

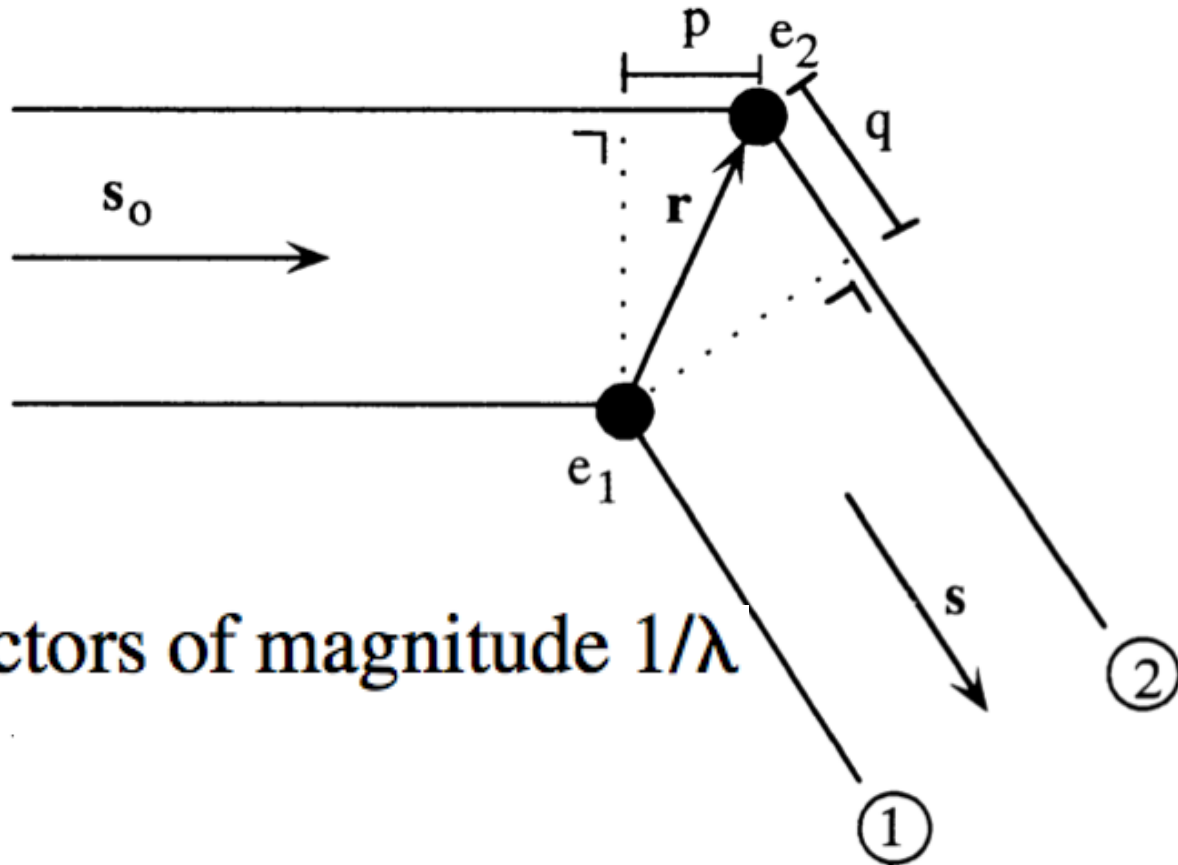
Structural Biology Methods

Fall 2020

Lecture #3

System of two electrons

Figure 4.4. A system of two electrons: e_1 and e_2 . The path difference between the scattered waves 1 and 2 is $p + q$.



\mathbf{s}_0 and \mathbf{s} are wave vectors of magnitude $1/\lambda$

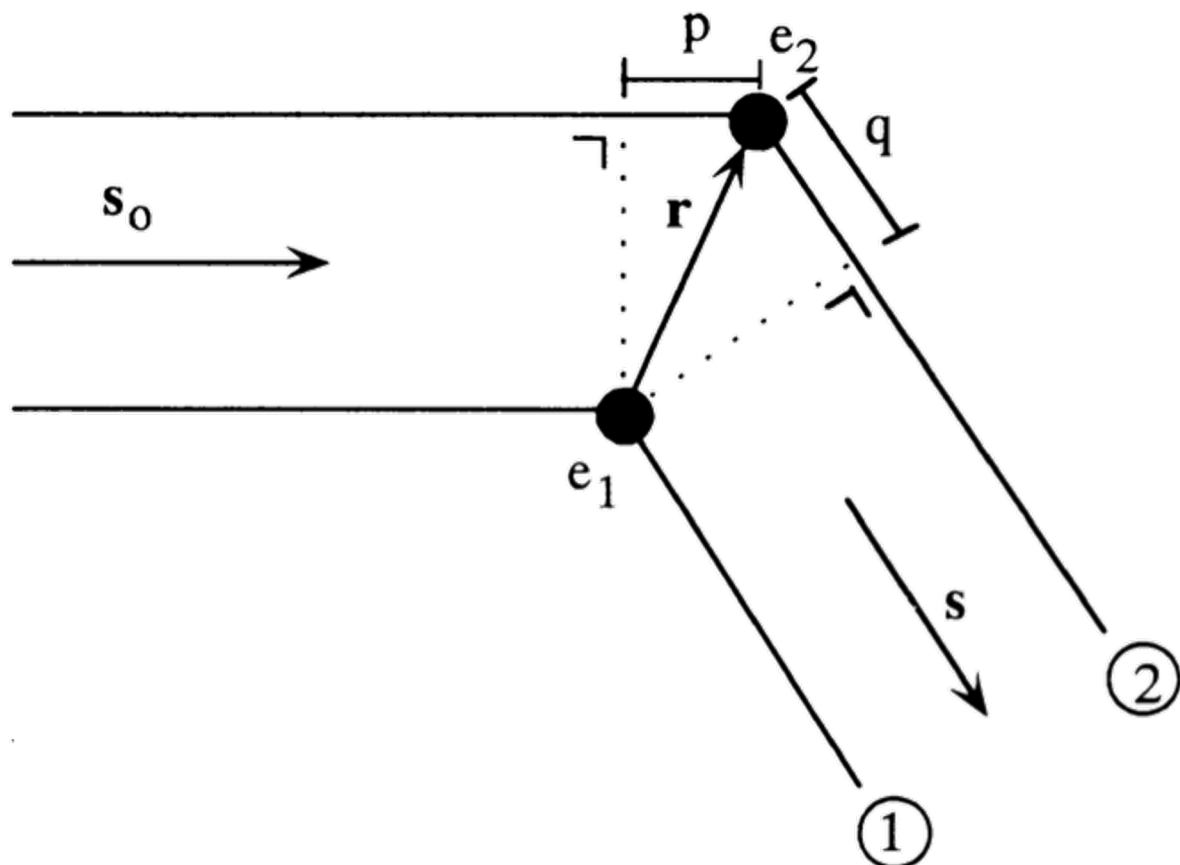
$$p = \lambda \cdot \mathbf{r} \cdot \mathbf{s}_0$$

$$q = -\lambda \cdot \mathbf{r} \cdot \mathbf{s}$$

minus sign is due to the fact that the projection of \mathbf{r} on \mathbf{s} has a direction opposite to \mathbf{s}

$$p + q = \lambda \cdot \mathbf{r} \cdot (\mathbf{s}_0 - \mathbf{s}).$$

Figure 4.4. A system of two electrons: e_1 and e_2 . The path difference between the scattered waves 1 and 2 is $p + q$.



The wave along electron e_2 is lagging behind in phase compared with the wave along e_1 . With respect to wave 1, the phase of wave 2 is

$$-\frac{2\pi\mathbf{r} \cdot (\mathbf{s}_0 - \mathbf{s}) \cdot \lambda}{\lambda} = 2\pi\mathbf{r} \cdot \mathbf{S},$$

where

$$\mathbf{S} = \mathbf{s} - \mathbf{s}_0 \tag{4.1}$$

Scattering by a unit cell

Suppose a unit cell has n atoms at positions \mathbf{r}_j ($j = 1, 2, 3, \dots, n$) with respect to the origin of the unit cell (Figure 4.12). With their own nuclei as origins, the atoms diffract according to their atomic scattering factor f . If the origin is now transferred to the origin of the unit cell, the phase angles change by $2\pi\mathbf{r}_j \cdot \mathbf{S}$. With respect to the new origin, the scattering is given by

