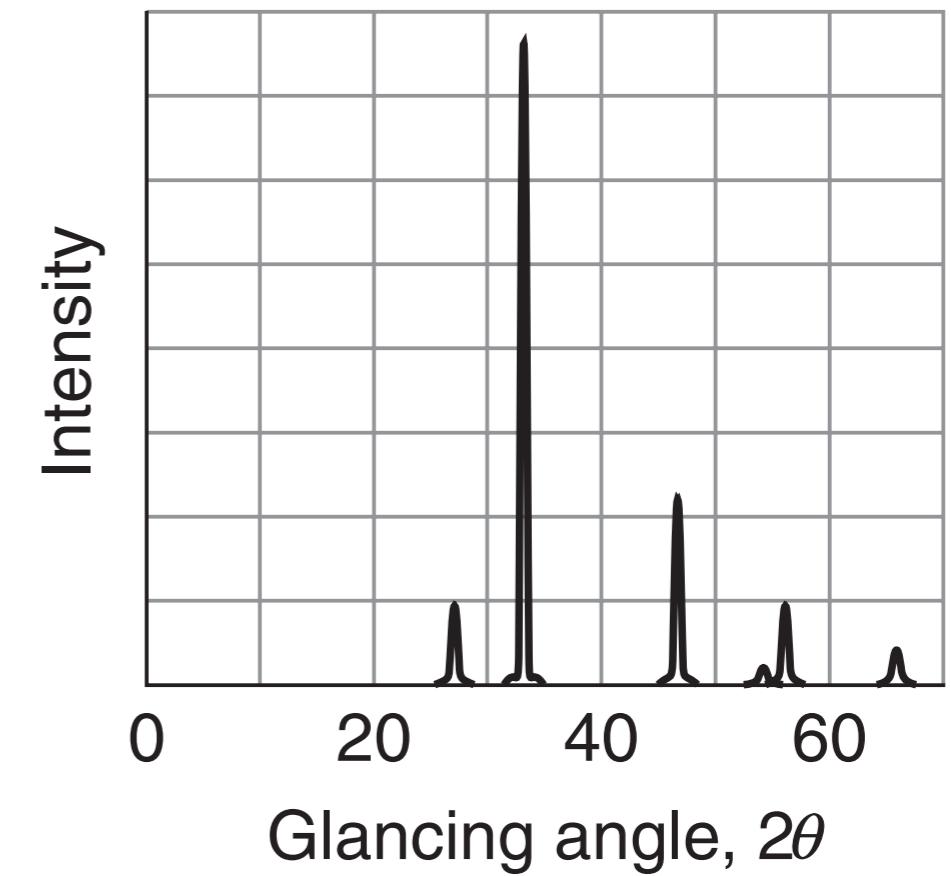
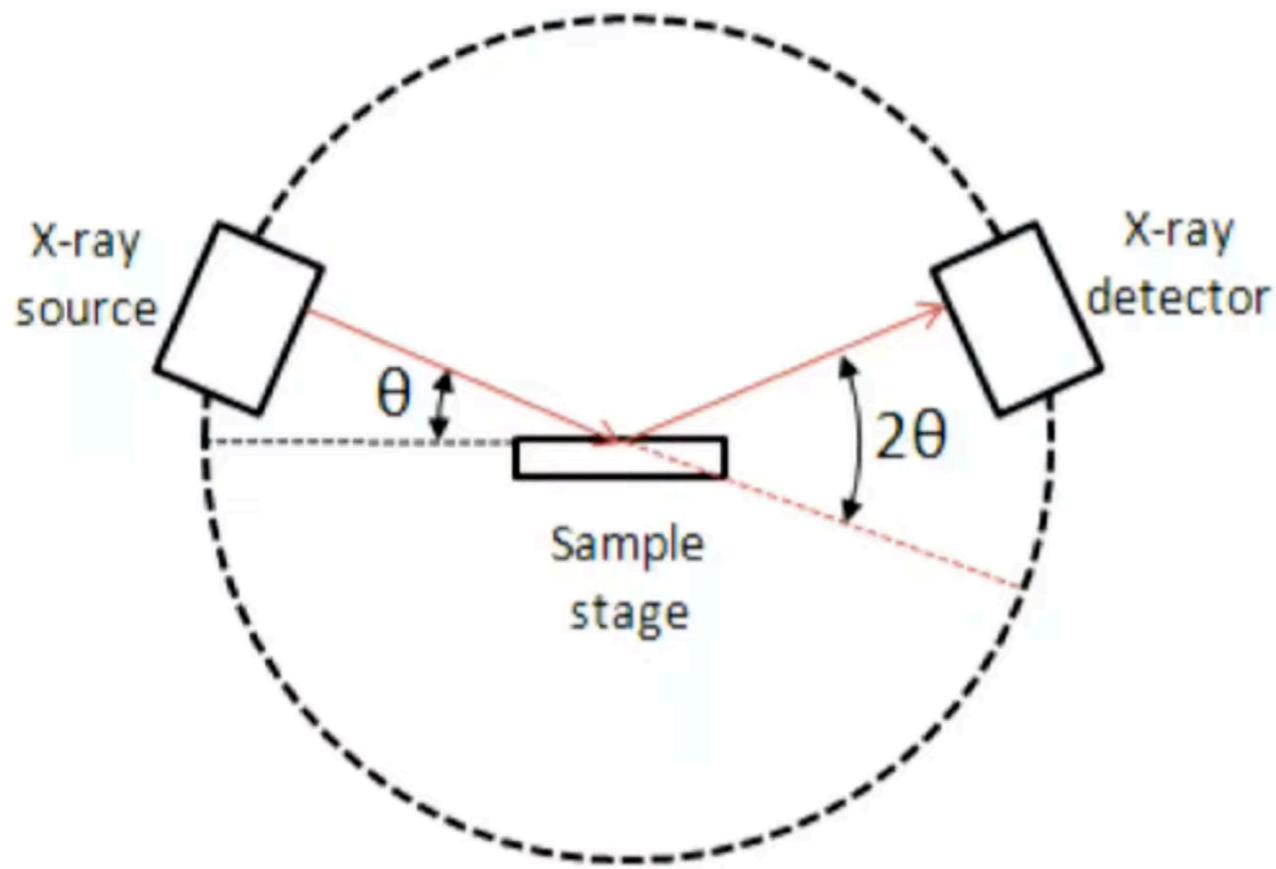
Condition for constructive interference

