Chapter 2

X-ray crystallography

Crystallography, the science concerned with the study of crystals, is a very old subject. It developed into a modern science, after the discovery of X-rays and their diffraction by crystals [1]. The important properties of X-rays were described by Röntgen, who discovered them in 1896. In 1912 M. von Laue, starting from an article by Ewald, a student of Sommerfield suggested the use of crystals as natural lattices for diffraction. This experiment was successfully performed by Friedrich and Knipping, both students of Laue. In 1913 W. L. Bragg and M. von Laue used X-ray diffraction patterns to deduce the structures of NaCl, KCl, KBr and KI.

2.1 Scattering of X-rays

The most powerful technique for investigating both the geometrical nature of the three dimensional array of molecules within a crystal and also the nature of the molecules themselves, is that of X-ray diffraction [2]. During the twentieth century, different types of diffraction have been found. Neutrons and electrons, which would be considered particles according to classical mechanics, can be diffracted[3]. These results complement those from X-ray diffraction studies, because mechanisms for the scattering of X-rays, neutrons and electron by atoms are different. X-rays are scattered by electrons, while neutrons are scattered by atomic nuclei. Neutron diffraction is important for the determination of the locations of light atoms like hydrogen atoms, which because of their low electron count are poor X-ray scatterers. Electron diffraction[4], while requiring much smaller crystals and therefore being potentially useful for the study of macromolecules, produces diffraction patterns that are more complicated. Their interpretation is hampered by the fact that the diffracted electron beams are re-diffracted within
the crystal much more than are X-ray beams. This has limited the practical use of electron diffraction in the determination of atomic arrangements in crystals to studies of surface structure.

Dorothy Hodgkin,[5] who obtained the Nobel Prize in 1965 for her X-ray diffraction work, wrote, "a great advantage of X-ray analysis as a method of chemical structure analysis is its power to show some totally unexpected and surprising structure with, at the same time, complete certainty".

The interaction of X-rays with electrons essentially occurs either by Thomson scattering or Compton scattering.

### 2.1.1 Thomson scattering

The physical explanation given for diffraction by atoms is this: when the X-rays hit an atom, the rapidly oscillating electric field of the radiation sets the electrons of the atom into oscillation about their nuclei. This oscillation has the same frequency as that of the incident radiation. The result is that the electron acts as an oscillating dipole that serves as a source of secondary radiation with the frequency of the incident beam. The effect of an incident radiation on an assembly of electrons ensures that the phase relationship between waves scattered from different electrons is well defined. Hence this is referred to as coherent scattering. Thomson scattering is often referred to as coherent scattering.

### 2.1.2 Compton scattering

The Compton effect requires a quantum mechanical explanation unlike the Thomson scattering which was dealt with as a wave-particle interaction. The model used to describe Compton scattering considers the collision between an electron and a quantum of electromagnetic radiation called a Photon. This may be considered as a particle-particle collision. Using this model, the scattered photons maybe characterised by two wavelengths just as observed experimentally. The Compton calculation uses a particle-particle collision model, therefore rejects the concept of phase. This implies that when we consider the combined effect of Compton scattering on an assembly of electrons, there are no phase relationships between the scattering events. Consequently, the Principle of Superposition cannot be used to assess the overall scattering effect of an assembly of electrons. Compton scattering is also referred to as incoherent scattering.

In a crystal no discrete diffraction effects result from Compton scattering. The sole effect of Compton scattering is to give rise to an overall background scattering, which has no direct influence on the diffraction pattern as such. The diffraction pattern of a crystal may be explained solely by the coherent Thomson scattering by electrons within the crystal.
2.2 Diffraction of X-rays by crystals

Crystals are solids characterised by long-range three-dimensional order. A crystal can be conceptually regarded as

\[ \text{crystal structure} = \text{lattice} \ast \text{motif} \]

where, the **motif** is the structural unit which is repeated regularly in space and **lattice** is a conceptual array of points in space which serves to define the geometrical relationship between the motifs in a structure. The symbol \( \ast \) implies convolution of the lattice with motif. The two-dimensional **Plane lattices** and three-dimensional **Space lattices** are defined in terms of **Crystallographic unit vectors** and **Crystallographic unit cells**.

The conventional **Crystallographic unit cell** is a parallelepiped defined by the space lattice of a crystal which serves to display the symmetry of the lattice in a convenient manner. The conventional **Crystallographic unit vectors** are a set of three non-coplanar lattice vectors which define the conventional unit cell. The directions defined by such crystallographic unit vectors specify the **Crystallographic axes**.

Within a single unit cell of a crystal, the atoms are distributed in a certain manner, and so there exists some electron density function \( \rho(r) \) which describes the location of the electrons within the atoms of the unit cell. The electron density function will have a large value at positions corresponding to atomic nuclei, and a relatively low value between nuclei. If we can find the electron density function, then we may infer the positions of atoms from the electron density maxima and derive data such as that pertaining to bond lengths and bond angles. All the fundamental structural information of the contents of the unit cell is contained within the electron density function, and it is this function which we wish to determine from X-ray analysis.

It is the three-dimensional ordering of the crystalline state which allows us to obtain clear diffraction data, giving us readily understandable information on the nature of the crystal and molecules which it contains. Waves can contain directional information only through their wave vectors \( \mathbf{k} \), implying that the diffraction pattern may be represented as a function of \( \mathbf{k} \), \( F(\mathbf{k}) \).