$$\mathbf{f}_j = f_j \exp[2\pi i \mathbf{r}_j \cdot \mathbf{S}],$$

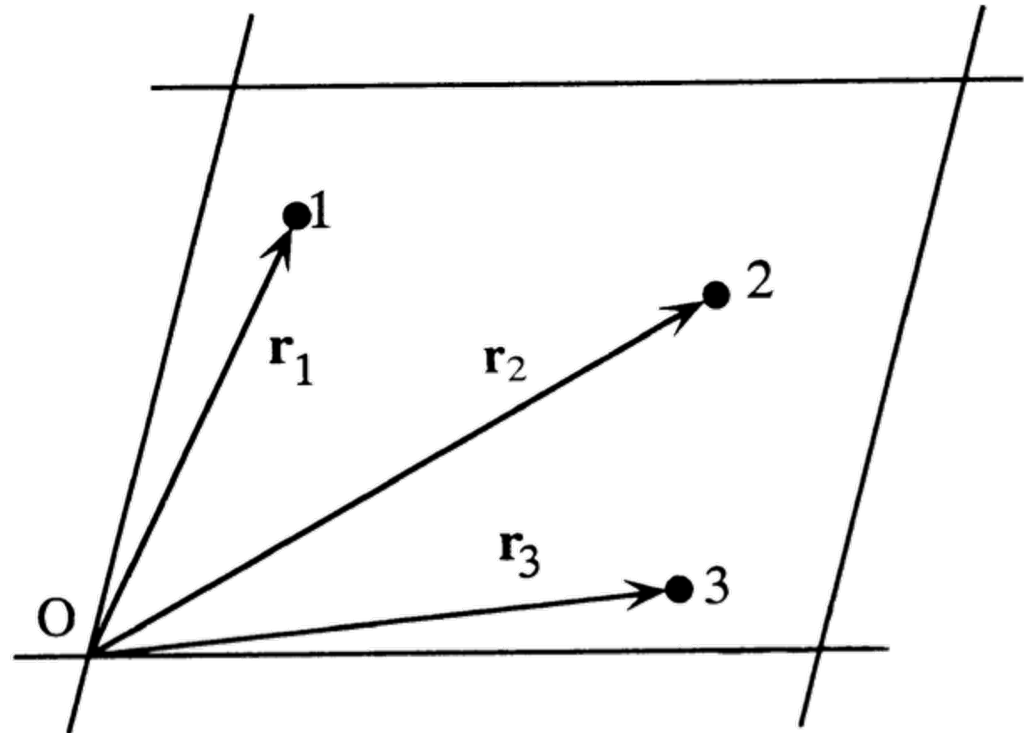
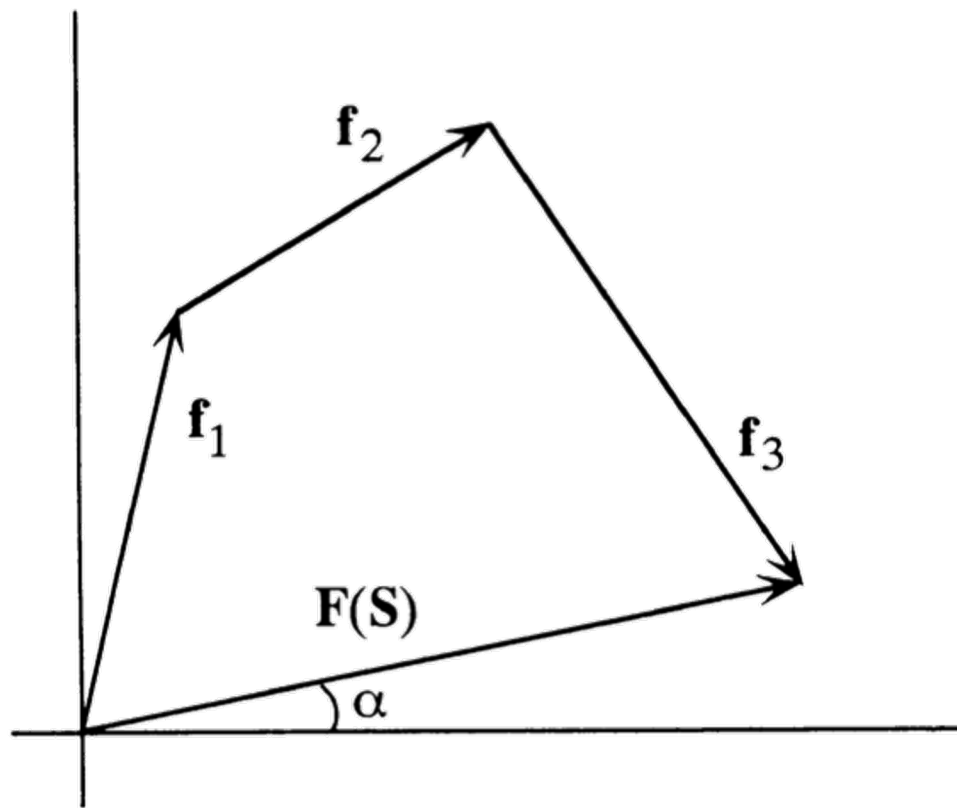


Figure 4.12. A unit cell with three atoms (1, 2, and 3) at positions \mathbf{r}_1 , \mathbf{r}_2 , and \mathbf{r}_3 .

Figure 4.13. The structure factor $\mathbf{F}(\mathbf{S})$ is the sum of the scattering by the separate atoms in the unit cell.



unit cell is

$$\mathbf{F}(\mathbf{S}) = \sum_{j=1}^n f_j \exp[2\pi i \mathbf{r}_j \cdot \mathbf{S}]. \quad (4.3)$$

$\mathbf{F}(\mathbf{S})$ is called the *structure factor* because it depends on the arrangement (structure) of the atoms in the unit cell (Figure 4.13).

Scattering by a crystal

Suppose that the crystal has translation vectors \mathbf{a} , \mathbf{b} , and \mathbf{c} and contains a large number of unit cells: n_1 in the \mathbf{a} direction, n_2 in the \mathbf{b} direction, and n_3 in the \mathbf{c} direction (Figure 4.14).

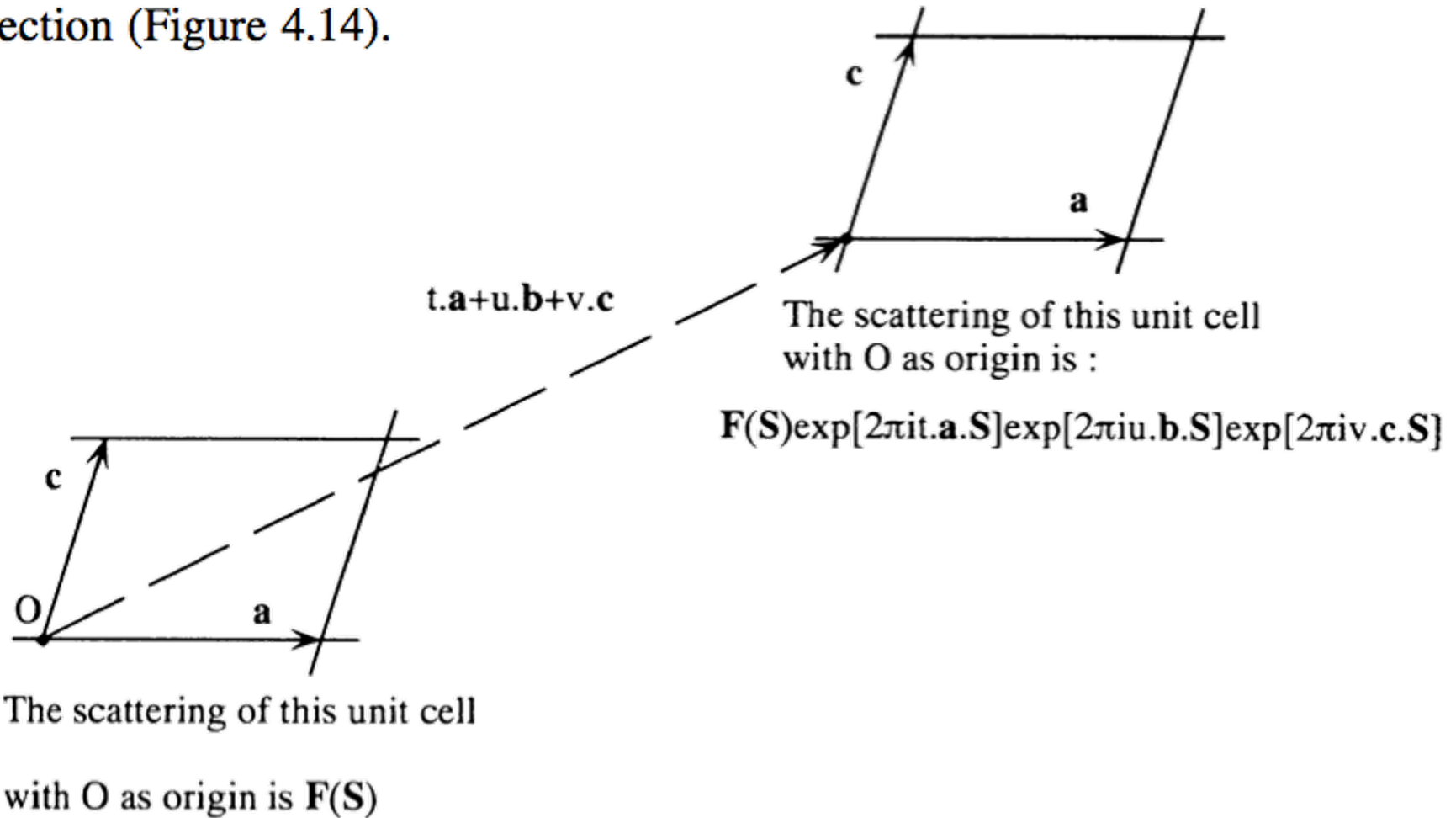


Figure 4.14. A crystal contains a large number of identical unit cells. Only two of them are drawn in this figure.

To obtain the scattering by the crystal, we must add the scattering by all unit cells with respect to a single origin. We choose the origin O in Figure 4.14. For a unit cell with its own origin at position $t \cdot \mathbf{a} + u \cdot \mathbf{b} + v \cdot \mathbf{c}$, in which t , u , and v are whole numbers, the scattering is

$$\mathbf{F}(\mathbf{S}) \times \exp[2\pi i t \mathbf{a} \cdot \mathbf{S}] \times \exp[2\pi i u \mathbf{b} \cdot \mathbf{S}] \times \exp[2\pi i v \mathbf{c} \cdot \mathbf{S}].$$

The total wave $\mathbf{K}(\mathbf{S})$ scattered by the crystal is obtained by a summation over all unit cells:

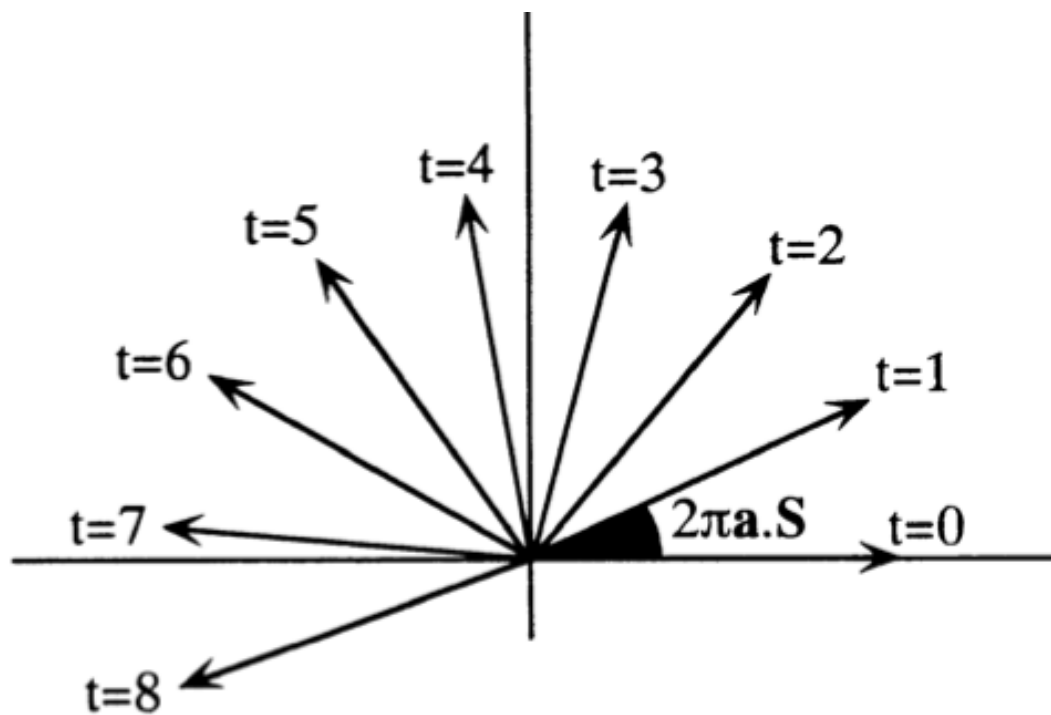
$$\mathbf{K}(\mathbf{S}) = \mathbf{F}(\mathbf{S}) \times \sum_{t=0}^{n_1} \exp[2\pi i t \mathbf{a} \cdot \mathbf{S}] \times \sum_{u=0}^{n_2} \exp[2\pi i u \mathbf{b} \cdot \mathbf{S}] \times \sum_{v=0}^{n_3} \exp[2\pi i v \mathbf{c} \cdot \mathbf{S}].$$

