CHAPTER- 2

This chapter covers the basic principles, theories and techniques of various physico-chemical methods involved in characterization of the coordination compounds
INSTRUMENTAL METHODS AND THEORIES

The basic purpose of this chapter is to amplify the methods that are used to identify the coordination compounds. No single technique is enough to provide a complete characterization of a compound rather, a variety of techniques are required in combination.

There are several physico-chemical methods available for the study of coordination compounds and a brief description of the techniques used in the investigation of the newly synthesized complexes described in the present work are given below:

1- Infrared Spectroscopy
2- Nuclear Magnetic Resonance Spectroscopy
3- Electron Paramagnetic Resonance Spectroscopy
4- Ultraviolet and Visible (Ligand Field) Spectroscopy
5- Magnetic Susceptibility Measurements
6- Molar Conductance Measurements
7- Elemental Analysis
8- Fluorescence study
9- Antibacterial activity
10- X-ray crystallography
INFRARED SPECTROSCOPY

When Infrared light is passed through a sample some of the frequencies are absorbed while other frequencies are transmitted through the sample without being absorbed. The plot of the percent absorbance or percent transmittance against frequency results is an infrared spectrum.

The IR radiation does not have enough energy to induce electronic transitions observed in UV spectroscopy. Absorption of IR radiation is restricted to the compounds with small energy differences in the possible vibrational and rotational states. For a molecule to absorb IR radiation, the vibrations or rotations within a molecule must cause a net change in the dipole moment of the molecule. The alternating electrical field of the radiation interacts with fluctuations in the dipole moment of the molecule. If the frequency of the radiation matches the vibrational frequency of the molecule then radiation will be absorbed, causing a net change in the amplitude of the molecular vibration.

In the absorption of the radiation, only transition for which change in the vibrational energy level is $\Delta V=1$ can occur, since most of the transition will occur from stable $V_0$ to $V_1$, the frequency corresponding to its energy is called the fundamental frequency.

The group frequency which are frequencies of certain groups are characteristic of the group irrespective of the nature of the molecule in which these groups are attached. The absence of any band in the approximate region indicates the absence of that particular group in the molecule.
The term “infrared” covers the range of electromagnetic spectrum between 0.78 and 1000 μm. In the context of infrared spectroscopy, wavelength is measured in “wavenumbers”, which have the units cm\(^{-1}\).

\[ \nu = \frac{1}{\lambda} \]

It is useful to divide the infrared region into three sections; near, mid and far infrared:

<table>
<thead>
<tr>
<th>Region</th>
<th>Wavelength range(μm)</th>
<th>Wavelength range (cm(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td>Near</td>
<td>0.78-2.5</td>
<td>12800-4000</td>
</tr>
<tr>
<td>Middle</td>
<td>2.5-50</td>
<td>4000-200</td>
</tr>
<tr>
<td>Far</td>
<td>50-1000</td>
<td>200-10</td>
</tr>
</tbody>
</table>

**Important Group Frequencies in the IR Spectra Pertinent to the Discussion of the Newly Synthesized Compounds.**

1. **Amines**

   a) **N-H Stretching Vibrations**

   The N-H Stretching vibrations occur in the region 3300-3500 cm\(^{-1}\) in the dilute solution\(^1\). The N-H stretching band shifts to lower value in the solid state due to the extensive hydrogen bonding. Primary amines in the dilute solutions, in non-polar solvents give two absorptions i.e. symmetric stretch found near 3400 cm\(^{-1}\) and asymmetric stretch mode found near 3500 cm\(^{-1}\). Secondary amines show only a single N-H stretching band in dilute solutions. The intensity and frequency
of N-H stretching vibrations of secondary amines are very sensitive to structural changes. The band is found in the range 3310-3350 cm\(^{-1}\) (low intensity) in aliphatic, secondary amines and near 3490 cm\(^{-1}\) (much higher intensity) in heterocyclic secondary amines such as pyrazole and imidazole.

b) N-H Bending Vibrations

The N-H bending (scissoring) vibration of primary amines is observed in the 1650-1580 cm\(^{-1}\) region of the spectrum. The band is medium to strong in intensity which shifts to slightly higher frequencies when the compound is associated. The N-H bending band is seldom detectable in the spectra of aliphatic secondary amines, whereas secondary aromatic amines absorb near 1515 cm\(^{-1}\).

2. Amine Salts

N-H stretching Vibration

The ammonium ion displays strong, broad absorption in the 3300-3030 cm\(^{-1}\) region because of N-H stretching vibrations. There is also a combination band in the 2000-1709 cm\(^{-1}\) region.

Salts of primary amines show\(^2\) strong, broad absorption between 3000-2800 cm\(^{-1}\) arising from asymmetrical and symmetrical stretching in the NH\(_3^+\) group. In addition, multiple combination bands of medium intensities occur in the 2800-2000 cm\(^{-1}\) region the most prominent being the band near 2000 cm\(^{-1}\).

Salts of secondary amines absorb\(^3\) strongly in the 3000-2700 cm\(^{-1}\) region with the multiple bands extending to 2273 cm\(^{-1}\).
Tertiary amine salts absorb at longer wavelengths (2700-2250) than the salts of primary and secondary amines.

Quaternary ammonium salts can have no N-H stretching vibrations.

3. Methyl Group Frequency

Absorption arising from C-H stretching in the alkanes occurs generally in the region of 2840-3000 cm\(^{-1}\). The position of C-H stretching vibrations are among the most stable in the spectrum. An examination of a large number of saturated hydrocarbons containing methyl group showed\(^1\) in all cases, two distinct bands occurring at 2960 cm\(^{-1}\) and 2870 cm\(^{-1}\). The first of these results from asymmetric stretching mode in which two C-H bonds of the methyl group are extending while the third one is contracting (v\(_{\text{asy}}\), CH\(_3\)). The second arises from symmetric stretching (v\(_{\text{sy}}\), CH\(_3\)) in which all three of the C-H bonds extend and contract in phase. The presence of several methyl groups in a molecule results in a strong absorption bands at these vibrational modes.