If the structure of the obstacle is represented by the amplitude function \( f(r) \), it is found that the diffraction pattern function \( F(\mathbf{k}) \) and the amplitude function \( f(r) \) are Fourier transform mates.

\[
F(\mathbf{k}) = \int f(r) e^{i\mathbf{k} \cdot \mathbf{r}} \, dr \\
f(\mathbf{r}) = \int F(\mathbf{k}) e^{-i\mathbf{k} \cdot \mathbf{r}} \, d\mathbf{k}
\]
The amplitude of the diffraction pattern resulting from the scattering of X-rays by a distribution of electrons characterised by an electron density function \( \rho(\mathbf{r}) \) is

\[
F(\Delta \mathbf{k}) = \int f_e \rho(\mathbf{r}) e^{i \Delta \mathbf{k} \cdot \mathbf{r}} d\mathbf{r}
\]

where, \( \rho(\mathbf{r}) \) represents the contents of the unit cell of a crystal, the limits of the integral correspond to those values of \( \mathbf{r} \) which define the unit cell. \( \Delta \mathbf{k} \) is the \textit{scattering vector}, which describes the change in direction which has occurred as a result of the diffraction. Since \( f_e \) is a constant we have,

\[
F(\Delta \mathbf{k}) = f_e \int \rho(\mathbf{r}) e^{i \Delta \mathbf{k} \cdot \mathbf{r}} d\mathbf{r}
\]

The ratio \( F(\Delta \mathbf{k})/f_e \) represents the relative scattering ability of the contents of the unit cell as compared to a single electron. This can be referred to as \( F_{\text{rel}}(\Delta \mathbf{k}) \). The relative intensity of the diffraction pattern of the unit cell is given by \( |F_{\text{rel}}(\Delta \mathbf{k})|^2 \).

In an X-ray diffraction experiment, we sample the diffraction pattern in space and obtain values of the time average of the intensity \( |F(\mathbf{k})|^2 \), corresponding to as many values of \( \mathbf{k} \) as we care to measure. Assuming that this allows us to find the corresponding values of \( F(\mathbf{k}) \), then we may obtain a complete set of \( F(\mathbf{k}) \) data.

We can then calculate \( f(\mathbf{r}) \) and the structure of the crystal is solved. It can be seen from the equation above that \( F(\mathbf{k}) \) is in general a complex quantity. Since we cannot measure complex numbers in any real experiment, the only measurable quantity is \( |F(\mathbf{k})|^2 \). \( F(\mathbf{k}) \) can be written in the amplitude-complex exponential form as:

\[
F(\mathbf{k}) = |F(\mathbf{k})| e^{i \delta}
\]

\( \delta \) is a phase factor. For any \( F(\mathbf{k}) \), about half the information is expressed in \( F(\mathbf{k}) \), and the other half in the phase factor \( \delta \). Thus the observed quantity \( |F(\mathbf{k})|^2 \) contains no information about \( \delta \). This ambiguity is known as the \textit{Phase Problem}.

2.2.1 Diffraction pattern

Information about the structure of a crystal is present in the diffraction pattern.

- The position of the main peaks gives information on the lattice.
- The shape of each main peak gives information on the overall object shape.
- The set of intensities of all the main peaks gives information on the structure of the motif.
2.2.2 Bragg's law and Ewald sphere

The phenomenon of X-ray diffraction can be treated as equivalent to cooperative reflection from planes of atoms. If X-rays are to be diffracted by a set planes, the incident ray should fall on the set of planes at an angle $\theta$ given by

$$\sin \theta = n\lambda/2d$$

where $n$ is an integer expressing the order of reflection from this set of planes whose interplanar spacing is $d$ and $\lambda$ is the wavelength of the incident X-rays. The above equation can be conveniently written as

$$\lambda = 2dhkl \sin \theta$$

where $n$ is contained in $dhkl$ and $(hkl)$ refers to the Miller indices of the reflecting planes. The spacing between the fictitious planes is $n$ times smaller than that between real planes, i.e.,

$$d_{hlkl} = n d_{hlkl}$$

The above equation is also referred to as Bragg's law.

One of the geometrical interpretations of Bragg’s law was propounded by the physicist P. P. Ewald. Since the wavelength of the radiation remains unchanged during diffraction, the locus of possible diffracted wave vectors will sweep out a sphere about a point $C$ which represents the position of the crystal. This is called the Ewald sphere or the Reflecting sphere.

Given a direct crystal lattice, a second imaginary lattice reciprocal to it can be built as follows: To every plane in the direct lattice a normal from the origin of the unit cell whose length is restricted to the reciprocal of the interplanar spacing can be drawn. If a point is placed at the end of each limited normal, then the collection of such points constitutes a new lattice called the reciprocal lattice. The concept of reciprocal lattice is very helpful in explaining the X-ray diffraction phenomenon. When a crystal is rotated in an X-ray beam, it can be assumed that the reciprocal lattice associated with it is also rotated. The Bragg’s law can be rearranged as

$$\sin \theta_{hlkl} = 1/d_{hlkl} = 2/\lambda$$

2.3 Instruments used

The experimental techniques in X-ray crystallography have come a long way in terms of quality, speed and accuracy of data collection. The instrumental setup consists of an X-ray source, a detector and a goniometer. The Facility at the Department of Studies in Physics, University of Mysore, Mysore, is equipped with two single crystal diffractometers viz.; The AFC7S Rigaku Single Crystal Diffractometer and DIPLabo Image Plate System.
2.3.1 AFC7S single crystal diffractometer

The AFC7S diffractometer is a Four Circle diffractometer. This is a mechanical assembly which permits total freedom of rotation about three mutually perpendicular axes, so that the crystal may be positioned with respect to the incident beam in any orientation. The angular position of the crystal with respect to the incident beam is specified by three Eulerian angles $\phi$, $\chi$ and $\Omega$, hence the name "Eulerian cradle". The incident beam and the photon counter are constrained to move only in the horizontal plane. The scattering angle $2\theta$ defines the position of the counter with respect to the straight-through beam. Since there are three circles which determine the position of the crystal, and one which specifies the location of the counter, it is called a four circle diffractometer. Figure 2.3.1 shows the schematic diagram of the four circle diffractometer. The operation of the instrument is to position the crystal in the incident beam so that the plane defined by the incident beam and diffracted beam under observation is horizontal. The counter is then positioned so as to detect the desired diffracted beam. The three coordinates $\phi$, $\chi$ and $\Omega$ of the crystal are read from the appropriate mounting circles, and the scattering angle $2\theta$ is measured from the counter circle. For each diffraction maximum the crystal position and the corresponding Bragg angle are recorded. The photon counter reading is fed directly into the computer where the intensity corrections are calculated. The procedure for data collection involves mounting a crystal on the diffractometer and physically centering it with the aid of the microscope. The MSC/AFC control package has an automatic data collection procedure. This option greatly reduces the amount of user time.