Path length difference between the waves has to be integer multiples of wavelength

$$AB + BC = n\lambda \implies 2d \sin \theta = n\lambda$$

Bragg law

Recording x-ray diffraction



For cubic crystal

$$\left(\frac{\lambda}{2a}\right)^2 = \frac{\sin^2\theta}{h^2 + k^2 + l^2}$$

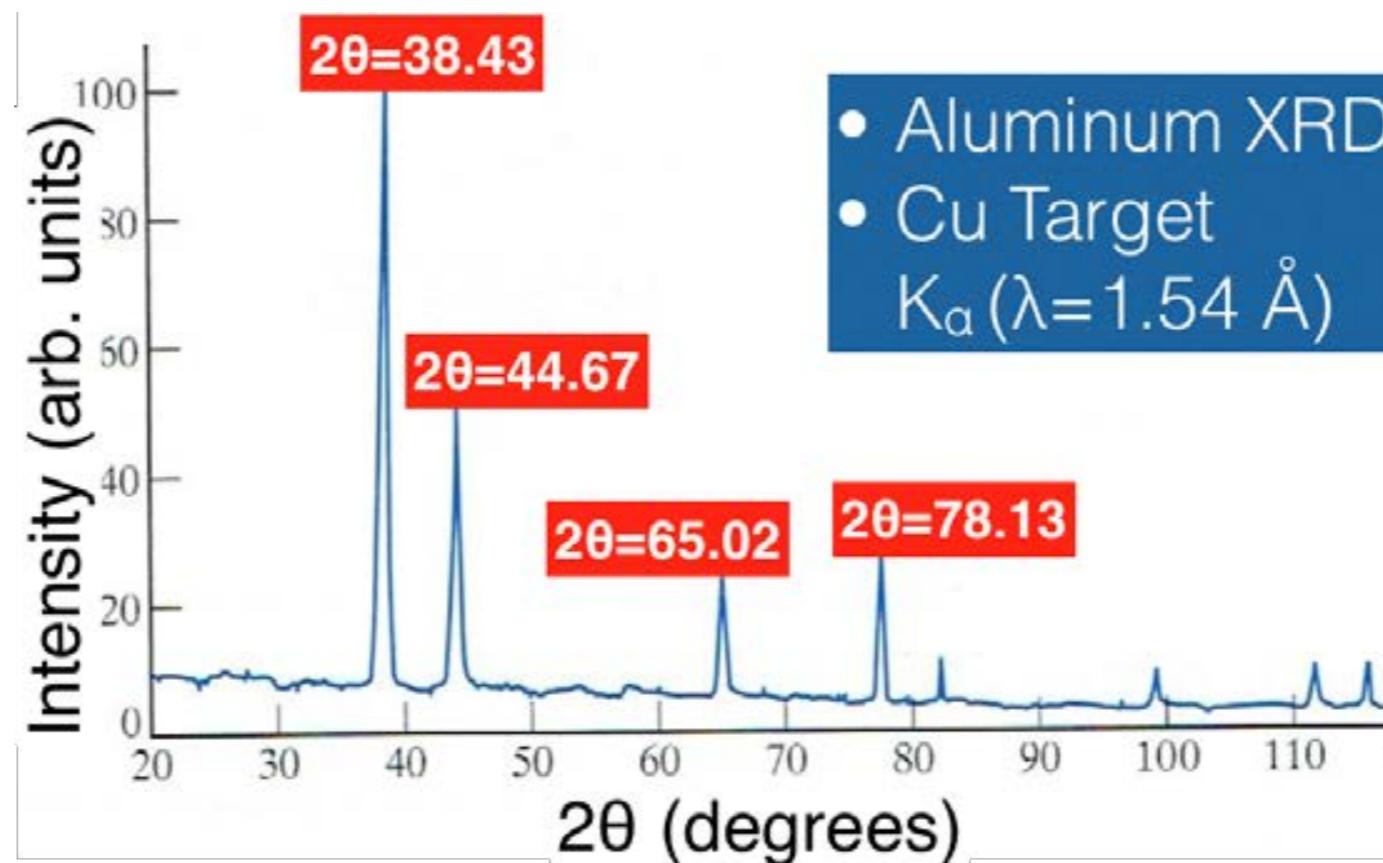
Selection rules for x-ray diffraction