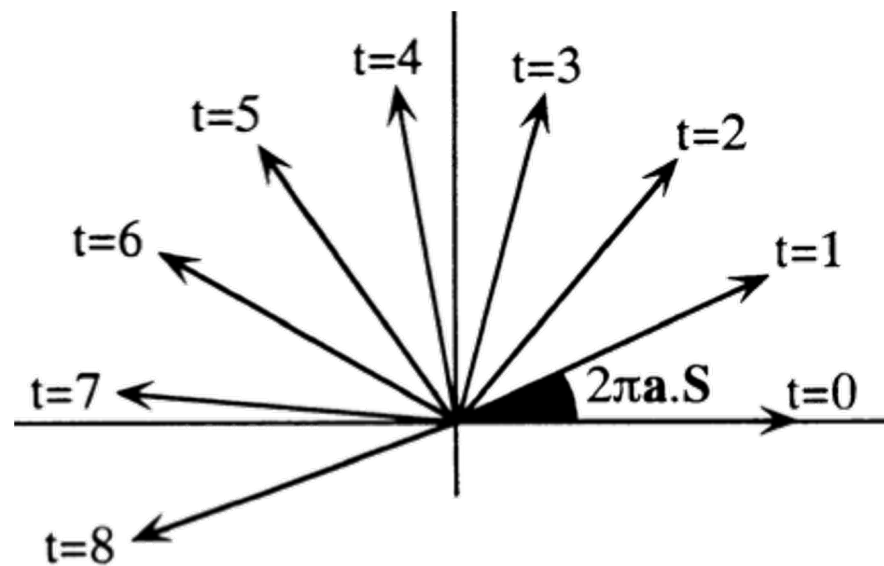
The total wave $\mathbf{K}(\mathbf{S})$ scattered by the crystal is obtained by a summation over all unit cells:

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Because n_1 , n_2 , and n_3 are very large, the summation $\sum_{t=0}^{n_1} \exp[2\pi i t \mathbf{a} \cdot \mathbf{S}]$ and the other two over u and v are almost always equal to zero unless $\mathbf{a} \cdot \mathbf{S}$ is an integer h , $\mathbf{b} \cdot \mathbf{S}$ is an integer k , and $\mathbf{c} \cdot \mathbf{S}$ is an integer l . This is easy to understand if we regard $\exp[2\pi i t \mathbf{a} \cdot \mathbf{S}]$ as a vector in the Argand diagram with a length of 1 and a phase angle $2\pi t \mathbf{a} \cdot \mathbf{S}$ (see Figure 4.15).

Figure 4.15. Each arrow represents the scattering by one unit cell in the crystal. Because of the huge number of unit cells and because their scattering vectors are pointing in different directions, the scattering by a crystal is, in general, zero. However, in the special case that $\mathbf{a} \cdot \mathbf{S}$ is an integer h , all vectors point to the right and the scattering by the crystal can be of appreciable intensity.





Conclusion: A crystal does not scatter X-rays, unless

$$\begin{aligned} \mathbf{a} \cdot \mathbf{S} &= h, \\ \mathbf{b} \cdot \mathbf{S} &= k, \\ \mathbf{c} \cdot \mathbf{S} &= l. \end{aligned} \tag{4.4}$$

These are known as the Laue conditions. h , k , and l are whole numbers, either positive, negative, or zero. The amplitude of the total scattered wave is proportional to the amplitude of the structure factor $\mathbf{F}(\mathbf{S})$ and the number of unit cells in the crystal.

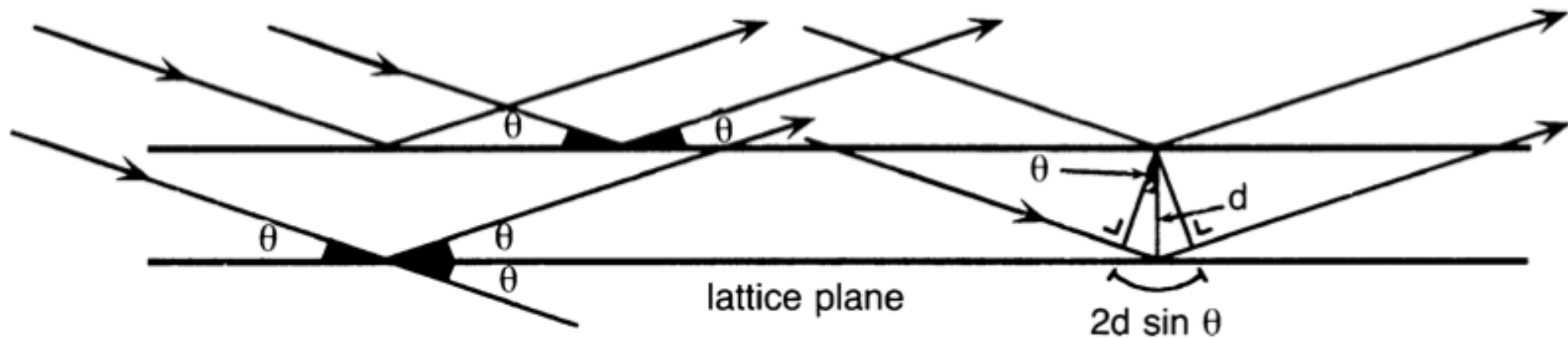
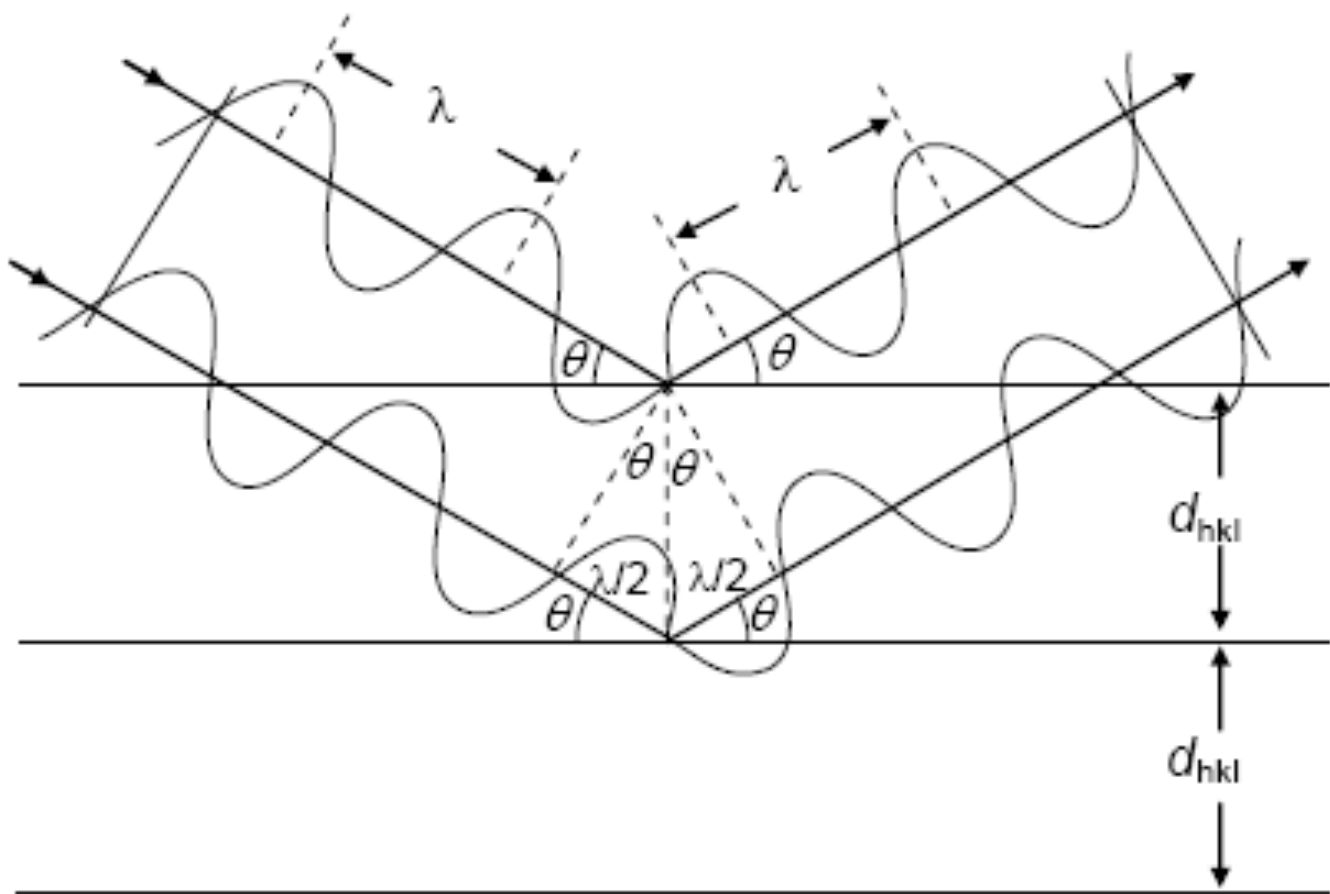


Figure 4.17. Two lattice planes are drawn separated by a distance d . The incident and the reflected beams make an angle θ with the lattice planes. Note that the beam is thus deflected through an angle of 2θ relative to its incident direction.

The incident and reflected beam make an equal angle with the plane (Figure 4.17). In a series of parallel reflecting planes (Bragg planes), the phase difference between the radiation from successive planes is 2π . The diffraction of X-rays by lattice planes can easily form the impression that only atoms on lattice planes contribute to the reflection. This is completely wrong! All atoms in the unit cell contribute to each reflection, atoms on lattice planes and in between. The advantage of lattice plane reflection and Bragg's law is that it offers a visual picture of the scattering process.



Reciprocal lattice and Ewald construction

There is a crystal lattice and a reciprocal lattice. The crystal lattice is real, but the reciprocal lattice is an imaginary lattice.

Question: What is the advantage of the reciprocal lattice?

Answer: With the reciprocal lattice, the directions of scattering can easily be constructed.