![Schematic diagram of a four circle diffractometer](image)
Figure 2.3.2: AFC7S Diffractometer.

Figure 2.3.3: Goniostat and X-ray tube of AFC7S.
required to collect a data set by sequentially performing all the necessary steps in an automatic fashion. The automatic procedure can be summed up thus:

1. **Search:** 25 Reflections are located by scanning through reciprocal space with the shutter open. When a reflection is found, accurate angular settings are determined by either half-slit centering algorithm or a $+\omega - \omega$ centering routine. Intensity profiles are plotted as each reflection is located. This gives an instant knowledge of the crystal quality. When half of the reflections have been found a preliminary indexing will occur.

2. **Index:** A primitive unit cell is determined from the centered peak positions. If the program is unable to find a reasonable unit cell that is consistent with all of the reflections, the crystal is more than likely non-single or twinned. A twin deconvolution routine is initiated automatically in such cases.

3. **Delaunay:** The unit cell of the highest metrical symmetry is determined and the unit cell parameters are transformed if necessary. The default cell reduction program is TRACER.

4. **Laue:** The implied symmetry of the unit cell is verified by measuring the intensities of symmetry equivalent reflections. A single $\psi$-scan can be performed at this point as a check for the effect of absorption and to verify crystal alignment. An empirical absorption curve is calculated and applied to the intensities when checking equivalent reflections. A statistical test is performed on the results to determine if the implied symmetry exists. If the symmetry is ambiguous then the program will stop so and the proper symmetry for data collection has to be set manually.

5. **Limits:** Data collection parameters are selected based on the observed Laue symmetry, the cell centering, and previously selected variables. The data collection speed can be selected based on the average intensity of the reflections found in **Search**. Standard reflections are selected based on both intensity and spatial distribution. The choice between $\omega$ or $\omega/2\theta$ scans is decided based on the size and centering of the unit cell.

6. **Precell:** This is an optional module that is used to collect high-angle data set. The data is collected using peak top measurements. A subset of strong reflections is selected and centered for the determination of a high-angle cell and orientation matrix.

7. **Collect:** Scan widths are determined by scanning the standard reflections. Data collection is performed in previously selected shells of $2\theta$ with routine orientation checking and measurements of standard reflections. Weak reflections are rescanned to accumulate counts thus improving intensity statistics.
The data output for each reflection contains calculated values of $F$ and $\sigma(F)$ for easy evaluation of the data as it is being collected.

8. **Cell:** An optional high-angle cell at the end of data collection can be obtained. The data set is scanned for the strong high-angle reflections and these reflections are subsequently centered and refined.

9. **Psi:** A list of reflections near $\chi = 90^\circ$ is generated and the relative intensities of these reflections are determined by a quick scan. Measurements of these reflections are made at multiple values of $\psi$ for the calculation of an empirical absorption curve.

The analysis and reduction of single crystal diffraction data is done using the teXsan software package.

Since the intensity maxima are measured either singly or in small groups, the time taken for the recording of the complete set of intensity data by a diffractometer is usually longer than if a photographic method (2-d detector) were employed. Also, this increases the probability of radiation damage to the crystal, and requires that the X-ray source is well stabilised so that the incident intensity is constant over the entire experiment.

The 2-d detectors are the most efficient in terms of measuring diffracted X-rays in all directions. Two dimensional detectors have been in use from the very beginning of X-ray diffraction studies, the year 1912. Initially 2-d detector was made of X-ray sensitive film. At present, electronic and IP (photoluminiscent Image Plate) detectors dominate.

### 2.3.2 DIPlabo image plate diffractometer

The imaging plate is essentially a storage phosphor. This means that the X-ray photons produce on the plate a latent image that is then excited by stimulation with a He-Ne laser. The photostimulable material covering the plate is Ba F Br:Eu$^{2+}$ crystals. An electronic transition in these ions generates the luminescence with an intensity proportional to that of the original X-rays. The storage phosphor is read by an image reader which releases the stored information by means of a laser and collects the emitted radiation and channels it into a photomultiplier tube which converts the radiation into an electrical signal. The plate can be used repeatedly, since exposure to visible radiation restores it to its initial condition.

The procedure for data collection involves mounting of the crystal and aligning it with the aid of a CCD. The mechanical assembly in this case comprises of a kappa goniostat. In this type of geometry the $\chi$ circle does not exist and the goniometer head carrying the crystal is mounted on an arm that can rotate about
the $\kappa$ axis, which forms an angle of $50^\circ$ with the main instrument axis as shown in figure 2.3.4. The advantage of this system is that it is less cumbersome than the Eulerian cradle and thus rotations that would cause collisions or would produce diffracted beams that would be blocked by the $\chi$ circle in the conventional four-circle diffractometer are still measurable. In the traditional approach, one collects the data first and then starts analyzing the results. This strategy has a risk that there may be a gross inefficiency in the setup of the experiment, for example: the data set may be incomplete, the reflections may overlap, the zones may overlap, a large percent of the reflections may be overloaded, etc. Data collection is therefore performed as a highly interactive process. Immediate data processing provides fast feedback during data collection. The image plate detector radius is 12 cm. The length and width of the image plate are 44.266 cm and 24.000 cm respectively. The oscillation range for data collection is set to $5^\circ$ by default.

The analysis and reduction of single crystal diffraction data consists of seven major steps. These are:

1. Visualization and preliminary analysis of the original, unprocessed, diffraction pattern.
2. Indexing of the diffraction pattern.
3. Refinement of the crystal and detector parameters.
4. Integration of the diffraction maxima.
5. Finding the relative scale factors between measurements.
2.3 Instruments used

Figure 2.3.5: DIPLabo Image Plate System.

Figure 2.3.6: Goniostat and X-ray tube of DIPLabo IP.
6. Precise refinement of crystal parameters using the entire data set.

7. Merging and statistical analysis of the measurements related by space group symmetry.

To analyze single crystal diffraction data collected with 2-d detectors several computer programs have been developed. The programs Denzo, XdisplayF and Scalepack together form the HKL package. Of these, Denzo and XdisplayF are used to perform the first four of the above mentioned processes. Scalepack is used for the tasks 5–7.