Crystal	Allowed reflections	Forbidden reflections
Simple cubic	Any	None
BCC	$h + k + l = \text{even}$	$h + k + l = \text{odd}$
FCC	$h, k, l - \text{ all odd}$ or all even	$h, k, l - \text{ mixed}$

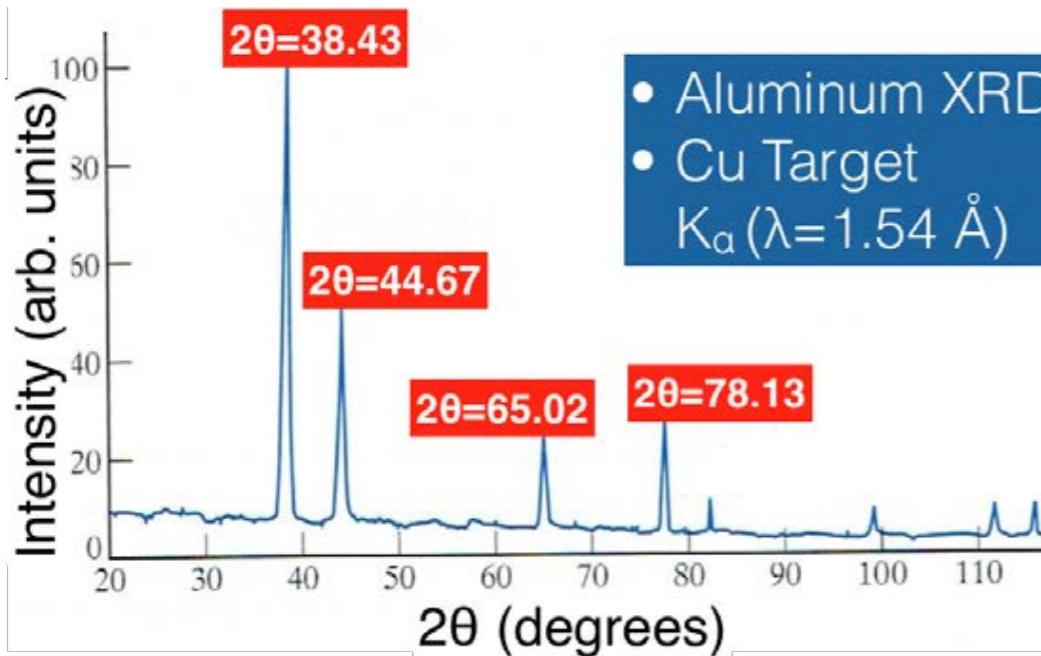
How can we use these information to determine crystal structure from x-ray diffraction patterns?