4. C=\(\equiv\)N Stretching Frequency

Schiff’s bases (RCH=NR, imines), oximes, thiazoles, iminocarbonates etc. show the C=\(\equiv\)N stretching frequency in the 1471-1689 cm\(^{-1}\) region\(^1\). Although the intensity of the C=\(\equiv\)N stretch is variable, however it is usually more intense than the C=C stretch.

5. N–N Stretching Frequency

A strong band appearing around 1000 cm\(^{-1}\) may reasonably be assigned\(^5\) to \(\nu\) (N–N) vibrations.
6. **M-N Stretching Frequency**

The M-N stretching frequency is of particular interest since it provides direct information regarding the metal-nitrogen coordinate bond. Different amines complexes exhibited the metal-nitrogen frequencies in the 215-465 cm\(^{-1}\) region.

7. **M-X Stretching Frequency**

Metal-halogen stretching bands appear in the region of 500-750 cm\(^{-1}\) for MF, 200-400 cm\(^{-1}\) for MCl, 200-300 cm\(^{-1}\) for MBr and 100-200 cm\(^{-1}\) for MI.

8. **M-O Stretching Frequency**

Metal-oxygen stretching frequency has been reported to appear in different regions for different metal complexes. The M-O stretching frequency of the complexes in which oxygen of the carbonyl group or carboxylic group is the donor site lie in the range 510-625 cm\(^{-1}\).

9. **Pyridine Ring Vibrations**

The vibrational spectra of pyridine shows three important vibrations, i.e. 16\(b\) (out of plane ring deformation), 6\(a\) and 8\(a\) (in plane ring deformation) observed at 403, 601 and 1578 cm\(^{-1}\) respectively. All the three bands suffer considerable positive shifts on coordination of pyridine to metal ions and magnitude of the shift depends on the stereochemistries of the complexes and on the metal atoms. The 601 cm\(^{-1}\) band has been suggested to be more sensitive to the stereochemistry of the complexes such that in tetrahedral, polymeric octahedral and octahedral complexes, it appears at 642, 631 and 625 cm\(^{-1}\), respectively. In a few complexes, the doubling of the above mentioned pyridine bands have also
been reported which has been suggested to be due to some interaction between different pyridine molecules either within a single molecule or between different molecules of the unit cell.

10. **C=O stretching vibrations**

Ketones, aldehydes, carboxylic acids, carboxylic esters, lactones, acid halides, anhydrides, amides and lactams show a strong stretching absorption band in the region of 1870-1540 cm⁻¹. Its relatively constant position and high intensity and relative freedom from interfering bands make it as one of the easiest band to recognize in infrared spectra.

11. **Carboxylate group Vibrations**

Extensive Infrared studies have been made on metal complexes of carboxylic acids. The asymmetric, \( v_{as}(CO_2^-) \) and the symmetric, \( v_s(CO_2^-) \) of free acetate ion have been reported to appear ca. 1560 and 1417 cm⁻¹, respectively. In case of unidentate mode of bonding of carboxylate group the \( v(C=O) \) is higher than \( v_{as}(CO_2^-) \) and \( v(C-O) \) is lower than \( v_s(CO_2^-) \) resulting in much larger separation between the two \( v(CO) \) in unidentate complexes than in the free ion. While a opposite trend is observed if carboxylate group is bonded in bidentate (chelate) fashion in the complex, the separation between the \( v(CO) \) is smaller than that of the free ion. In the bridging complex however, two \( v(CO) \) are close to the free carboxylate ion value.

In a series of unidentate complexes, \( v(C=O) \) increases and \( v(C-O) \) decreases as the M-O bond becomes stronger.
12. Nitrate group frequencies

Free nitrate group exhibits bands in the 1008-1041 cm\(^{-1}\), 1271-1306 cm\(^{-1}\), and 715-728 cm\(^{-1}\) regions.

13. Perchlorate vibrations

The perchlorate (\(\text{ClO}_4^{2-}\)) group exhibits medium intensity bands in the 1089-1386 and 920-948 cm\(^{-1}\) regions.

FTIR spectra (4000-200 cm\(^{-1}\)) were recorded as KBr and/or CsCl, CsI discs on a Perkin Elmer-2400 spectrometer.

NUCLEAR MAGNETIC RESONANCE SPECTROSCOPY

\(^1\text{H}-\text{NMR Spectroscopy}\)

As implied from the name, nuclear magnetic resonance (or NMR) is concerned with the magnetic properties of certain nuclei e.g. \(^1\text{H},\ ^{13}\text{C}\). The nuclei for which spin quantum number \(I > 0\) exhibits the NMR phenomenon where \(I\) is associated with the mass number and atomic number of the nuclei as shown below:

<table>
<thead>
<tr>
<th>Mass number</th>
<th>Atomic number</th>
<th>Spin quantum number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Odd</td>
<td>Odd or Even</td>
<td>(1/2, 3/2, 5/2, \ldots)</td>
</tr>
<tr>
<td>Even</td>
<td>Even</td>
<td>0</td>
</tr>
<tr>
<td>Even</td>
<td>Odd</td>
<td>1, 2, 3</td>
</tr>
</tbody>
</table>
The nucleus of the $^1\text{H}$, the proton has $I = 1/2$, whereas, $^{12}\text{C}$ and $^{16}\text{O}$ have $I = 0$ and therefore are nonmagnetic. Other important magnetic nuclei that have been studied extensively by NMR are $^{11}\text{B}$, $^{13}\text{C}$, $^{14}\text{N}$, $^{15}\text{N}$, $^{17}\text{O}$, $^{19}\text{F}$ and $^{31}\text{P}$.