2.4 Data reduction

The procedure followed to extract relative structure factor amplitudes from raw integrated intensities is called data reduction. In data reduction, the different reflection dependent parameters are taken into account by multiplying the relative intensities by suitable correction factors.

The relative amplitude of the diffraction spot associated with the reciprocal lattice point \( hkl \) is given by the structure factor \( F_{hkl} \)

\[
F_{hkl} = \sum_j f_j e^{2\pi i(hx_j+kx_j+lz_j)}
\]

and the relative intensity \( I_{hkl} \) is

\[
I_{hkl} = |F_{hkl}|^2
\]

\( I_{hkl} \) represents the intensity of the scattering by a unit cell as compared to that by a single electron, and so \( I_{hkl} \) is a relative, and not an absolute measurement of intensity. In order to derive the correct value of \( I_{hkl} \) from the observed intensity data, we must apply certain correction factors that allow for various geometrical and physical factors viz., the Lorentz factor, the polarisation factor, absorption correction, anomalous scattering, extinction factor and correction for the thermal effects.

2.4.1 Lorentz factor

The Lorentz factor corrects for the different geometries of intersection of a reciprocal lattice point with Ewald sphere. The Lorentz factor \( L_{hkl} \) is defined as

\[
L_{hkl} = \frac{1}{\lambda \nu_\alpha} = \frac{1}{\sin \theta}
\]
2.4 Data reduction

where, $\lambda$ is the wavelength of the incident radiation, $\omega$ is the angular velocity with which the crystal rotates, $\bar{v}_n$ is the velocity component perpendicular to the Ewald sphere surface and $2\theta$ is the scattering angle appropriate to the diffraction maximum indexed as $hkl$.

Thus the Lorentz factor is due to the consideration of the normal velocity with which a reciprocal lattice point passes through the Ewald sphere.

2.4.2 Polarisation factor

The state of polarisation of the incident X-ray beam is dependent on the scattering angle, $\theta$ of the diffracted beam. The polarisation factor $P_{hkl}$ is given by,

$$P_{hkl} = \frac{1 + \cos^2 2\theta}{2}$$

where $\theta$ is the Bragg angle and the diffracting crystal is tacitly assumed to be ideally mosaic. This simple expression for the polarisation correction can be applied whenever the incident radiation is produced by a conventional source and monochromatised using an appropriate filter. The more general form of the polarisation correction for an incident beam monochromated with a crystal is

$$P_{hkl} = \frac{(\cos^2 2\theta \cos^2 \rho + \sin^2 \rho) |\cos^n 2\theta M| + \cos^2 2\theta \sin^2 \rho + \cos^2 \rho}{1 + |\cos^n 2\theta M|}$$

where $\theta$ is the Bragg angle of the reflection produced by the specimen and $\theta_M$ the angle of reflection of the monochromator crystal which was used to select the wavelength. $\rho$ is the angle between the projection of the normal to the reflecting plane on to a plane perpendicular to the incident monochromated X-rays and the plane of incidence[6]. When the original X-ray beam, the monochromated beam and the scattered beam all lie in the same plane this angle is equal to 0 and the polarisation factor takes the simpler form

$$P_{hkl} = \frac{\cos^2 2\theta |\cos^n 2\theta M| + 1}{1 + |\cos^n 2\theta M|}$$

The exponent $n$ depends on the characteristics of the monochromating crystal. If the crystal is assumed to be a perfectly mosaic it is equal to 2, whereas for an ideal crystal it is equal to 1.

2.4.3 Absorption corrections

According to Beer's law, absorption reduces the intensity of an X-ray beam travelling through a given material by an amount which depends on the material and
the length of the path travelled. The path lengths are dependent on the location of the point scattering the X-rays and on the incident and scattering angle, that is, on the reflection considered.

The intensity of the diffracted X-rays is thus reduced, with respect to what it would be without absorption by the factor

\[ \frac{I}{I_0} = e^{-\mu z} \]

which is valid for every point in the crystal. Here \( z \) is the total path length and \( \mu \) is the linear absorption coefficient of the crystal.

The linear absorption coefficient for the crystal can be calculated from the mass absorption coefficients of the atoms present in the unit cell. No structural knowledge is required, only the values of mass absorption coefficients of the elements(\( \mu_m \)) which can be found in the *International tables for X-ray crystallography* are sufficient. From the values of \( \mu_m \) for a given wavelength, \( \mu \) can be calculated by the following equation:

\[ \mu = \rho \sum_i g_i \mu_m^i \]

where \( g_i \) is the mass fraction of element \( i \) present in the unit cell, \( \mu_m^i \) is its mass absorption coefficient, and \( \rho \) is the crystal density.

The absorption correction may be determined either by analytical or by experimental procedures. The analytical methods require a careful measurement of the crystal size and shape and an estimation of the path length of X-ray for each Bragg reflection as it passes through the crystal. If the absorption is high (for large crystals or for crystal containing heavy metals), it is important to use a crystal that is as equidimensional as possible. Sometimes it is possible to grind a crystal to a spherical shape in order to minimise absorption effects. Another method of applying an absorption correction is to use an experimental procedure, an *Azimuthal scan* (\( \psi \)-scan). This is a scan of diffraction data measured as the crystal rotates about the diffraction vector. The intensities can be corrected for absorption as a function of the orientation of the crystal when the reflection under consideration was measured.

### 2.4.4 Extinction corrections

Extinction coefficient depends on the mosaic structure of the crystal and has two components. The most important one called, the *secondary extinction*, takes into account the fact that the lattice planes first encountered by the primary beam will reflect a significant fraction of the primary intensity and the deeper planes receive less primary radiation. This causes a weakening of the diffracted intensity, mainly observable for high intensity reflections at low \( \sin \theta/\lambda \) values in sufficiently perfect
crystals. If the mosaic blocks are misoriented then they do not diffract together and shielding of deeper planes is consequently reduced. Secondary extinction is equivalent to an increase of the linear absorption coefficient. Thus it is negligible for sufficiently small crystals.