X-ray diffraction pattern to the crystal structure

- $2\theta \rightarrow \sin^2\theta$ for all peaks in the x-ray diffraction pattern.
- Normalize all $\sin^2\theta$ by the smallest value.
- Clear fractions from the normalized $\sin^2\theta$. Let's call this z .
- Speculate all the hkl values that can satisfy $h^2 + k^2 + l^2 = z$.
- Combining this hkl with the selection rules predicts the lattice type.
- Calculate for each θ , the quantity: $\sin^2\theta/(h^2 + k^2 + l^2)$. If each result is identical, then it is validated and gives lattice constant a .



X-ray diffraction pattern to the crystal structure: example



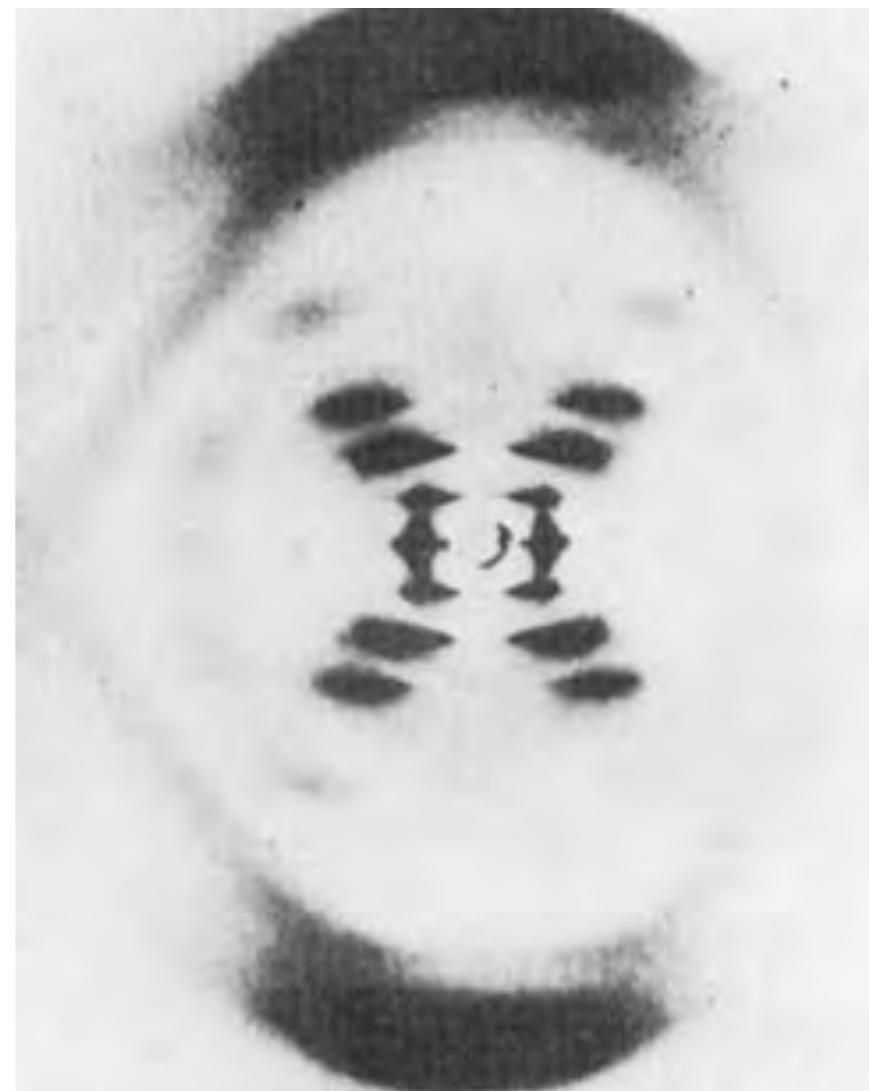
$$\left(\frac{\lambda}{2a}\right)^2 = \frac{\sin^2\theta}{h^2 + k^2 + l^2} \rightarrow \text{Lattice constant, } a$$

2θ	$\sin^2\theta$	Normalized	Z	hkl	hkl type	$\frac{\sin^2\theta}{h^2 + k^2 + l^2}$
38.43	0.1083	1.0	3	111	all odd	0.0361
44.67	0.1444	1.33	4	200	all even	0.0361
65.02	0.2888	2.67	8	220	all even	0.0361
78.13	0.3972	3.67	11	311	all odd	0.0361