Since atomic nuclei are associated with charge, a spinning nucleus generates a small electric current and has a finite magnetic field associated with it. When a spinning nucleus is placed in a magnetic field the nuclear magnet experiences a torque which tend to align it with the external field. The number of possible orientations for a magnetic nucleus under the influence of an external magnetic field is given by $(2I+1)$ so that for nuclei with spin $1/2$ ($^1\text{H}$, $^3\text{C}$, $^9\text{F}$ etc.) only two orientations are allowed, parallel to the field (low energy) and against the field (high energy). If the precessing nuclei are irradiated with a beam of radiofrequency energy of the correct frequency, the low energy nuclei may absorb this energy and move to a higher energy state. The precessing nuclei will only absorb energy from the radiofrequency source if the precessing frequency is the same as the frequency of the radiofrequency beam, the nucleus and the radiofrequency beam are said to be in resonance, hence the term nuclear magnetic resonance.

The precessional frequency, $\nu$ is directly proportional to the strength of the external field, $B_0$, i.e.

$$\nu \propto B_0$$

This is one of the most important relationships in NMR spectroscopy.
A proton exposed to an external magnetic force 1.4 T (14,000 gauss) will press a ≈ 60 million times per second, so that $\nu = 60$ MHz. The precessional frequency of all protons in same external applied field is not, however the same and the precise value for any proton depends on a number of factors such as electronegativity of the attached groups, van der waals deshielding and anisotropic effects explained by Packard in 1951, who detected three different values for the precessional frequencies of the protons in ethanol which have been rationalized for the three different chemical environments for the protons in ethanol (CH$_3$, CH$_2$ and OH) since then NMR has become an important tool for the chemists. As the shift in frequency depends on chemical environment, the term chemical shift was coined. Chemical shift positions are normally expressed in $\delta$ (delta units) which are defined as proportional differences in parts per million (ppm) from an appropriate reference standard (TMS in the case of proton and Carbon-13 NMR).

Since the $\delta$ unit is proportionality, it is a dimensionless number it is independent of field strength.

The $^1$HNMR spectra were recorded in DMSO-d$_6$ and D$_2$O using a Jeol Eclipse-400 spectrophotometer with Me$_4$Si as an internal standard.

**ELECTRON SPIN RESONANCE SPECTROSCOPY**

Electron resonance is a branch of absorption spectroscopy in which radiation of microwave frequency is absorbed by molecule possessing electrons with unpaired spins. The phenomenon has been designated by different names:
Electron paramagnetic resonance (EPR), Electron spin resonance (ESR) and Electron magnetic resonance.

For an electron of spin $S = 1/2$, the spin angular momentum quantum number can have values of $m_s = \pm 1/2$, which in the absence of a magnetic field leads to doubly degenerate spin energy state. When a magnetic field is applied this degeneracy is resolved. The low energy state has the spin magnetic moment aligned with the field and corresponds to the quantum number, $m_s = -1/2$, while the high energy state $m_s = +1/2$ has its moment opposed to the field. In EPR, a transition between the two different electron spin energy states occurs upon absorption of a quantum of radiation in the radiofrequency or microwave region. The energy, $E$ of transition is given by

$$E = h\nu = g\beta H_0$$

Where $h$ is Planck's constant, $\nu$ the frequency of radiation, $\beta$ the Bohr magneton, $H_0$ the field strength, and $g$ the spectroscopic splitting factor. The quantity $g$ is not constant, but tensor quantity. For a free electron $g$ has the value of 2.0023. In many free radicals, the $g$ value of the odd electron is close to that of a free electron, but in metal ions $g$ values are often greatly different from the free electron value. In general the magnitude of $g$ depends upon the orientation of the molecule containing the unpaired electron with respect to the magnetic field. In a solution or in gas phase, $g$ is averaged over all orientations because of the free motion of the molecule, but in a crystal, movement is restricted. If the paramagnetic radical or ion is located in a perfectly cubic site (e.g. an octahedral or tetrahedral site) the $g$ value is independent of the orientation of the crystal and
is said to be isotropic. In a crystal site of lower symmetry the g value depends upon the orientation of the crystal and is said to be anisotropic. The Z direction is defined coincident with the highest fold rotation axis. The $g_z$ value is equivalent to $g_{II}$, the g value obtained when the Z axis is parallel with the external magnetic field. The g values along the x and y axes are $g_x$ and $g_y$, which in a tetragonal site are equal and referred to as $g_\perp$, the g value obtained with the external magnetic field perpendicular to the z axis, the experimental g value is given by the following equation for a system with axial symmetry:

$$g^2 = g_{II}^2 \cos^2 \theta + g_{\perp}^2 \sin^2 \theta$$

small distortions which go undetected by X-ray methods can sometimes be determined by EPR from the inequality of the g values.

EPR spectrum can be represented by plotting intensity against the strength of the applied field, but EPR spectra are commonly presented as derivative curves i.e. the first derivative (the slope of the absorption curve) is plotted against the strength of the magnetic field. Much greater sensitivity can be achieved by this detection method if the line shape is broad.