The second component of the extinction coefficient, called *primary extinction*, takes into account the loss of intensity due to dynamic effects inside every single block. At the Bragg angle every incident wave can suffer multiple reflections from different lattice planes. After an odd number of reflections the direction will be the same as the diffracted beam. After an even number of reflections the direction will be the same as the primary beam. Each scattering causes a phase lag of $\lambda/2$. Consequently, destructive interference will result. The same consideration holds for waves propagating along the direction of the diffracted beam. The result is that both primary and diffracted beams are weakened because of dynamical effects.

Corrections for this effect are generally made after refinement, and involve finding a factor needed to adjust the most intense Bragg reflections so that they agree with those calculated (which are initially larger). Sometimes, these corrections are not physically reasonable and care should be exercised in their interpretation.

### 2.4.5 Anomalous dispersion

The extent that X-rays are absorbed by an atom normally increases slowly as the wavelength of the X-rays increases. Discontinuities in the amount of absorption are found at certain wavelengths. The absorption drops suddenly and then starts to rise again. These discontinuities are called absorption edges and occur at wavelengths that represent the energy necessary to excite a bound electron to a vacant higher energy level or to eject it all together. At wavelengths just below the absorption edge, i.e. at energies higher than the absorption edge, the scattering of X-rays becomes complex. This means that the scattering factor $f_j$ must be replaced by a complex value

$$f_j + \Delta f'_j + i \Delta f''_j$$

where $i = \sqrt{-1}$, $\Delta f'_j$ and $\Delta f''_j$ are called the real and imaginary dispersion corrections. $\Delta f'_j$ and $\Delta f''_j$ vary with the wavelength of the incident radiation. If an atom absorbs X-rays strongly, there will be a phase change (related to $\Delta f'_j + i \Delta f''_j$) for the X-rays scattered by the absorbing atom relative to the phases of the X-rays scattered by other atoms. This is called *anomalous scattering*. As a result, for a crystal in a non-centrosymmetric space group, the intensity of a Bragg reflection, $hkl$, is different from that of $-h, -k, -l$ Bragg reflection.

*Friedel's Law* states that $|F|^2$ values of centroymetrically related Bragg reflections $(h, k, l$ and $-h, -k, -l$) are equal. Thus, for a non-centrosymmetric
structure, this law does not apply if there is an anomalous scatterer in the structure. The Friedel law is satisfied if the structure is centrosymmetric.

2.4.6 Thermal effects

The assumption that the nuclei are fixed at single points is valid only at temperature near to absolute zero. This assumption is not true at room temperature. Approximate corrections need to be applied. As the temperature increases, the ambient thermal energy causes the atoms to oscillate about their equilibrium positions and so we cannot strictly assign point coordinates \((x_j, y_j, z_j)\) to the nucleus of any atom. Rather, \((x_j, y_j, z_j)\) should be taken as the average position of the nucleus of the \(j^{th}\) atom. Since the thermal motions are very rapid, we may represent their effect by associating the nucleus with a position characterised by \((x_j, y_j, z_j)\) but which in reality is somewhat blurred and not an ideal mathematical point. As the nucleus oscillates about its average position, the electrons associated with the atom also move, so the interaction of the electrons with the incident X-rays is changed.

The effect of thermal vibrations is to reduce the effective scattering power of an atom. If the atom is oscillating, then the expression for \(f_j\) must be modified to account of this. The temperature corrected atomic scattering factor \((f_j)_T\) is defined as:

\[
(f_j)_T = f_j e^{-B_j (\sin^2 \theta / \lambda^2)}
\]

in which \(\theta\) is the Bragg angle and \(\lambda\) is the wavelength of the incident radiation. \(B_j\) is called temperature factor, and its value depends on the atom under study and the temperature at which the diffraction event takes place. An implicit assumption in the form of the above equation is that the thermal vibrations of the atom \(j\) are spherically symmetric. The equation accounts for the thermal motion by introducing a negative exponential. The term \(\sin^2 \theta / \lambda^2\) is necessarily positive, and \(B_j\) is always positive. Since \(B_j (\sin^2 \theta / \lambda^2)\) must be dimensionless, the dimensions of \(B_j\) are \([\text{length}]^2\). The value of \(B_j\) is given by:

\[
B_j = \frac{8 \pi^2}{3} \langle u^2 \rangle
\]

where \(\langle u^2 \rangle\) represents the mean square displacement of the \(j^{th}\) atom. This equation shows that \(B_j\) is necessarily positive.

Hence, \((f_j)_T\) is always less than \(f_j\). When \(\theta\) is zero, \((f_j)_T\) equals \(f_j\). Also, the value of the exponential decreases quite rapidly as \(\theta\) increases causing \((f_j)_T\) to fall off sharply as compared to \(f_j\).
2.5 Structure solution

The goal of structural analysis is to obtain the distribution of electron density in the unit cell (atomic positions) starting from the diffraction data which is given by,

$$\rho(x, y, z) = \rho(x, y, z) = \frac{1}{V} \sum_{h} \sum_{k} \sum_{l} F_{hkl} e^{-2\pi i(hx + ky + lz)}$$

It is not possible to reach this goal in a unique and automatic way, because from the experimental data only the magnitudes, not the phases, of the structure factors are obtained. Therefore, in order to compute the electron density, we must somehow derive the missing information. The derivation of the correct values of the structure factors $F_{hkl}$, both magnitude and phase, from the relative intensities $|F_{hkl}|^2$ constitutes the Phase Problem.

There are two principal methods to tackle the phase problem. The first of these the chemical modification method involves the chemical modification of the molecules within the crystal. The second approach is a more mathematical one in which the relative intensity $|F_{hkl}|^2$ data are treated statistically and analytically in order to search for certain relationships such as inequalities, which have been theoretically predicted. This in turn is used to determine the phases $\alpha_{hkl}$. These mathematical techniques of solving the phase problem are called Direct methods.

Two important properties of the electron density function that are helpful in obtaining the phase information are:

- it is everywhere positive, i.e. $\rho(r) \geq 0$ (positivity).
- it is composed of discrete atoms (atomicity).

Possible phase angles are constrained by these two conditions, so that relative phase determination hinges on the mathematical expressions for Fourier series. In the total Fourier synthesis involving $F_{hkl}$ with the correct value of $\alpha_{hkl}$, these two conditions should apply.