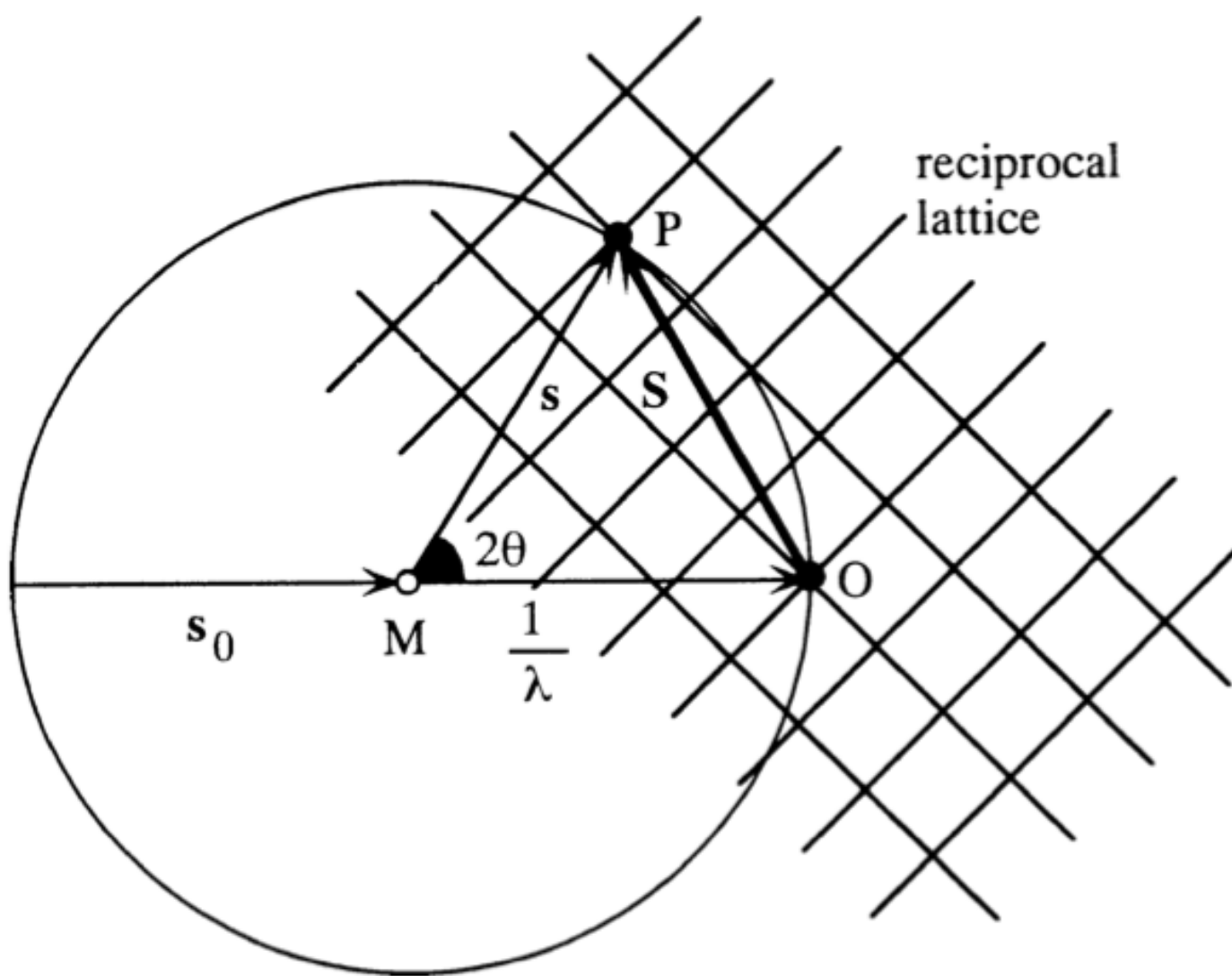
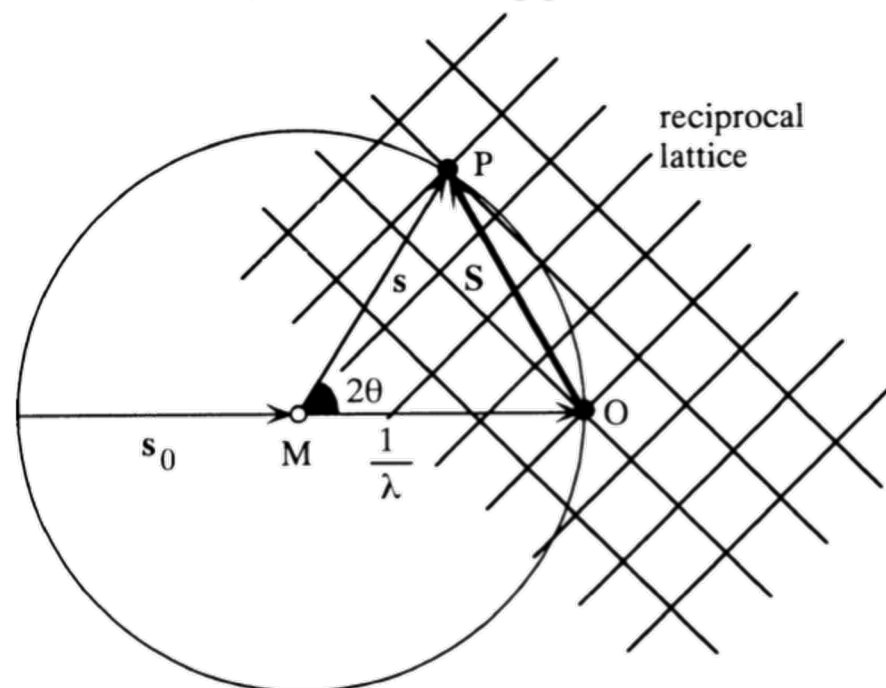


Figure 4.19. The Ewald sphere as a tool to construct the direction of the scattered beam. The sphere has radius $1/\lambda$. The origin of the reciprocal lattice is at O . s_0 indicates the direction of the incident beam; s indicates the direction of the scattered beam.

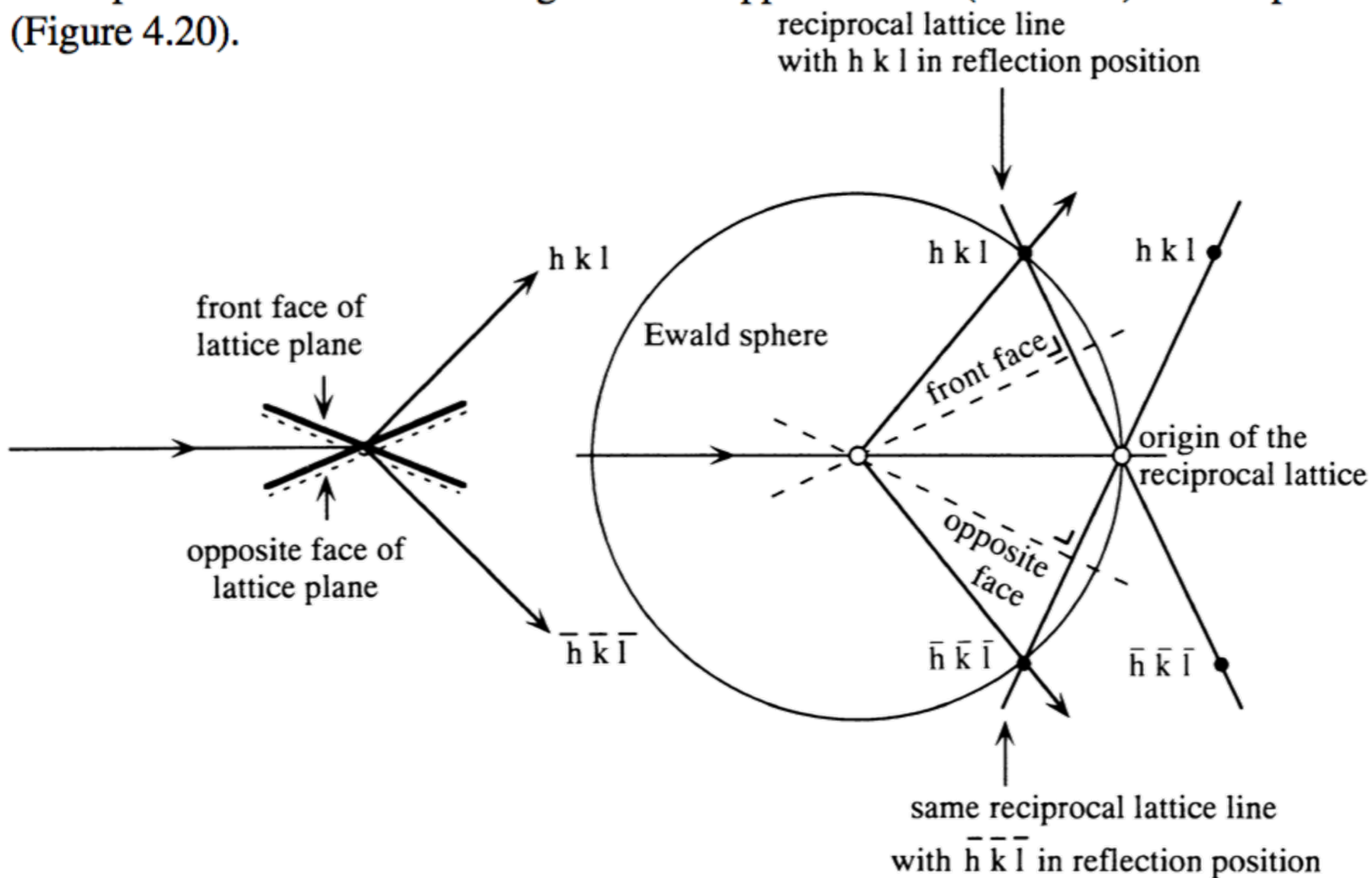
- The reciprocal lattice rotates exactly as the crystal does.
- The direction of the beam diffracted from the crystal is parallel to MP for the orientation of the crystal, which corresponds to the orientation of the reciprocal lattice.

From Figure 4.19, two properties of $\mathbf{S}(hkl)$ can easily be derived:

1. The reciprocal space vector $\mathbf{S}(hkl) = \mathbf{OP}(hkl)$ is perpendicular to the reflecting plane hkl , which is in agreement with the definition of \mathbf{S} in Section 4.3.
2. $|S(hkl)| = 2(\sin \theta)/\lambda = 1/d$ and Bragg's law is fulfilled.