A system of charges exhibits paramagnetism whenever it has a resultant angular momentum. Such paramagnetic system includes elements containing 3d, 4d, 4f, 5d, 5f, 6d etc, electrons, atoms having an odd number of electrons like hydrogen, molecules containing odd number of electrons such as NO₂, NO etc. and free radicals which possess an unpaired electron like methyl, Diphenyl picryl hydazide free radical etc. are among the suitable reagents for EPR investigation.
Splitting of energy levels in EPR occurs under the effect of two types of fields, namely the internal crystalline field and applied magnetic field. While studying a paramagnetic ion in a diamagnetic crystal lattice, two types of interactions are observed, i.e. interaction between the paramagnetic ions called dipolar interaction and the interactions between the paramagnetic ion and the diamagnetic neighbour called crystal field interaction. For small doping amount of paramagnetic ion in the diamagnetic host, the dipolar interaction will be negligibly small. The latter interaction of paramagnetic ion with diamagnetic ligands modifies the magnetic properties of the paramagnetic ions. According to crystal field theory, the ligand influence the magnetic ion through the electric field which they produce at its site and their orbital motion get modified. The crystal field interaction is affected by the outer electronic shells.

The dipole-dipole interaction arises from the influence of magnetic field of one paramagnetic ion on the dipole moments of the neighboring, smaller ions. The local field at any given site will depend on the arrangements of the neighbors and the direction of their dipole moments. Thus the resultant magnetic field on the paramagnetic ion will be the vector sum of the external field and the local field. Thus resultant field varies from site to site giving a random displacement of the resonance frequency of each ions and thus broadening the line widths.

Hyperfine interactions are mainly magnetic dipole interactions between the electron magnetic moment and the nuclear magnetic moment of the paramagnetic ion. The quartet structure in the EPR of vanadyl ion are the results of hyperfine interactions. The origin of this can be understood simply by assuming that the
nuclear moment produces a magnetic field $B_N$ at the magnetic electrons and the modified resonance condition will be $E = h\nu = g\beta |B + B_N|$ where $B_N$ takes up $2I + 1$, where $I$ is the nuclear spin. There may be an additional hyperfine structure also due to interaction between magnetic electrons and the surrounding nuclei called superhyperfine structure. The effect was first observed by Owens and Stevens in ammonium hexa chloroiridate$^{13}$ and subsequently for a number of transition metal ions in various hosts$^{14,15}$.

The EPR spectra of complexes were recorded on a JEOL JES RE2X EPR spectrometer.

**ULTRA-VIOLET AND VISIBLE (LIGAND FIELD) SPECTROSCOPY**

Most of the compounds absorb light somewhere in the spectral region between 200 and 1000 nm. These transitions correspond to the excitation of electrons of the molecules from ground state to higher electronic states. In a free transition metal ion all the five d-orbitals viz. $d_{xy}$, $d_{yz}$, $d_{xz}$, $d_{z2}$ and $d_{x2-y2}$ are degenerate. However, in coordination compounds due to the presence of ligands this degeneracy is lifted and d-orbitals split into two groups called $t_{2g}$ ($d_{xy}$, $d_{yz}$ and $d_{xz}$) and $e_g$ ($d_{z2}$ and $d_{x2-y2}$) in an octahedral complex and $t$ and $e$ in a tetrahedral complex. The set of $t_{2g}$ orbitals goes below and the set of $e_g$ orbitals goes above the original level of the degenerate orbitals in an octahedral complex. In case of the tetrahedral complexes the position of the two sets of the orbitals is reversed. When a molecule absorbs radiation, its energy equal in magnitude to $h\nu$ can be expressed by the relation:
\[ E = hv \]

or \[ E = \frac{hc}{\lambda}. \]

Where \( h \) is Planck’s constant, \( v \) and \( \lambda \) are the frequency and wavelength of the radiation, respectively and \( c \) is the velocity of the light.

In order to interpret the electronic spectra of transition metal complexes, the device of energy level diagram based upon ‘Russell Saunder Scheme’ must be introduced. This has the effect of splitting the highly degenerate configurations into groups of levels having lower degeneracies known as ‘Term Symbols’.

The orbital angular momentum of electrons in a filled shell vectorically adds up to zero. The total orbital angular momentum of an incomplete d shell electron is observed by adding \( L \) value of the individual electrons, which are treated as a vector with a component \( m_l \) in the direction of the applied field. Thus

\[ L = \sum m_l = 0, 1, 2, 3, 4, 5, 6, \]

\( S, P, D, F, G, H, I \)

The total spin angular momentum \( S = \sum s_i \) where \( s_i \) is the value of spin angular momentum of the individual electrons. \( S \) has a degeneracy \( \tau \) equal to \( 2S + 1 \), which is also known as ‘Spin Multiplicity’. Thus a term is finally denoted as \( \tau L \). For example, if \( S = 1 \) and \( L = 1 \), the term will be \(^3P\) and similarly if \( S = 1 \frac{1}{2} \), and \( L = 3 \), the term will be \(^4F\).

In general the terms arising from a \( d^n \) configuration are as follows:
Coupling of L and S also occurs, because both L and S if non-zero, generate magnetic fields and thus tend to orient their moments with respect to each other in the direction where their interaction energy is least. This coupling is known as 'LS coupling' and gives rise to resultant angular momentum denoted by quantum number \( J \) which may have quantized positive values from \(|L + S|\) up to \(|L - S|\). For example, in the case of \( ^3P \) (\( L = 1, S = 1 \)) and \( ^4F \) (\( L = 3, S = 1 \)) possible values of \( J \) representing state, arising from term splitting are 2, 1 and 0 and \( 4, 3, 2, 1 \) respectively. Each state is specified by \( J \) is \( 2J + 1 \) fold degenerate. The total number of states obtained from a term is called the multiplet and each value of \( J \) associated with a given value of \( L \) is called component. Spectral transitions due to spin-orbit coupling in an atom or ion occurs between the components of two different multiplets while LS coupling scheme is used for the elements having atomic number less than 30, in that case spin-orbital interactions are large and electron repulsion parameters decrease. The spin-angular momentum of an individual electron couples with its orbital momentum to give an individual \( J \) for that electron. The individual \( J \)'s couple to produce a resultant \( J \) for the atom. The
electronic transitions taking place in an atom or ion are governed by certain Selection Rules which are as follows:

1. Transitions between states of different multiplicity are forbidden.
2. Transitions involving the excitation of more than one electron are forbidden.
3. In a molecule, which has a centre of symmetry, transitions between two gerade or two ungerade states are forbidden.