2.5.1 Development of phase information

The methods used to ensure that the Fourier summation does not give a negative electron-density map are mathematical in nature. In order to generalise the mathematics, a quantity unitary structure factor is defined as,

$$U_{hkl} = \frac{F_{hkl}}{\sum_j F_j}$$
Further, unitary atomic scattering factor, \( n_j \) is defined as,

\[
n_j = \frac{f_j}{\sum_j f_j}
\]

Therefore

\[
U_{hkl} = \sum_j n_j e^{2\pi i(hx_j + ky_j + lz_j)}
\]

which is analogous to the expression for \( F_{hkl} \).

Another useful quantity is the normalised structure factor \( E_{hkl} \), which is defined as,

\[
E_{hkl} = \frac{U_{hkl}}{\langle |U_{hkl}|^2 \rangle^{1/2}}
\]

where

\[
\langle |U_{hkl}|^2 \rangle = \frac{1}{N} \sum_{hkl} |U_{hkl}|^2
\]

in which the summation is over the \( N \) values of \( |U_{hkl}|^2 \) corresponding to all reciprocal lattice sites, \( hkl \). \( \langle |U_{hkl}|^2 \rangle \) therefore represents the average value of \( |U_{hkl}|^2 \).

The direct methods may be classified according to the means by which structure factor magnitudes are compared as follows:

- Inequalities
- Equalities
- Probabilities

### 2.5.2 Inequalities

The Harker-Kasper inequalities were the first to be introduced. These are based on the symmetry of the crystal. In a centrosymmetric crystal, the phase problem reduces to the allocation of the appropriate sign to each structure factor. By making use of the Cauchy inequality, a relation between the unitary structure factors is obtained as follows:

\[
U_{2h 2k 2l} \geq 2U_{hkl}^2 - 1
\]

This equation relates information concerning the reciprocal lattice site \( h k l \) to that for the site \( 2k 2h 2l \), and if the intensity at \( hkl \) is strong enough, then the sign of \( U_{2h 2k 2l} \) may be unambiguously deduced. In general, they give unambiguous information only if there are a relatively large number of intensity maxima associated with unitary structure factors of about one half or greater. Another limitation is that the use of inequalities is restricted to centrosymmetric crystals.
2.5 Structure solution

2.5.3 Equalities

The equality relationships refer only to centrosymmetric crystals. Since only the signs of the structure factors are to be determined, we may represent the sign associated with the structure factor $F_{hkl}$ by $s(hkl)$. By considering a structure of identical, resolved (i.e., non-overlapping) atoms, Sayre derived the following relationships between the signs of three reflections indexed as $hkl$, $h'k'l'$ and $h + h' k + k' l + l'$:

$$s(hkl) = s(h'k'l') \cdot s(h + h' k + k' l + l')$$
$$s(h + h' k + k' l + l') = s(hkl) \cdot (h'k'l')$$

Thus, knowledge of the signs $s(hkl)$ and $s(h'k'l')$ permit the unambiguous determination of the sign $s(h + h' k + k' l + l')$. These relationships are true only for the hypothetical case of a structure composed of identical atoms. For organic crystals, the predominant components carbon, oxygen and nitrogen have about the same atomic weight and hence above equations have been shown to hold if the associated structure factors are large.

2.5.4 Probabilities

The use of probabilities as opposed to equalities is a more general approach although subject to errors. But if a set of signs can be identified, the errors in the electron density may can be removed by computing the Fourier synthesis. For a general centrosymmetric crystal the equation is as follows:

$$s(hkl) \cdot s(h'k'l') \cdot s(h + h' k + k' l + l') \approx 1$$

One of the problems, however, associated with the use of probabilities is that they can give the signs of a certain structure factor only if those associated with two others are already known.

The problem is more difficult for the non-centrosymmetric crystals. However, the tangent formula is applicable to these cases and has been widely used. If the phase angle associated with the structure factor $F_{hkl}$ is represented for brevity as $\alpha_h$, then

$$\tan \alpha_h \approx \frac{\sum_{h'} |E_{h'}||E_{h-h'}| \sin(\alpha_h + \alpha_{h-h'})}{\sum_{h'} |E_{h'}||E_{h-h'}| \cos(\alpha_h + \alpha_{h-h'})}$$

The estimated phase is $\alpha_h$ is thus the average taken over all the triplet reflections involving $h'$. 
2.5.5 Patterson methods

The Patterson function is a map that indicates all the possible relationships (vectors) between the atoms in a crystal structure. The Patterson map, \( P(uvw) \), is a Fourier synthesis that uses the indices, \( h, k, l \) and the square of the structure factor amplitude, \( |F_{hkl}|^2 \), of each diffracted beam.

\[
P(uvw) = \frac{1}{V} \sum_{h} \sum_{k} \sum_{l} |F_{hkl}|^2 e^{-2\pi i(hu + kv + lw)}
\]

This equation is same as the equation for electron density, but there is no phase angle in the expression. The coefficients of Patterson function are the observed intensities. As the Patterson function uses coefficients that are the squares of the structure factor expression, only the cosine terms of the structure factor are conserved and no phases are needed.

\[
P(uvw) = \frac{1}{V} \sum_{h} \sum_{k} \sum_{l} |F_{hkl}|^2 \cos 2\pi i(hu + kv + lw)
\]

The Patterson function can also be described as the Fourier transform of \( F_{hkl}^2 \).

If the electron density is known, the Patterson function at any point \( u, v, w \) may be calculated by multiplying the electron density at a point \( x, y, z \) with that at \( x + u, y + v, z + w \). Doing this for all values of \( x, y \) and \( z \) and summing the products

\[
P(uvw) = \int \rho(xyz) \rho(x + u, y + v, z + w) \, dV
\]

The Patterson function is the convolution of the electron density at all points \( x, y, z \) in the unit cell with the electron density at points \( x + u, y + v, z + w \). A peak at \( u, v, w \) in the Patterson map represents a vector from the origin of the Patterson function to the point \( u, v, w \). This means that if any two atoms in the unit cell are separated by a vector \( u, v, w \) then there will be a peak in the Patterson map at \( u, v, w \). This peak represents the vector between the two atoms, at \( x_1, y_1, z_1 \) and \( x_1 + u, y_1 + v, z_1 + w \). The important feature is that since there is a peak at this location in the Patterson map, then, within the actual arrangement of atoms in the crystal structure, there are at least two atoms separated by this particular vector. The height of a peak in a Patterson map is approximately proportional to the products of the atomic numbers, \( Z_iZ_j \), of the atoms at the two ends of the vector. The Patterson map has a very high peak at the origin representing a vector between each atom and itself.