One more comment on lattice planes: If the beam hkl corresponds to reflection against one face (let us say the front) of a lattice plane, then $(\bar{h}\bar{k}\bar{l})$ [or $(-h, -k, -l)$] corresponds to the reflection against the opposite face (the back) of the plane (Figure 4.20).



Temperature (B) factor

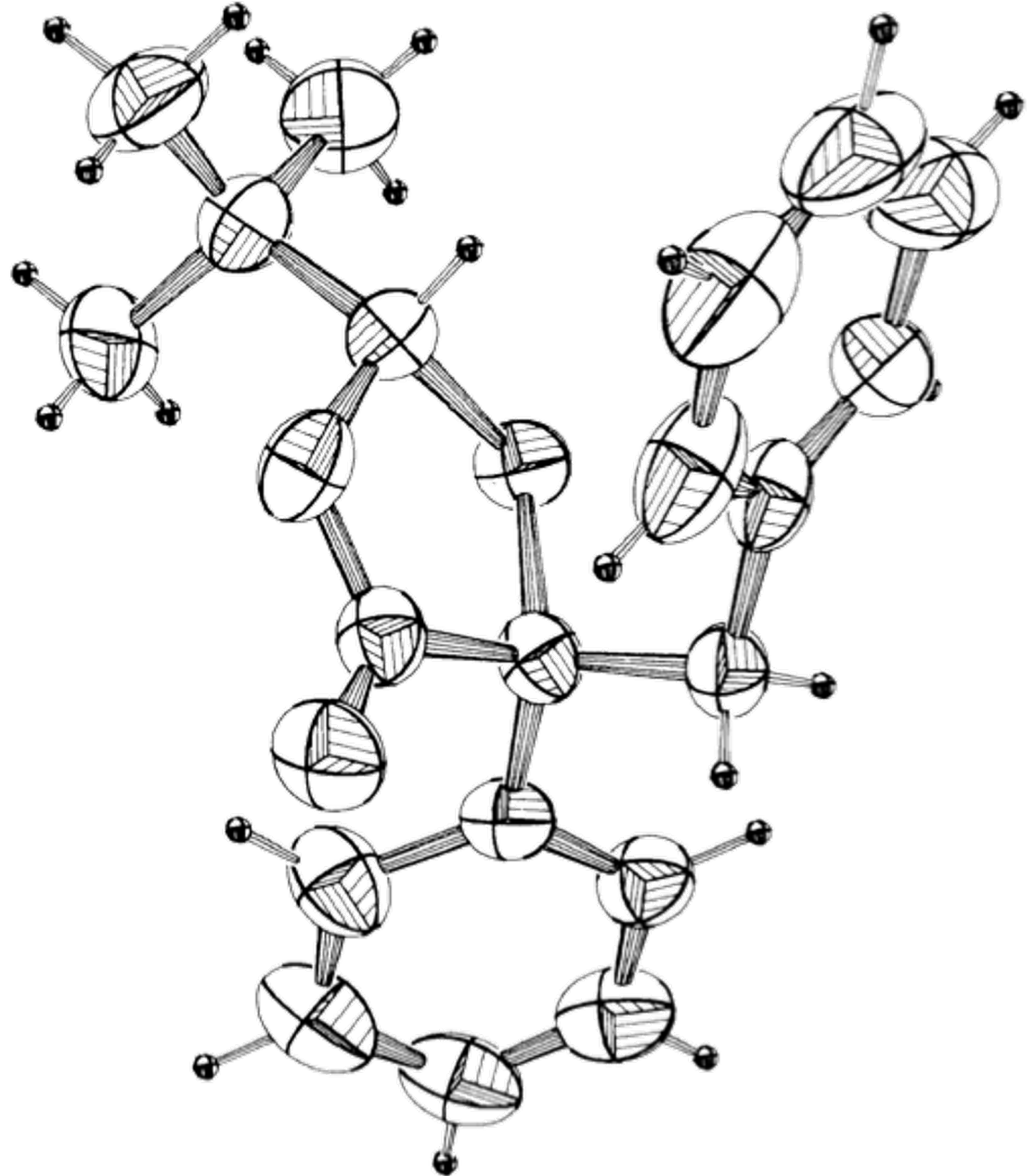


Figure 4.22. The plot of an organic molecule with 50% probability of thermal ellipsoids. (Reproduced with permission from Strijtveen and Kellogg © 1987 Pergamon Press PLC.)

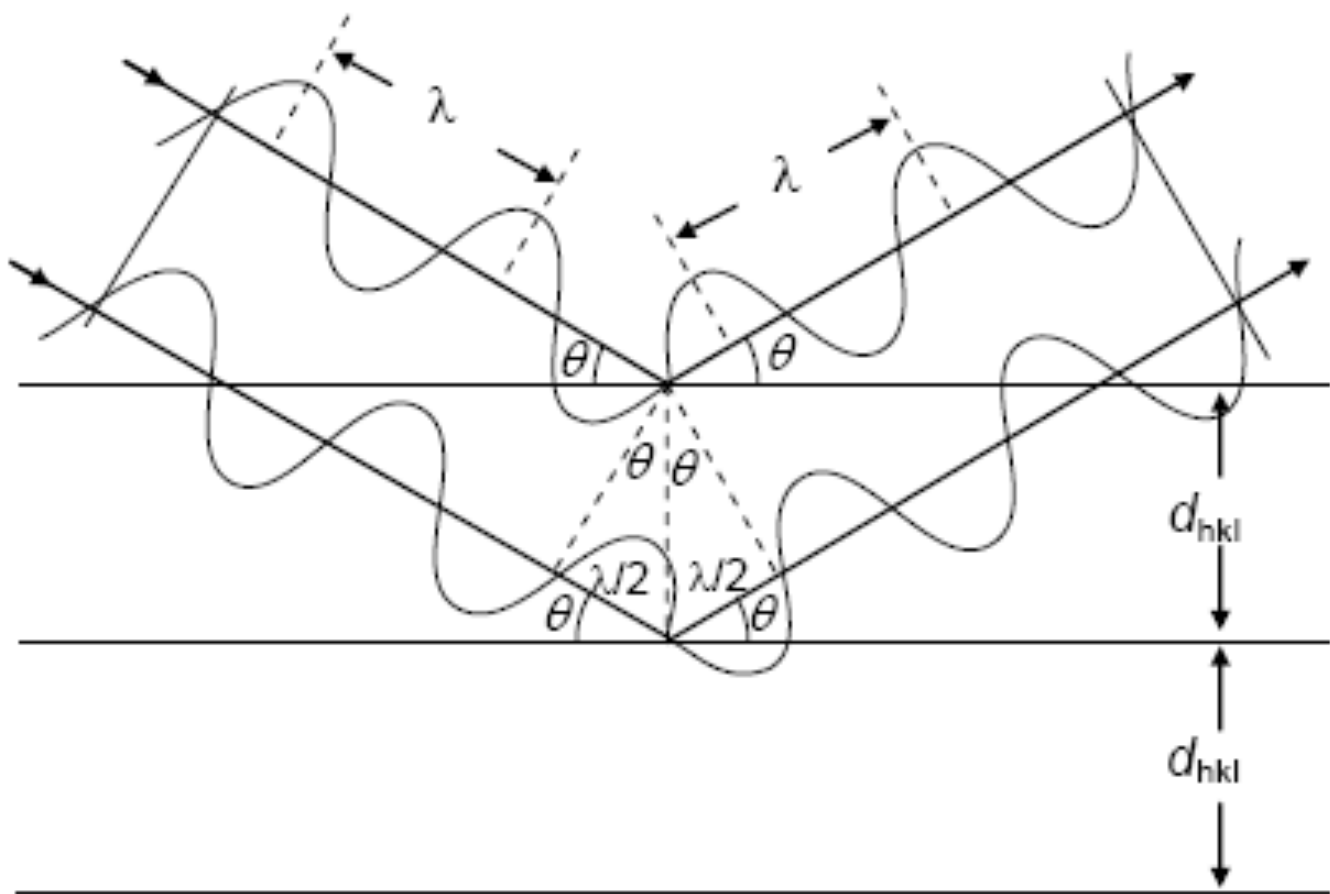
The vibration of an atom in a reflecting plane hkl has no effect on the intensity of the reflection (hkl). Atoms in a plane diffract in phase and, therefore, a displacement in that plane has no effect on the scattered intensity. The component of the vibration perpendicular to the reflecting plane does have an effect. In the simple case in which the components of vibration are the same in all directions, the vibration is called *isotropic*.

Then the component perpendicular to the reflecting plane and thus along \mathbf{S} is equal for each (hkl), and the correction factor for the atomic scattering factor is

$$\begin{aligned}
 T(\text{iso}) &= \exp \left[-B \frac{\sin^2 \theta}{\lambda^2} \right] = \exp \left[-\frac{B}{4} \left(\frac{2 \sin \theta}{\lambda} \right)^2 \right] \\
 &= \exp \left[-\frac{B}{4} \left(\frac{1}{d} \right)^2 \right].
 \end{aligned}
 \tag{4.6}$$

Assuming isotropic and harmonic vibration, it can be shown that the thermal parameter B is related to the mean square displacement \bar{u}^2 of the atomic vibration:

$$B = 8\pi^2 \times \bar{u}^2
 \tag{4.7}$$



Calculation of electron density

The structure factor is a function of the electron density distribution in the unit cell:

$$\mathbf{F}(\mathbf{S}) = \sum_j f_j \exp[2\pi i \mathbf{r}_j \cdot \mathbf{S}]. \quad (4.3)$$

$$\mathbf{F}(\mathbf{S}) = \int_{\text{cell}} \rho(\mathbf{r}) \exp[2\pi i \mathbf{r}_j \cdot \mathbf{S}] d\nu. \quad (4.8)$$

where $\rho(\mathbf{r})$ is the electron density at position \mathbf{r} in the unit cell. If x , y , and z are fractional coordinates in the unit cell ($0 \leq x < 1$; the same for y and z) and V is the volume of the unit cell, we have

$$d\nu = V \cdot dx dy dz$$

and

$$\begin{aligned} \mathbf{r} \cdot \mathbf{S} &= (\mathbf{a} \cdot x + \mathbf{b} \cdot y + \mathbf{c} \cdot z) \cdot \mathbf{S} = \mathbf{a} \cdot \mathbf{S} \cdot x + \mathbf{b} \cdot \mathbf{S} \cdot y + \mathbf{c} \cdot \mathbf{S} \cdot z \\ &= hx + ky + lz. \end{aligned}$$