It is possible to examine the effects of crystal field on a polyelectron configuration. The ligand field splitting due to cubic field can be obtained by considerations of group theory. It has been shown that an S state remains unchanged. P states does not split, and D state splits into two and F state into three and G state into four states as tabulated below:

\[
\begin{align*}
S & \quad A_1 \\
P & \quad T_1 \\
D & \quad E + T_2 \\
F & \quad A_2 + T_1 + T_2 \\
G & \quad A_2 + E + T_1 + T_2 \\
\end{align*}
\]

(Applicable for an octahedral 'Oh' as well as tetrahedral 'Td' symmetry).

Transitions from the ground state to the excited state occur according to the selection rules described earlier. The energy level order of the states arising from the splitting of a term state for a particular ion in an octahedral field is the
reverse that of the ion in a tetrahedral field. However, due to transfer of charge from ligand to metal or metal to ligand, sometimes bands appear in the ultraviolet region of the spectrum. These spectra are known as ‘Charge Transfer Spectra’ or Redox Spectra. In metal complexes there are often possibilities that charge transfer spectra extend into the visible region to obscure d-d transition. However, these should be clearly discerned from the ligand bands, which might also occur in the same region.

The electronic Spectra of complexes were recorded in DMSO on a Pye- Unicam 8800 spectrophotometer at room temperature.

MAGNETIC SUSCEPTIBILITY MEASUREMENTS

The determination of magnetic moments of transition metal complexes have been found to provide ample information in assigning their structure. The main contribution to bulk magnetic properties arises from magnetic moment resulting from the motion of electrons. It is possible to calculate the magnetic moments of known compounds from the measured values of magnetic susceptibility.

There are several kinds of magnetic phenomenon observed in chemical substances viz., diamagnetism, paramagnetism and ferromagnetism or antiferromagnetism. Mostly compounds of the transition elements are paramagnetic. Diamagnetism is associated with substances having closed shell electrons in an applied magnetic field. In the closed shell the electron spin moment and orbital moment of the individual electrons balance one another so
that there is no magnetic moment. Ferromagnetism and antiferromagnetism arise as a result of interaction between dipoles of neighboring atoms.

If a substance is placed in a magnetic field $H$, the magnetic induction $B$ with the substance is given by

$$B = H + 4\pi I$$

Where $I$ is the intensity of magnetization. The ratio $B/H$ is called magnetic permeability of the material and is given by

$$\frac{B}{H} = I + 4\pi I/H = I + 4\pi K\kappa$$

Where $\kappa$ is called the magnetic susceptibility per unit volume or volume susceptibility. $B/H$ is the ratio of the density of lines of force within the substance to the density of such lines in the same region in the absence of sample. Thus the volume susceptibility in vacuum is zero by definition since in vacuum $B/H = 1$.

When magnetic susceptibility is considered on the weight basis, the term gram susceptibility ($\chi_g$) is used instead of volume susceptibility. The $\mu_{\text{eff}}$ value can then be calculated from the gram susceptibility multiplied by the molecular weight corrected for diamagnetism

$$\mu_{\text{eff}} = 2 \cdot 84\sqrt{\chi_g^{\text{corr}} \cdot T \cdot BM}$$

where $T$ is the absolute temperature at which the experiment is performed.

The magnetic properties of any individual atom or ion will result from some combination of two properties, that is the inherent spin moment of the electron and the orbital moment resulting from the motion of the electron around
the nucleus. The magnetic moments are usually expressed in Bohr Magnetons (BM). The magnetic moment of a single electron is given by

\[ \mu_s = g \sqrt{S(S+1)} \text{ BM} \]

Where \( S \) is the total spin quantum number arising from unpaired electrons and \( g \) is the gyromagnetic ratio. For \( \text{Mn}^{2+}, \text{Fe}^{3+} \) and other ions having ground states as \( S \) there is no orbital angular momentum. While the transition metal ion in their ground state \( D \) or \( F \) being most common, do possess orbital angular momentum. The magnetic moment for such ions, eg. \( \text{Co}^{2+} \) and \( \text{Ni}^{2+} \) is given by

\[ \mu_{s+L} = \sqrt{4S(S+1) + L(L+1)} \]

where \( L \) represents the total orbital angular momentum quantum number for the ion.

The spin magnetic moment is insensitive to the environment of metal ion but the orbital magnetic moment is not. In order for an electron to have an orbital angular momentum and thereby an orbital magnetic moment with reference to a given axis, it must be possible to transform the orbital into a fully equivalent orbital by rotation about that axis. For octahedral complexes the orbital angular momentum is absent for \( A_{1g}, A_{2g} \) and \( E_g \) term, but can be present for \( T_{1g} \) and \( T_{2g} \) terms. Magnetic moments of the complex ions with \( A_{2g} \) and \( E_g \) ground terms may depart from the spin-only value by a small amount. The magnetic moments of the complexes possessing \( T \) ground terms usually differ from the high spin value and
vary with temperature. The magnetic moments of the complexes having a $^6A_{1g}$
ground term are very close to the spin-only value and are independent of the
temperature.

For octahedral and tetrahedral complexes in which spin-orbit coupling
causes a split in the ground state, an orbital moment contribution is expected.
Even no splitting of the ground state appears in cases having no orbital moment
contribution, an interaction with higher states can appear due to spin-orbit
coupling giving an orbital moment contribution.