Peaks of the Patterson function occur centrosymmetrically about the origin, and the width of each peak is equal to the sum of the widths of two relevant electron density peaks. There will be \( N(N - 1) \) peaks at points other than the
origin for any real unit cell containing \( N \) atoms. Since each Patterson peak is wider than the electron density peaks, it is usually unlikely for these \( N(N - 1) \) Patterson peaks to be separately resolved. Lines or planes within the Patterson map which contain local concentrations of peaks are called *Harker lines* or *Harker planes*, and identification of these is often useful in obtaining the symmetry information in addition to that derived from diffraction pattern and recognition of systematic absences.

### 2.6 Structure refinement

The structural models obtained both by Patterson and direct methods are often incomplete and in most cases they represent a crude first approximation of the real structure. The inaccuracies inherent in the observed structure factor magnitudes and phases, coupled with series termination errors, imply that the first derived electron density map is not an accurate representation of the true structure. The process of obtaining atomic parameters that are more precise than those obtained from an initial model, referred to as *refinement of the crystal structure*, is an essential part of any crystal structure analysis.

The positions \( x, y \) and \( z \) and the atomic displacement (thermal) parameters \( B \), derived for each atom in a preliminary crystal structure are adjusted so as to improve the agreement between the observed structure amplitudes, \( |F_{hkl}|_o \) and those calculated from the determined model, \( |F_{hkl}|_c \). The progress in improving this agreement is usually monitored by a residual index, \( R \), defined as

\[
R = \frac{\sum_{\text{all } hkl} |F_{hkl}|_o - |F_{hkl}|_c}{\sum_{\text{all } hkl} |F_{hkl}|_o}
\]

The \( R \) value is calculated as a measure of the precision of the results of the refinement. If \( R \) value is high, the derived structure is most likely incorrect. The refinement procedure takes place in four stages:

- **The derivation of the trial structure**: This is deduced from the first calculated electron density function using the observed structure factor magnitudes with the observed phases.

- **Cyclic Fourier refinement**: A set of structure factors including the thermal effects are obtained from the trial model. A new Fourier synthesis using the observed structure factor magnitudes with the calculated phases is computed. This procedure is repeated until a model which reproduces itself is obtained. The cyclic Fourier refinement process primarily gives us better values of the phases and allows us to draw an electron density map with more accurately located atomic sites.
• **Difference Fourier synthesis:** Having obtained a more reliable set of phases, a Fourier synthesis using the quantities \(|F_o| - |F_c|\) is calculated. This gives a difference Fourier map which refines the correct model. Also, this enables the identification of missing atoms and refinement of thermal parameters.

• **Least squares refinement:** This is a statistical treatment of data such that a model which represents the best fit with the observed data is obtained. In the most sophisticated treatment, three positional and six thermal parameters are fitted for each atom.

In order to get meaningful values of the estimated standard deviations of the varied parameters, the observational equations should be properly weighted. Several weighting functions have been proposed and first scheme suggested by Hughes [7] was:

\[
w = \text{constant} = 1/16F_{\text{min}}^2 \quad \text{for } |F_o| \leq 4F_{\text{min}}
\]

\[
w = 16F_{\text{min}}^2 / |F_o|^2 \quad \text{for } |F_o| > 4F_{\text{min}}
\]

where \(F_{\text{min}}\) is close to the minimum observed amplitude. Later Cruickshank [8] proposed the scheme

\[w = 1/(a_0 + |F_o| + a_2|F_o|^2)\]

with \(a_0 = 2F_{\text{min}}\) and \(a_2 = 2/F_{\text{max}}\) as initial guesses.

The weighting function available in the SHELX [3] program is:

\[w = 1/[\sigma^2(F_o^2) + (aP)^2 + bP]\]

where \(P = [2F_o^2 + \max(F_o^2, 0)]/3\).

The main disadvantage of refinement against \(F^2\) is that \(R\) indices based on \(F^2\) are larger than those based on \(F\). In SHELXL a conventional index \(R_1\) based on observed \(F\) values larger than \(4\sigma(F_o)\) is also calculated.

\[wR2 = \sum [w(F_o^2 - F_c^2)^2] / \sum [w(F_o^2)^2]^{1/2}
\]

\[R1 = \sum ||F_o| - |F_c|| / \sum |F_o|
\]

The Goodness of fit is always based on \(F^2\) and is defined as

\[GooF = S = \sum [w(F_o^2 - F_c^2)^2] / (n - p)^{1/2}\]

where \(n\) is the number of reflections and \(p\) is the total number of parameters refined.
2.7 Structure: constitution, configuration and conformation

X-ray crystallography produces a detailed description of the spatial arrangement of atoms in molecules.

Molecular constitution indicates the way in which atoms are interconnected, making due distinction between single and multiple bonds.

The configurations of a molecule of given constitution indicate all possible spatial arrangements of its atoms, ignoring those derived from rotation around single bonds.

The word conformation refers to the different spatial arrangements of a molecule of given configuration produced by rotation (torsion) around single bonds.

The term structure includes the constitutional, configurational and conformational aspects of the molecule.

2.7.1 Bond lengths and angles

A bond length is the distance between two atomic nuclei that are joined together by some type of electronic bonding. Because atoms undergo vibrational and other motions, reported bond lengths are the average or equilibrium bond lengths.