Scattering by a unit cell

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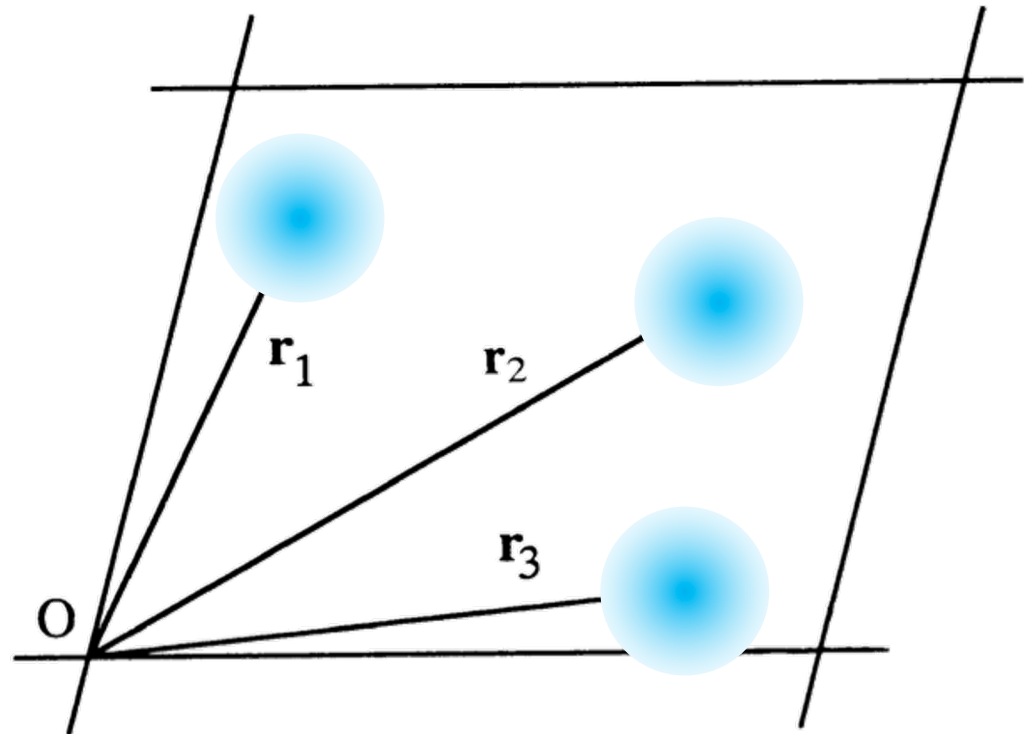
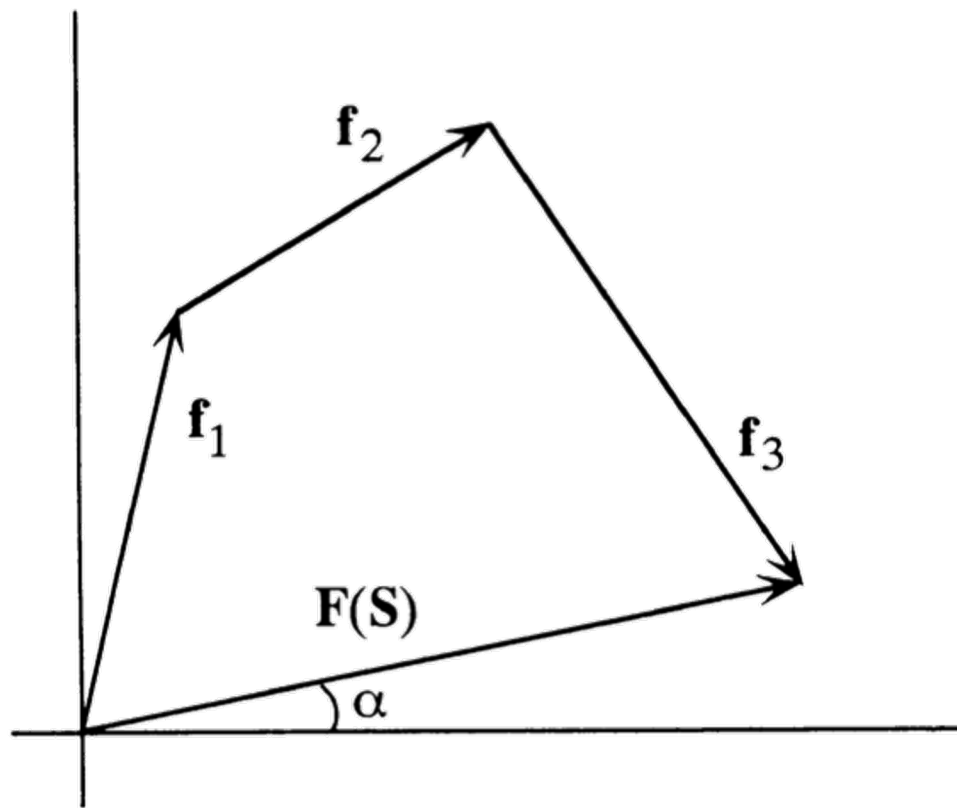


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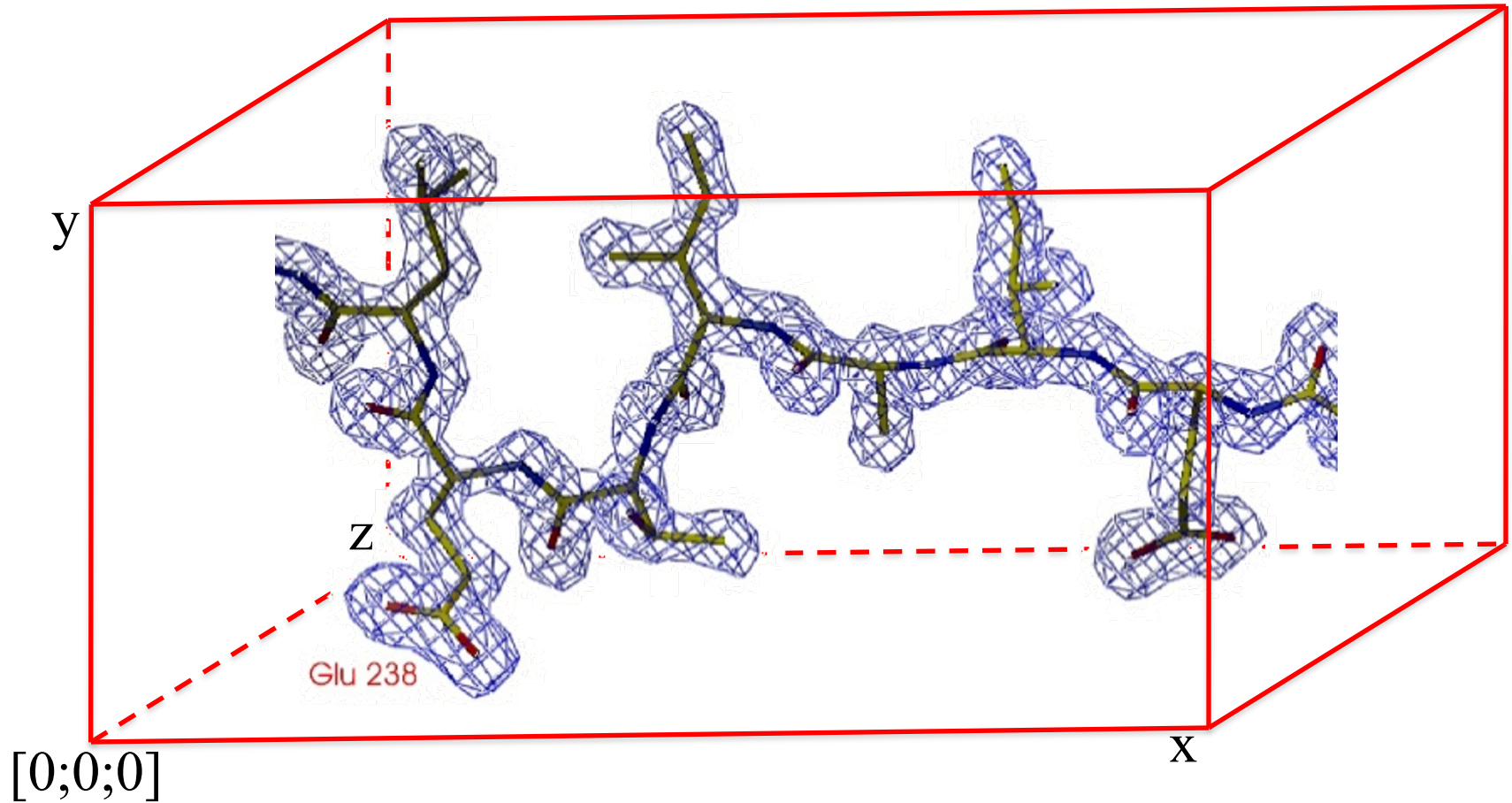
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and

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Information from X-ray diffraction experiment



$$\rho(x y z) = \frac{1}{V} \sum_h \sum_k \sum_l |F(h k l)| \exp[-2\pi i(hx + ky + lz) + i\alpha(h k l)]$$

$$\mathbf{F}(\mathbf{S}) = \int_{\text{cell}} \rho(\mathbf{r}) \exp[2\pi i \mathbf{r}_j \cdot \mathbf{S}] d\nu. \quad (4.8)$$

$$F(hkl) = V \int_{x=0}^1 \int_{y=0}^1 \int_{z=0}^1 \rho(xyz) \exp[2\pi i(hx + ky + lz)] dx dy dz. \quad (4.9)$$

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$F(hkl)$ is the Fourier transform of $\rho(xyz)$, but the reverse is also true: $\rho(xyz)$ is the Fourier transform of $F(hkl)$ and, therefore, $\rho(xyz)$ can be written as a function of all $F(hkl)$:

$$\rho(xyz) = \frac{1}{V} \sum_h \sum_k \sum_l F(hkl) \exp[-2\pi i(hx + ky + lz)]. \quad (4.10)$$

The Laue conditions tell us that diffraction occurs only in discrete directions and, therefore, in Equation (4.10), the integration has been replaced by a summation. Because $F = |F| \exp[i\alpha]$, we can also write

$$\rho(xyz) = \frac{1}{V} \sum_h \sum_k \sum_l |F(hkl)| \exp[-2\pi i(hx + ky + lz) + i\alpha(hkl)] \quad (4.11)$$

Notes

1. $\mathbf{F}(h k l)$ is the Fourier transform of the electron density $\rho(x y z)$ in the entire unit cell. Often the unit cell contains more than one molecule. Then $\mathbf{F}(h k l)$ is composed of the sum of the transforms of the separate molecules at position $(h k l)$ in reciprocal space.
2. Because of the crystallographic repeat of the unit cells, the value of the transform $\mathbf{F}(h k l)$ is zero in between the reciprocal space positions $(h k l)$. If there were no crystallographic repeat, the transform would be spread over the entire reciprocal space and its value is not restricted to reciprocal space positions $(h, k \ell)$.