Practically the magnetic moment value of the unknown complex is
obtained on Gouy Magnetic balance. Faraday method can also be applied for the
magnetic susceptibility measurement of small quantity of solid samples.

The gram susceptibility is measured by the following formula.

$$\chi_g = \frac{\Delta W}{W} \cdot \frac{W_{std}}{\Delta W_{std}} \cdot \chi_{std}$$

Where $\chi_g$ = Gram Susceptibility

$\Delta W$ = Change in weight of the unknown sample with magnet on and off.

$W$ = Weight of the known sample

$\Delta W_{std}$ = Change in weight of standard sample with magnets on and off.

$W_{std}$ = Weight of standard sample.

$\chi_{std}$ = Gram susceptibility of the standard sample.
Temperature Dependence of Magnetic Susceptibility: Curie and Curie-Weiss laws

The susceptibilities of the different kinds of magnetic material are distinguished by their different temperature dependences as well as by their absolute magnitudes. Many paramagnetic substances obey Curie Law, especially at high temperatures. This states that the magnetic susceptibility is inversely proportional to temperature:

$$\chi = \frac{C}{T}$$

Where $C$ is the curie constant. Often, however a better fit to the experimental data is provided by the Curie-Weiss Law:

$$\chi = \frac{C}{T+\theta}$$

Where $\theta$ is the Weiss constant.

Ferromagnetic materials show a very large susceptibility at low temperatures that decreases increasingly rapidly with rising temperature. Above a certain temperature (ferromagnetic Curie Temperature, $T_c$), the material is no longer ferromagnetic but reverts to paramagnetic, where Curie-Weiss Law behavior is usually observed.

For antiferromagnetic materials, the value of $\chi$ actually increases with rising temperature up to a critical temperature known as Neel point, $T_N$. Above $T_N$, the material again reverts to paramagnetic behavior.
The paramagnetic $\chi$ values correspond to the situation where unpaired electrons are present in the material and show some tendency to align themselves in a magnetic field.

In ferromagnetic materials, the electron spins are aligned parallel due to the cooperative interactions between spins of the neighboring ions in the crystal structure. The large $\chi$ values represent this parallel alignment of a large number of spins. In general, not all spins are parallel in a given material unless (a) very high magnetic fields and (b) low temperature are used.

In antiferromagnetic materials, the electrons spins are aligned antiparallel and have a canceling effect on $\chi$. Hence small $\chi$ values are expected.

For all materials, the effect of increasing the temperature is to increase the thermal energy possessed by ions and electrons. There is, therefore, a natural tendency for increasing structural disorder with increasing temperature.

For paramagnetic materials, the thermal energy of ions and electrons act to partially cancel the ordering effect of the applied magnetic field. Indeed, as soon as the magnetic field is removed, the orientation of the spins becomes disordered. Hence, for paramagnetic materials, $\chi$ decreases with increasing temperature, in Curie/Curie-Weiss Law fashion.

Magnetic susceptibility measurements were carried out using Faraday balance at 298°K. However, the temperature dependence of magnetic susceptibility was recorded on a quantum design SQUID magnetometer under field cooled conditions between 298 and 4.5 K at 1000G.
CONDUCTIVITY

The resistance of a sample of an electrolytic solution is defined by

\[ R = \rho \left( \frac{l}{A} \right) \]

Where \( l \) is the length of a sample of electrolyte and \( A \) is the cross sectional area. The symbol \( \rho \) is the proportionality constant and is a property of a solution. This property is called resistivity or specific resistance. The reciprocal of resistivity is called conductivity, \( \kappa \)

\[ \kappa = \frac{l}{\rho} = \frac{l}{RA} \]

Since \( l \) is in cm, \( A \) is in cm\(^2\) and \( R \) in ohms (Ω), the units of \( \kappa \) is Ω\(^{-1}\) cm\(^{-1}\) or S cm\(^{-1}\) (Siemens per cm)

Molar Conductivity

If the conductivity \( \kappa \) is in Ω\(^{-1}\) cm\(^{-1}\) and the concentration \( C \) is in mol cm\(^{-3}\), then the molar conductivity \( \Lambda \) is in Ω\(^{-1}\) cm\(^2\) mol\(^{-1}\) and is defined by

\[ \Lambda = \frac{\kappa}{C} \]

Where \( C \) is the concentration of solute in mol cm\(^{-3}\).

Conventionally solutions of 10\(^{-3}\) M concentration are used for the conductance measurement. Molar conductance values of different types of electrolytes in a few solvents are given below:

A 1:1 electrolyte may have a value of 70-95 ohm\(^{-1}\) cm\(^2\) mol\(^{-1}\) in nitromethane, 50-75 ohm\(^{-1}\) cm\(^2\) mol\(^{-1}\) in dimethyl formamide and D\(_2\)O and 100-160 ohm\(^{-1}\) cm\(^2\) mol\(^{-1}\) in methyl cyanide. Similarly a solution of 2:1 electrolyte may
have a value of 150-180 ohm$^{-1}$ cm$^2$ mol$^{-1}$ in nitromethane, 130-170 ohm$^{-1}$ cm$^2$ mol$^{-1}$ in dimethylformamide and 140-220 ohm$^{-1}$ cm$^2$ mol$^{-1}$ in methyl cyanide.$^{16-18}$

The electrical conductivities of 10$^{-3}$ M solutions in DMSO and D$_2$O were obtained on a digital APX 185 conductivity bridge equilibrated at 25±0.01°C.

**ELEMENTAL ANALYSIS**

The chemical analysis is quite helpful in fixing the stoichiometric composition of the ligand as well as its metal complexes. Carbon, hydrogen and nitrogen analyses were carried out on a Perkin Elmer-2400 analyzer. Chlorine was analyzed by conventional method$^{19}$ for chlorine estimation, a known amount of the sample was decomposed in a platinum crucible and dissolved in water with a little concentrated nitric acid. The solution was then treated with silver nitrate solution. The precipitate was then dried and weighed.