If two atoms at $x_1, y_1, z_1$ and $x_2, y_2, z_2$, lie in a unit cell that has edges with lengths $a$, $b$ and $c$ and interaxial angles of $\alpha$, $\beta$ and $\gamma$, the square of the distance $r$ between these two atoms is

$$r^2 = [(x_1 - x_2)a]^2 + [(y_1 - y_2)b]^2 + [(z_1 - z_2)c]^2 +
[2ab \cos \gamma (x_1 - x_2)(y_1 - y_2)] +
[2ac \cos \beta (x_1 - x_2)(z_1 - z_2)] +
[2bc \cos \alpha (y_1 - y_2)(z_1 - z_2)].$$

The internuclear bond angle, $\angle ABC$, is the angle between the bonds $A-B$ and $B-C$ formed by three atoms $A$, $B$ and $C$, connected in that order. If the length of $A-B = l_1$, $B-C = l_2$ and $A-C = l_3$, then the angle $A - B - C = \delta$ may be calculated from the formula

$$\cos \delta = \frac{l_1^2 + l_2^2 - l_3^2}{2l_1 l_2}.$$

2.7.2 Estimated standard deviations

One way to assess the precision of a structure determination is to use the estimated standard deviation (e.s.d.) of the geometric quantity of interest. This is obtained from the least squares refinement. The more precisely a measurement is made, the smaller is the e.s.d. of that measurement.
Estimated standard deviations of bond lengths

For two atoms, A (x_A, y_A, z_A) and B (x_B, y_B, z_B), σ_xA is the e.s.d. of x for atom A and Δx = x_B - x_A. The required e.s.d. values are obtained by least squares refinement. The e.s.d. of the bond distance between A and B is:

$$\sigma_d^2 = \frac{1}{\tau^2} \left[ \left( \sigma_{x_A}^2 + \sigma_{x_B}^2 \right)(\Delta x + \Delta y \cos \gamma + \Delta z \cos \beta)^2 \right] +$$

$$+ \left[ \left( \sigma_{y_A}^2 + \sigma_{y_B}^2 \right)(\Delta y + \Delta x \cos \gamma + \Delta z \cos \alpha)^2 \right] +$$

$$+ \left[ \left( \sigma_{z_A}^2 + \sigma_{z_B}^2 \right)(\Delta z + \Delta x \cos \beta + \Delta y \cos \alpha)^2 \right]$$

Estimated standard deviations of inter bond angles

The e.s.d. of the angle between the bonds A-B and B-C is calculated by:

$$\sigma_\theta = \left( \frac{\sigma_A^2}{(AB)^2} + \frac{\sigma_B^2(AC)^2}{(AB)^2(BC)^2} + \frac{\sigma_C^2}{(BC)^2} \right)^{1/2}$$

where σ_A, σ_B and σ_C are the e.s.d.s of the positions of atoms A, B and C.

2.7.3 Torsion angles

The torsion angle describes steric relationships across single bonds and the ring geometry. A torsion is the extent of the twist of constituents about a bond. It will indicate if the bonds to the substituent groups are eclipsed, staggered or intermediate between these. For a sequence of four atoms A, B, C, D, the torsion angle ω(ABCD) is defined as the angle between the normals to the planes ABC and BCD (Fig. 2.7.1). By convention, ω is positive if the sense of rotation from BA to CD, viewed down BC, is clockwise, otherwise it is negative (Fig. 2.7.1). The sign of a torsion angle does not change by rotation or translation, and is

![Diagram of torsion angle ω](image-url)

Figure 2.7.1: Definition of the torsion angle ω
reversed by reflection or inversion. According to definition,

\[
\cos \omega = \frac{(\mathbf{a} \wedge \mathbf{b}) \cdot (\mathbf{b} \wedge \mathbf{c})}{ab^2 \sin \alpha \sin \gamma}, \quad \frac{b}{b} \sin \omega = \frac{(\mathbf{a} \wedge \mathbf{b}) \wedge (\mathbf{b} \wedge \mathbf{c})}{ab^2 \sin \alpha \sin \gamma}
\]

which become

\[
\cos \omega = \frac{\cos \alpha \cos \gamma - \sin \beta}{\sin \alpha \sin \beta}, \quad \sin \omega = \frac{Vb}{ab^2 \sin \alpha \sin \gamma}
\]

### 2.7.4 Ring conformations

A ring containing single bonds, could have different conformations which can be described by a potential energy curve having minima and maxima corresponding to ring conformers and transition states.

A unique mean plane is defined for a general monocyclic puckered ring. The geometry of the puckering relative to this plane is described by amplitude and phase coordinates.

For six-membered rings \((N = 6)\), there are three puckering degrees of freedom [8]. These are described by a single amplitude-phase pair \((q_2, \phi_2)\) and a single puckering coordinate \(q_3\). Alternatively, these coordinates may be replaced by a “spherical polar set” \((Q, \theta, \phi)\), where \(Q\) is the total puckering amplitude and \(\theta\) an angle \((0 \leq \theta \leq \pi)\) such that

\[
q_2 = Q \sin \theta \quad q_3 = Q \cos \theta
\]

Classical examples of degenerate conformations are the envelope \((E)\) and twisted \((T)\) conformations of cyclopentane and the boat \((B)\) and twist-boat or twisted \((T)\) conformations of cyclohexane.

\[
B_{2g} : \phi_B(i) = z_i = 6^{-1/2} q_3 \cos[3 \pi(i - 1)/6] = 6^{-1/2} q_3(-1)^{i-1}
\]

\[
E_{2u} : \phi_{B,T}(i) = z_i = 3^{-1/2} q_2 \cos[4 \pi(i - 1)/6 + \phi_2]
\]

where \(\phi_2\) is a phase angle having values of 0, 60, 120, 180, 240 or 300° for the pure \(B\) conformations and 30, 90, 150, 210, 270 or 330° for the pure \(T\) conformations. The characterising parameters \((q_3, q_2, \phi_2)\) are more generally identified as the \(N - 3\) generalised puckering coordinates.

The conformations of a six-membered ring in terms of the parameters \(q_3, q_2\) and \(\phi_2\) and the \(N - 3\) possible conformations remain characterised as the three \(C, B\) and \(T\) conformers.

### Least Squares Plane

A least squares plane is defined as the plane which minimizes \(\sum d_m^2\), where \(d_m\) are the perpendicular distances of the \(m\) atoms from the plane [11].
2.8 References