$$F(hkl) = V \int_{x=0}^1 \int_{y=0}^1 \int_{z=0}^1 \rho(xyz) \exp[2\pi i(hx + ky + lz)] dx dy dz. \quad (4.9)$$

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Intensity diffracted by a crystal

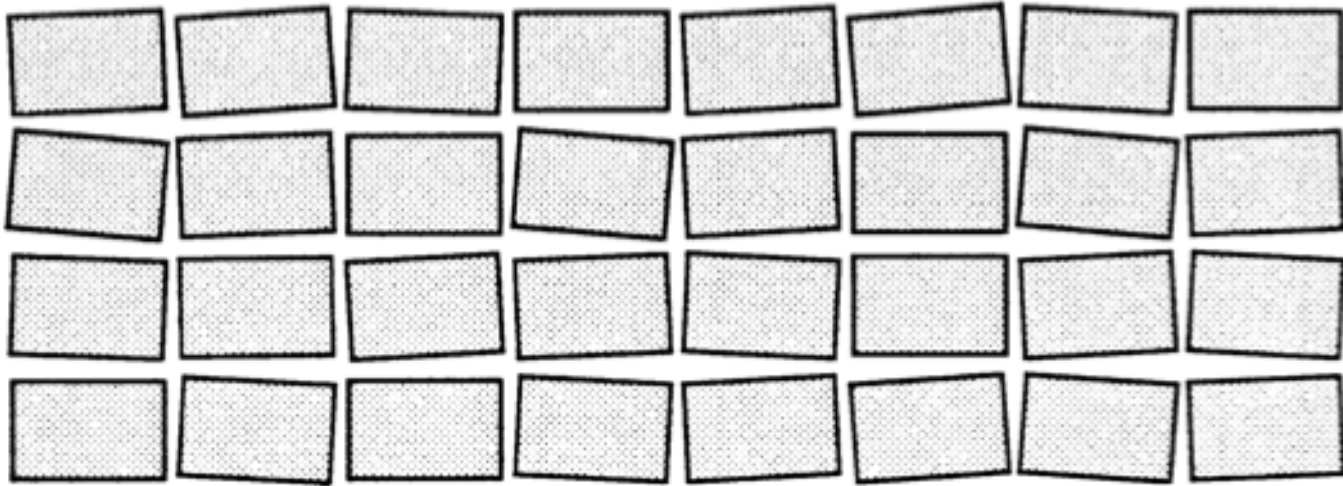


Figure 4.29. Most crystals are imperfect and can be regarded as being composed of small mosaic blocks.

We have the following assumptions:

1. Apart from ordinary absorption, the intensity I_0 of the incident beam is the same throughout the crystal.
2. The mosaic blocks are so small that a scattered wave is not scattered again (i.e., multiple scattering does not occur).
3. The mosaic blocks scatter independently of each other.

With these assumptions, the expression for I (int., $h k l$), if the crystal is rotated with an angular velocity ω through the reflection position, is

$$I(\text{int.}, h k l) = \frac{\lambda^3}{\omega \cdot V^2} \times \left(\frac{e^2}{mc^2} \right)^2 \times V_{\text{cr}} \times I_0 \times L \times P \times T_r \times |F(h k l)|^2 . \quad (4.32)$$

λ – wavelength

ω – angular velocity of crystal rotation

V – unit cell volume

e – electron charge

m – electron mass

c – speed of light

V_{cr} – crystal volume

I_0 – intensity of the excitation beam

L – Lorentz coefficient

P – polarization coefficient

T_r – transmission coefficient

$|F(hkl)|$ - structure factor amplitude

Effect of the unit cell size on the diffraction intensity

$$I(\text{int.}, hkl) = \frac{\lambda^3}{\omega \cdot V^2} \times \left(\frac{e^2}{mc^2} \right)^2 \times V_{\text{cr}} \times I_0 \times L \times P \times T_r \times |F(hkl)|^2$$

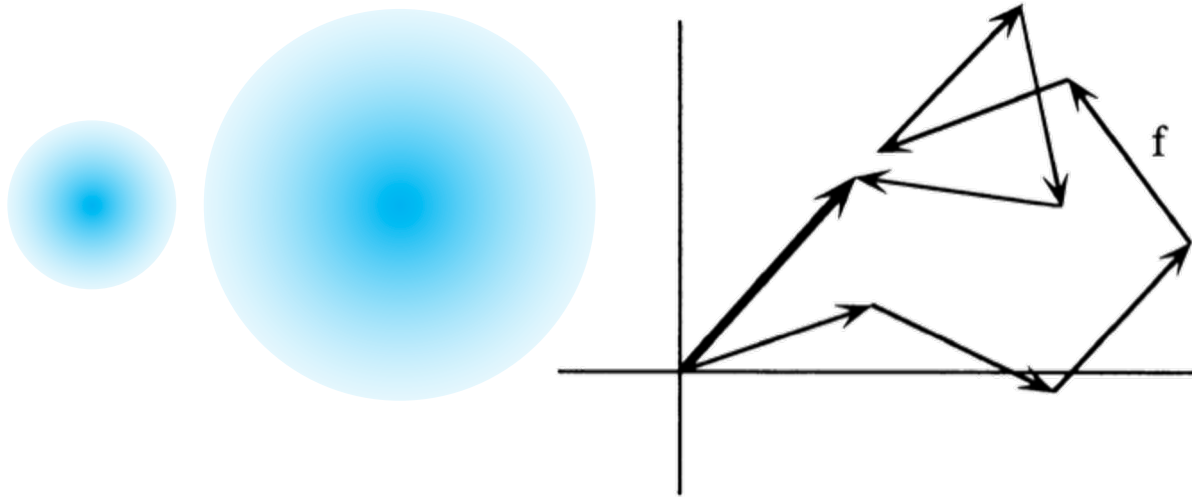


Figure 4.36. The displacement of a particle under the influence of Brownian motion. For n steps, where n is very large and each step has a length f , the final distance to the origin is $f\sqrt{n}$.

$$\sqrt{\overline{|F(hkl)|^2}} = f \times \sqrt{n} \quad \text{and} \quad \overline{|F(hkl)|^2} = f^2 \times n.$$

Combining the effect of the unit cell volume V and $|F(hkl)|$ in the scattering equation (4.32) leads to

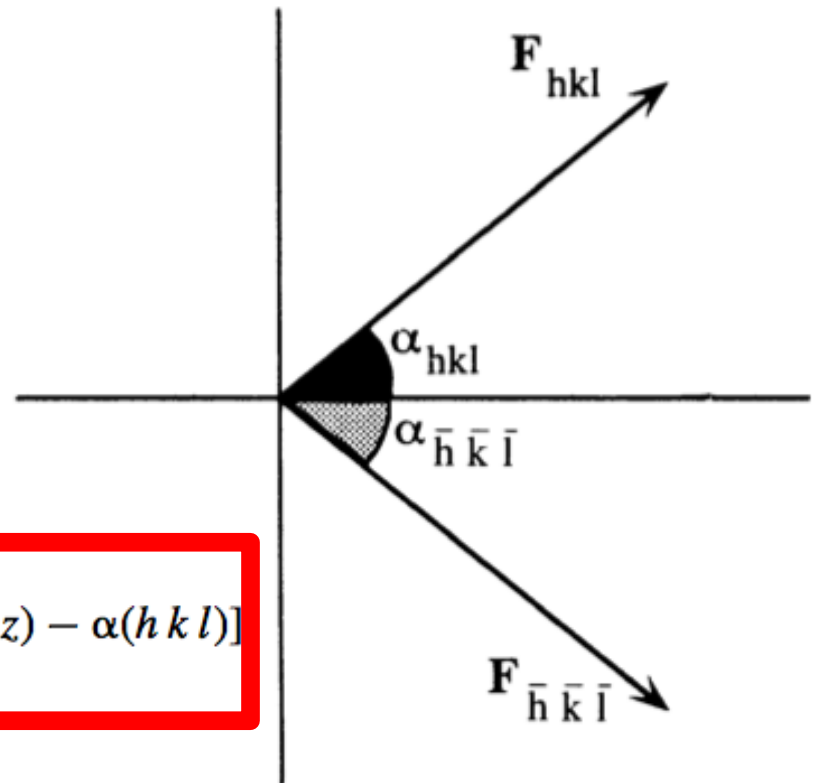
$$\overline{I(\text{int.}, hkl)} \text{ is proportional to } \frac{\overline{|F(hkl)|^2}}{V^2} = \frac{f^2}{V^2} \times n. \quad (4.36)$$

Friedel pairs

$$\mathbf{F}(h k l) = V \int_{\text{cell}} \rho(x y z) \exp[2\pi i(hx + ky + lz)] dx dy dz$$