For the metal estimation$^{20}$, a known amount of complex was decomposed with mixture of nitric, perchloric and sulfuric acids in a beaker. It was then dissolved in water and made up to known volume so as to titrate it with standard EDTA.

**FLUORESCENCE SPECTROSCOPY**

With some molecules, the absorption of a photon is followed by the emission of light of a longer wavelength (i.e. lower energy). This emission is called fluorescence (or phosphorescence, if the emission is long lived). There are many environmental factors that effect the fluorescence spectrum; furthermore, fluorescence efficiency is also environmentally dependent. Because these
parameters of fluorescence are more sensitive to the environment than are those of absorbance and because smaller amounts of material are required, fluorescence spectroscopy is frequently of greater value than absorbance measurement. With macromolecules, fluorescence measurements can give information about conformation, binding sites, solvent interactions, degree of flexibility, intermolecular distances and rotational diffusion coefficient of macromolecules. Furthermore, with living cells, fluorescence can be used to localize otherwise undetectable substances.

As with other physical methods, the theory of fluorescence is not yet adequate to permit a positive correlation between fluorescent spectrum and the properties of the immediate environment of the emitter; hence the utility of the procedure is based on establishing empirical principles from studies with model compounds.

The excited molecule does not always fluoresce. The probability of fluorescence is described by the quantum yield, Q that is the ratio of the number of emitted to absorbed photons. Several factors determine Q, some of these are properties of the molecule itself (internal factors) and some are environmental.

The internal factors are not generally of interest to biochemist concerned with the properties of macromolecules, environmental factors are more important. The effect of the environment is primarily to provide radiationless processes that compete with fluorescence and thereby reduce Q, this reduction in Q is called quenching. In biological systems, quenching is usually a result of either collisional processes (either a chemical reaction or simply collision with exchange of energy)
or a long range radiative process called resonance energy transfer. These three factors are usually expressed in an experimental situation involving solutions as an effect of the solvent or dissolved compounds (called quenchers), temperature, pH, neighboring chemical groups, or the concentration of the fluor.

It is important to know that distinction between a corrected spectrum and an uncorrected one is not often made in the presentation of fluorescence spectra in journal articles. It is common to plot a spectrum as the photomultiplier output versus wavelength. This is an uncorrected spectrum. Plotting fluorescence intensity or quantum yield produces a corrected spectrum. Invariably, when photomultiplier output is plotted, it is incorrectly called fluorescence or fluorescence intensity.

To measure $Q$ requires the counting of photons because

$$Q = \frac{\text{photons emitted}}{\text{photons absorbed}}$$

$Q$ is a dimensionless quantity

Because the energy, $E$, of one photon is related to the frequency, $v$ of the light by the relation $E = hv$, a measurement of the number of photons requires measuring the energy of the radiation and correcting for frequency. This usual method for determining $Q$ requires a comparison with a fluor of known $Q$, two solutions are prepared—one of the sample and one of the standard fluor—and, with the same exciting source, the integrated fluorescence (i.e. the area of the spectrum) of each is measured.

The quantum yield, $Q_x$, of a sample $X$ is
Chapter 2

\[ Q_s = \frac{I_x Q_s A_s}{I_s A_x} \]

where \( Q_s \) is the quantum yield of the standard, \( I_x \) and \( I_s \) are the integrated fluorescence intensities of the sample and the standard, respectively and \( A_x \) and \( A_s \) are the percentage of absorption of each solution at the exciting wavelength. Usually the solutions are adjusted so that \( A_x = A_s \).

Two types of fluors are used in fluorescence analysis of macromolecules- intrinsic fluors (contained in the macromolecules themselves) and extrinsic fluors (added to the system, usually binding to one of the components).

For proteins, there are only three intrinsic fluors- tryptophan, tyrosine and phenylalanine. The fluorescence of each of them can be distinguished by exciting with and observing at the appropriate wavelength. In practice, tryptophan fluorescence is most commonly studied, because phenylalanine has a very low Q and tyrocine fluorescence is frequently very weak due to quenching. The fluorescence of tyrosine is almost totally quenched if it is ionized, or near an amino group, a carboxyl group, or a tryptophan. In special situations, however, it can be detected by excitation at 280 nm. The principle reason for studying the intrinsic fluorescence of proteins is to obtain information about conformation. This is possible because the fluorescence of both tryptophan and tyrosine depends significantly on their environment (i.e. solvent, pH and presence of a quencher, a small molecule, or a neighboring group in the protein).

Fluorescence measurements were made on a Shimadzu RF-5301PC spectrofluorometer, equipped with microcomputer.
ANTIBACTERIAL ACTIVITY

The antibacterial activity of the compounds was determined by using agar well diffusion method\textsuperscript{21}. Cells were grown overnight at 37°C in Luria broth. Nutrient agar plates were prepared with small wells in it. Cells were spreaded on to the plates and different concentrations of compounds were kept in the wells. Plates were incubated overnight at 37°. Zone of inhibition around the well were calculated/measured.

X-RAY CRYSTALLOGRAPHY

Undoubtedly the most important and useful technique, X-ray diffraction, has been in use since the early part of this century for the fingerprint characterization of crystalline materials and for the determination of their crystal structures.

\textit{X-rays and their generation}

X-rays are electromagnetic radiations of wavelength \( \sim 1\text{Å} \) \((10^{-10}\text{m})\) occurring between γ-rays and the ultraviolet region of electromagnetic spectrum. The X-rays used in almost all diffraction experiments are produced by a process that leads to monochromatic X-rays.