$$a = 4.1 \text{ \AA}$$

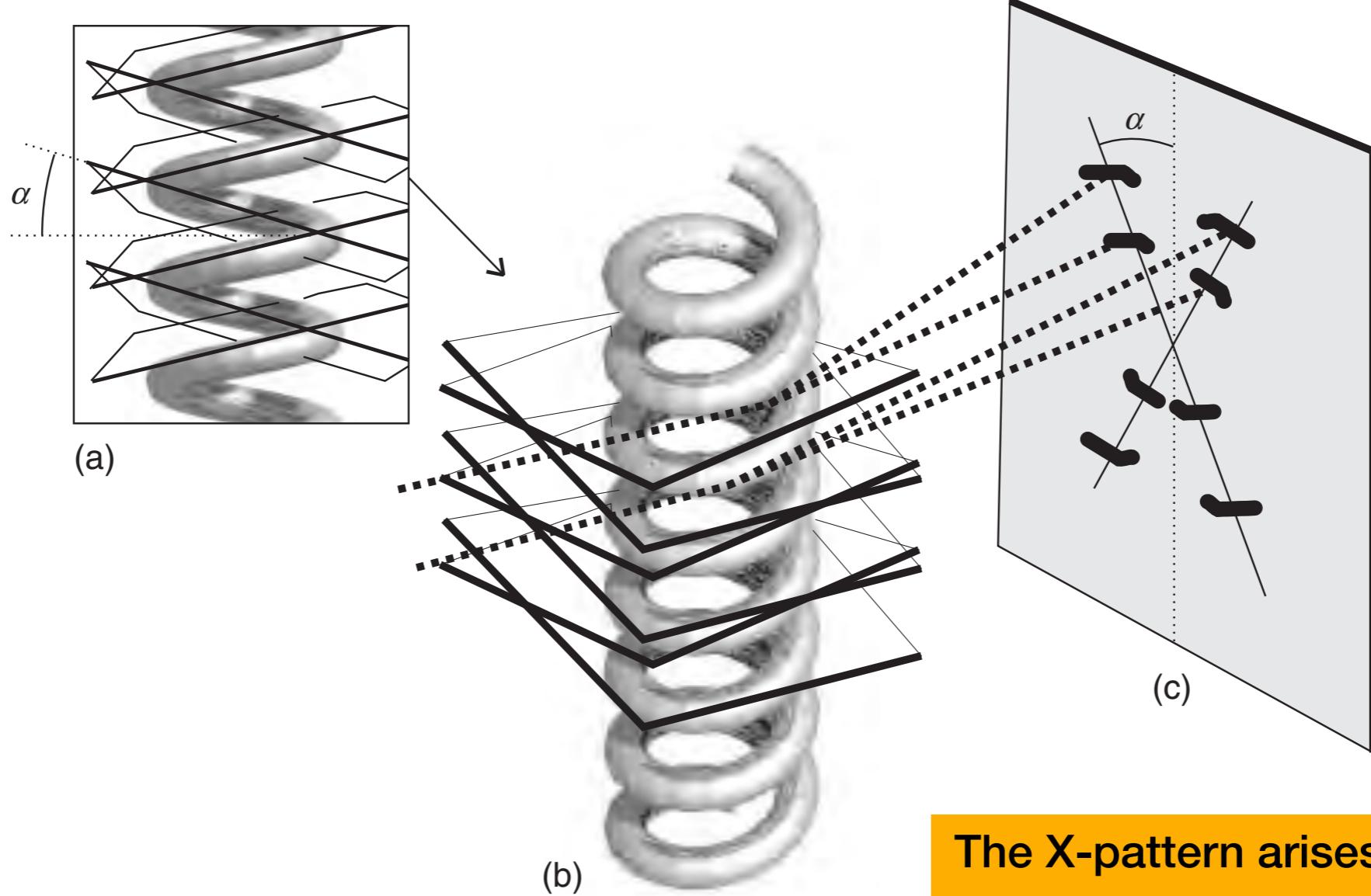
FCC lattice

X-ray diffraction patterns of DNA



The X-ray diffraction pattern obtained from a fiber of B-DNA. The black dots are the reflections, the points of maximum constructive interference, that are used to determine the structure of the molecule

X-ray diffraction data of a helix



The X-pattern arises from a set of planes at angle α and another set at angle $-\alpha$

How to prepare crystals of proteins or DNA

Common strategies for biopolymer crystallization

- Salt precipitation
- Dialysis
- Vapor diffusion

Covered already in L22

vapor diffusion method of biopolymer crystallization

- A single drop of dilute biopolymer solution hangs above a reservoir solution that is very concentrated in a non-volatile solute.
- Solvent evaporates from the more dilute drop until the vapor pressure of water in the closed container reaches a constant equilibrium value.
- In the course of evaporation, the biopolymer solution becomes more concentrated
- Biopolymers pack into repeating arrays and finally form crystals