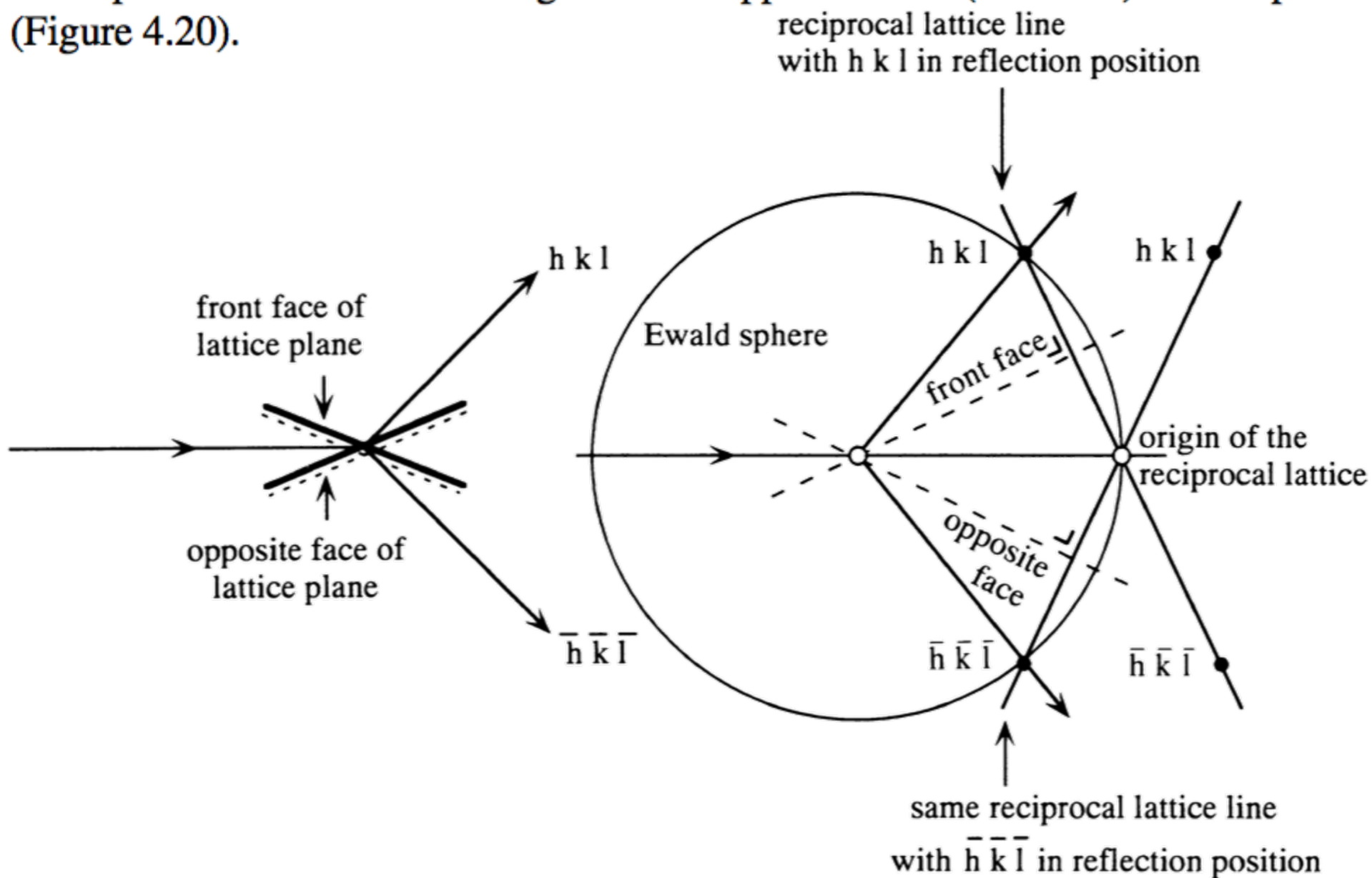
$$\mathbf{F}(\bar{h} \bar{k} \bar{l}) = V \int_{\text{cell}} \rho(x y z) \exp[2\pi i(-hx - ky - lz)] dx dy dz. \quad (4.25)$$

Figure 4.24. Argand diagram for the structure factors of the reflections $\mathbf{F}(h k l)$ and $\mathbf{F}(\bar{h} \bar{k} \bar{l})$.



$$\rho(x y z) = \frac{2}{V} \sum_{hkl=0}^{+\infty} |F(h k l)| \cos[2\pi(hx + ky + lz) - \alpha(h k l)]$$

One more comment on lattice planes: If the beam hkl corresponds to reflection against one face (let us say the front) of a lattice plane, then $(\bar{h}\bar{k}\bar{l})$ [or $(-h, -k, -l)$] corresponds to the reflection against the opposite face (the back) of the plane (Figure 4.20).



Symmetry in the diffraction pattern

4.12.1. A 2-Fold Axis Along y

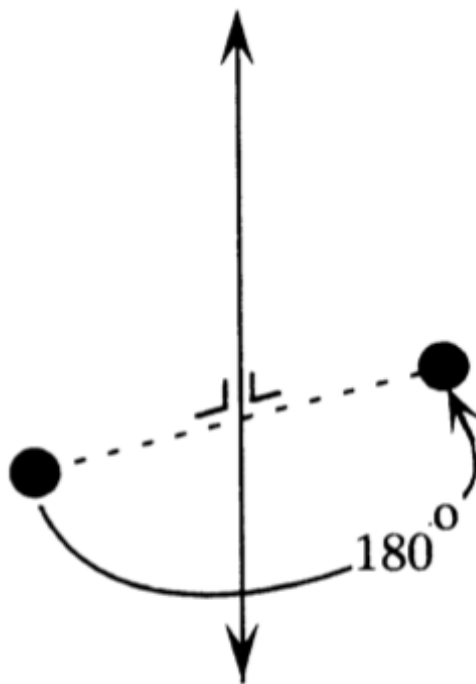
If a 2-fold axis through the origin and along y is present, then the electron density $\rho(x y z) = \rho(\bar{x} y \bar{z})$ (Figure 4.25). Therefore,

$$\begin{aligned} \mathbf{F}(h k l) = V \int_{\text{asymm}}^{\text{unit}} \rho(x y z) \{ \exp[2\pi i(hx + ky + lz)] \\ + \exp[2\pi i(-hx + ky - lz)] \} dx dy dz \end{aligned} \quad (4.26)$$

The integration in Eq. (4.26) is over one asymmetric unit (half of the cell), because the presence of the second term under the integral takes care of the other half of the cell.

$$\begin{aligned} \mathbf{F}(\bar{h} k \bar{l}) = V \int_{\text{asymm}}^{\text{unit}} \rho(x y z) \{ \exp[2\pi i(-hx + ky - lz)] \\ + \exp[2\pi i(hx + ky + lz)] \} dx dy dz \end{aligned} \quad (4.27)$$

It follows that $\mathbf{F}(h k l) = \mathbf{F}(\bar{h} k \bar{l})$ and also $I(h k l) = I(\bar{h} k \bar{l})$,



4.12.2. A 2-Fold Screw Axis Along y

For a 2-fold screw axis along y (Figure 4.26),

$$\rho(x y z) = \rho\{\bar{x}(y + 1/2)\bar{z}\}$$

term I ↓

$$\mathbf{F}(h k l) = V \int_{\text{asymm unit}} \rho(x y z) \{ \exp[2\pi i(hx + ky + lz)] + \exp[2\pi i(-hx + k(y + 1/2) - lz)] \} dx dy dz \quad (4.28)$$

term II ↑

term III ↓

$$\mathbf{F}(\bar{h} k \bar{l}) = V \int_{\text{asymm unit}} \rho(x y z) \{ \exp[2\pi i(-hx + ky - lz)] + \exp[2\pi i(hx + k(y + 1/2) + lz)] \} dx dy dz. \quad (4.29)$$

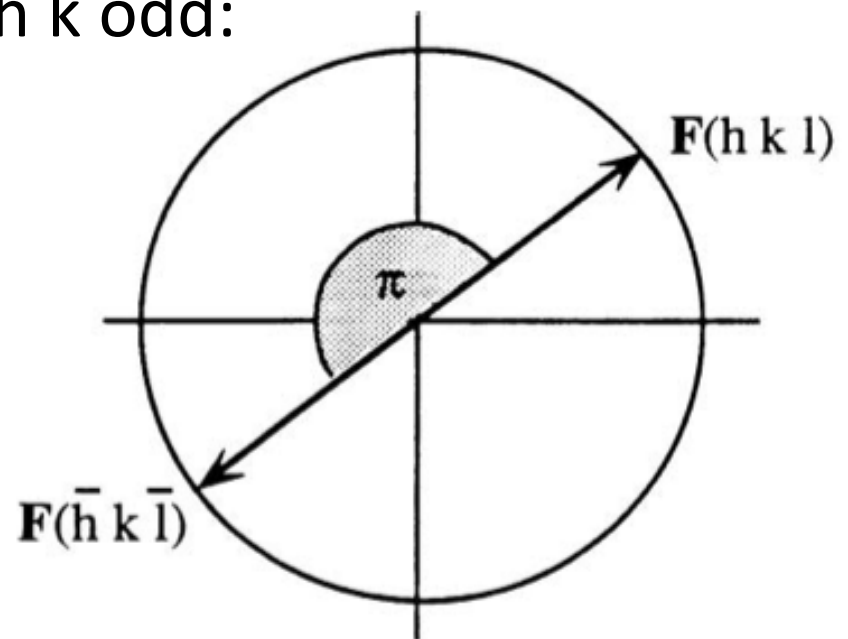
term IV ↑

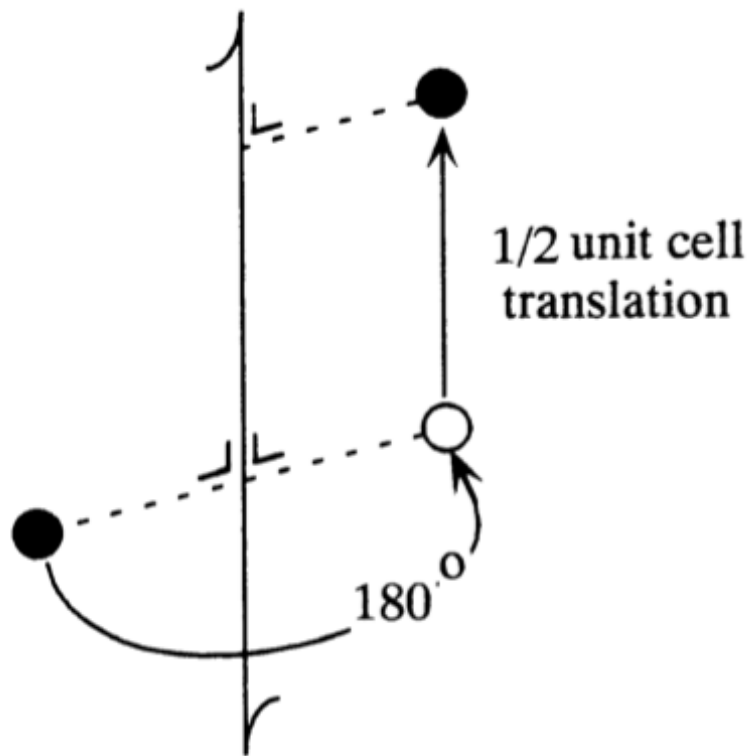
In Equation (4.28), term II is

$$\exp\{2\pi i[-hx + k(y + 1/2) - lz]\} = \exp[2\pi i(-hx + ky - lz + 1/2k)].$$

For k even, this is equal to term III in Equation (4.29). The same is true for term IV in Equation (4.29) and term I in Equation (4.28). Therefore, when k is even, $\mathbf{F}(h k l) = \mathbf{F}(\bar{h} k \bar{l})$ and $I(h k l) = I(\bar{h} k \bar{l})$. When k is odd, terms I and IV have a difference of π in their phase angles: $2\pi(hx + ky + lz)$ and $2\pi(hx + ky + lz + 1/2k)$.

For $\mathbf{F}(hkl)$ with k odd:





Systematic absences in P2(1)

$$\mathbf{F}(0k0) = V \int_{\text{asymm unit}} \rho(xyz) \{ \exp[2\piiky] + \exp[2\piik(y + 1/2)] \} dx dy dz. \quad (4.30)$$

When k is even, this is $2 \times V \int \rho(xyz) \exp[2\piiky] dx dy dz$. However, when k is odd, the two terms in Equation (4.30) cancel and $\mathbf{F}(0k0) = 0$