Commonly, two approaches have been used to treat diffraction by crystals as mentioned below:

1. The Laue equations

2. and Bragg’s Law
The Laue equations provide a rigorous and mathematically correct way to describe diffraction by crystals. The drawback is that they are cumbersome to use. The alternative theory of diffraction based on Bragg's law is much simpler and is used almost universally in solid state chemistry.

**Structure factor**

The X-ray scattering power of an atom is directly proportional to the number of electrons composing it and can be expressed by a scattering factor $f$. In order to determine the combined scattering power of all the atoms in a unit cell, it is necessary to relate the differences between the pathlengths of X-rays scattered by each atom. This is done most conveniently by a geometric factor which is a function of the position of each atom among the equipoints. When the amplitudes of the wavelets scattered by each atom in the unit cell are added, one obtains the so-called structure factor or structure amplitude.

$$F_{hkl} = f_1 e^{2\pi i (hx_1 + ky_1 + lx_1)} + \ldots + f_N e^{2\pi i (hx_N + ky_N + lx_N)}$$

$$= \sum_{n=1}^{N} f_n e^{2\pi i (hx_n + ky_n + lx_n)}$$

where $N$ is the total number of atoms contained in a unit cell. The exponential term expresses the relative phase of the radiation scattered by each atom, $n$ as a function of its position in the unit cell $x_n, y_n, z_n$.

In an actual x-ray diffraction experiment one measures the intensities rather than the amplitudes of the reflected beam. The intensity is directly proportional to the square of the amplitude

$$I_{hkl} \sim F_{hkl}^2$$
So that it is possible to determine the positions of the atoms in a unit cell, that is, the crystal structure, directly from the observed intensity values.

**Factors that effect intensities**

Intensities depend on several factors other than structure factors. The main factors are:


2. *Structure factor* – dependence on the position of atoms in the unit cell and their scattering power.

3. *Lorenz factor* – a geometric factor that depends on the particular type of instrument used and varies with $\theta$. Usually lumped with polarization factor to give $L_p$ factor.

4. *Temperature factor* – thermal vibrations of atoms cause a decrease in the intensities of diffracted beams and an increase in background scatter.

5. *Absorption factor* – absorption of X-rays by the sample and depends on the form of the sample and geometry of the instrument. Ideally, for single crystal work, crystal should be spherical so as to have the same absorption factor in all directions.

**R- Factors and structure determination**
The measure of agreement between the individual, scaled $F_{\text{obs}}^{\text{shi}}$ and $F_{\text{calc}}^{\text{shi}}$ values is given by the residual factor or R-factor defined as follows:

$$R = \frac{\sum |F_{\text{obs}} - |F_{\text{calc}}| |}{\sum |F_{\text{obs}}|}$$

The values of R guides one to solve the unknown crystal structures among other parameters. The lower the value of R, more likely the structure to be correct.

It is not possible to give hard and fast rules about the relation between the magnitude of R and the likely correctness of the structure, but, usually, when R is less than 0.1 to 0.2, the proposed structure is essentially correct. A structure which has been solved fully using good quality intensity data has R typically in the range 0.02 to 0.06.

*Space lattice and Unit Cell*

Crystals have definite orderly arrangements of their constituents (atoms, molecules or ions) in three dimensions. The position of atoms, molecules or ions in a crystal relative to one another in space, are designated usually by points. Such a representation is called space lattice (i.e. an array of points showing how molecules, atoms or ions are arranged at different sites in a three dimensional space).

A unit cell is the smallest repeating unit in space lattice which when repeated over again results in a crystal of the given substance.
The three translations selected as the edges of the unit cell are called the crystallographic axes $a$, $b$, $c$ and the angles between them are called the interfacial angles, $\alpha$, $\beta$, $\gamma$ where

- $\alpha$ is the angle between $b$ and $c$ (opposite the $a$ axis)
- $\beta$ is the angle between $c$ and $a$ (opposite the $b$ axis)
- $\gamma$ is the angle between $a$ and $b$ (opposite the $c$ axis)

**Determination of unit cell contents**

The unit cell, by definition must contain at least one formula unit, whether it is be an atom, ion pair, molecule, etc. In centered cells and sometimes in primitive cells, the unit cell contains more than one formula unit. A simple relation may be derived between the cell volume, the number of formula units in the cell, the formula weights and the bulk crystal density. The density is given by

$$D = \frac{\text{mass}}{\text{volume}} = \frac{\text{formula weight}}{\text{molar volume}}$$

$$= \frac{FW}{\text{volume of formula unit} \times N}$$

Where $N$ is Avogadro’s number. If the unit cell, of volume $V$, contains $Z$ formula unit then

$$V = \text{volume of one formula unit} \times Z$$

Therefore,

$$D = \frac{FW \times Z}{V \times N}$$

$V$ is usually expressed as A$^{3}$ and density in grams per cubic centimeters.
Symmetry in crystals

Symmetry is the most characteristic property of the crystals. Symmetry in crystals may be understood in terms of symmetry operation and symmetry elements.

There are two nomenclatures for labelling symmetry element.

a. The Hermann–Mauguin system used in crystallography

b. And the schofield system used in spectroscopy

Both systems are well established, crystallographer require elements of space symmetry that spectroscopist do not, and vice versa and spectroscopist use a more extensive range of point symmetry than crystallographer.

The choice of unit cell and crystal system

Theoretically, there can be 32 different combinations of elements of symmetry of a crystal. These are called 32 point groups or 32 systems. Some of the systems however have been grouped together so that we have only seven basic crystal systems viz., Cubic, Orthorhombic, Tetragonal, Monoclinic, Triclinic, Hexagonal and Rhombohedral.

All crystallographic data were collected on a Rigaku AFC7R diffractometer with graphite monochromated Mo Kα radiation.
REFERENCES


