1.1 Introduction

Over the past ten years there has been a resurgence of interest in the coordination chemistry of lanthanide complexes in solution in general and in aqueous solution in particular. Enthusiasm in this work may be related to the enhanced appreciation of rich functionality of the ground and excited states of lanthanide complex.

Rare earth metal cation, Ln (III) are of great importance for their industrial [1], chemical [2], biochemical and medicinal application[3] due to their specific spectroscopic and magnetic properties. Lanthanide ion is a subject of increasing interest in bioinorganic and coordination chemistry. Lanthanide ions possess fascinating optical properties and their discovery, first industrial uses and present high technological applications are largely governed by their interaction with light. Lighting devices (economical luminescent lamps, light emitting diodes), television and computer displays, optical fibres, optical amplifiers, lasers, as well as responsive luminescent stains for biomedical analysis,
medical diagnosis, and cell imaging rely heavily on lanthanide ions. This critical review has been tailored for a broad audience of chemists, biochemists and materials scientists; the basics of lanthanide photophysics are highlighted together with the synthetic strategies used to insert these ions into mono- and polymetallic molecular edifices. Recent advances in NIR-emitting materials, including liquid crystals, and in the control of luminescent properties in polymetallic assemblies are also presented. The lanthanides (Ce–Lu) are unique among the elements, barring the actinides, in resembling each other so markedly in their chemical properties, particularly oxidation states [4,5]. The forbidden nature of the 4f-4f transition requires the use of antenna ligands which serve to facilitate energy transfer from the ligands excited state stimulating metal-centered luminescence [6]. The characteristic absorption and emission spectra of lanthanide compounds in the visible, near ultra-violet and near infra-red is attributed to transitions between 4f levels due to the fact that they present sharp lines, specially at low temperature, with oscillator strengths typically of the order of these transitions are electric dipole forbidden to first-order, but are allowed by the electric quadrupole, vibronic, magnetic dipole and forced electric dipole mechanisms.
It has been known for more than 50 years, that among these mechanisms only the magnetic dipole and forced electric dipole ones could account for the observed intensities. Trivalent state LnIII ([Xe] 4f n,n50–14) in aqueous solutions, in view of the various degrees of stabilization experienced by the 4f, 5d, and 6s orbitals upon ionization. The shielding of the 4f orbitals by the filled 5p 6s 2 sub-shells results in special spectroscopic properties with parity-forbidden 4f–4f absorptions having very low molar absorption coefficients and characteristic narrow-line emission, mostly in the visible and near infrared ranges. Luminescence has been instrumental in the discovery of several lanthanide elements. In turn, these elements have always played a prominent role in lighting and light conversion technologies (Auer mantles, incandescent lamps, lasers) and more recently in both cathode-ray and plasma displays. Presently, attention focuses on several potential applications of luminescent lanthanide ions[7].

The search for strongly luminescent lanthanide complexes, which can be excited near UV spectral region, motivated us to synthesize lanthanide (III) complexes contain aromatic N-donor ligands [8-9].
The coordinating chemistry of lanthanides, relevant to the biological, biochemical and medical aspects, makes a significant contribution to understanding the basis of application of lanthanide, particularly in biological and medical system. The importance of the application of lanthanides, as an excellent diagnostic and prognostic probe in clinical diagnostics and on anticancer material is remarkably increasing lanthanide chelates based contrast enhancing agents for MRI are being excessively used in radiological analysis in our body systems. Several hetrocyclic compounds containing pyrimidine nucleus in their structures have remarkable therapeutic significance due to their biological activity. In quest of exploring the chelating behaviour of some N,N,O and N,N,S-donor semicarbazone and thiosemicarbazones in several metal complexes, we could acquire more information about their nature of coordination and related structural, spectral and biological properties.[10].

The most important property of chelating agents, in lanthanide chelate complex, is its ability to alter the behavior of lanthanide ion with which it binds in biological systems and the chelation, markedly modifies the bio distribution and excretion
profile of the lanthanide ions. The chelating agents, especially aminopoly carboxylic acids, being hydrophilic, increase the proportion of their complex excreted from complexed lanthanide ion from biological systems. Lanthanide polyamino carboxylate chelate complexes are used as contrast enhancing agents for MRI. Conjugation of antibodies and other tissue specific molecules to lanthanide chelates has led to a new type of specific MRI contrast agents and their conjugated MRI contrast agents with improved relaxivity.

1.2 Object and Scope

It is reported that s-alkyl dithiocarbazate, thiosemicarbazone and benzimidazoles possess biochemical and biomedical and industrial uses, their activity further enhances because of ligand with F-block elements particularly La, Sm, Pr and Nd. This is supported by the fact that s-alkyl dithiocarbazates, thiosemicarbazones and benzimidazoles, the toxicity may be due to =N-C-S groups and -N = C group respectively. Which ligand with the active centers of the cell constituents resulting in interference with the normal cell process. The higher bacteriotoxicity experienced by the compounds / complexes may
be ascribed to the fact that the ligand are more susceptible towards the bacterial cells than fungicidal cells.

Various physicochemical methods have employed to characterize the complexes. The spectral data suggests that coordination of the ligand to central Ln atom takes place in a tridentate fashion coordinating to the metal ion through ONO donor sequence. In view of the great deal of importance lanthanide complexes including s-alkyl thiocarbazate, thiosemi carbazole and benzimidazole. The spectral properties of some complexes have been undertaken and the results of this investigations are presented here.

1.3 Review Literatures

1.3.1 Schiff bases

Schiff bases are typically formed by the condensation of a primary amine and an aldehyde/ketone. The resultant compound, \( R_1R_2C=NR_3 \), is called a Schiff base (named after Hugo Schiff), where \( R_1 \) is an aryl group, \( R_2 \) is a hydrogen atom and \( R_3 \) is either an alkyl or aryl group. However, usually compounds where \( R_3 \) is an alkyl or aryl group and \( R_2 \) is an alkyl or aromatic group are also regarded as Schiff bases. Schiff bases that contain aryl
substituents are substantially more stable and more readily synthesized, while those which contain alkyl substituents are relatively unstable. Schiff bases of aliphatic aldehydes are relatively unstable and readily polymerizable [11], while those of aromatic aldehydes having effective conjugation are more stable. In general, aldehydes react faster than ketones in condensation reactions, leading to the formation of Schiff bases as the reaction centre of aldehyde are sterically less hindered than that of ketone. Furthermore, the extra carbon of ketone donates electron density to the azomethine carbon and thus makes the ketone less electrophilic compared to aldehyde [12]. Schiff bases are generally bidentate (1), tridentate (2), tetradequate (3) or polydentate (4) ligands capable of forming very stable complexes with transition metals. They can only act as coordinating ligands if they bear a functional group, usually the hydroxyl, sufficiently near the site of condensation in such a way that a five or six membered ring can be formed when reacting with a metal ion (Fig. 1.1). Schiff bases derived from aromatic amines and aromatic aldehydes have a wide variety of applications in many fields, e.g., biological, inorganic and analytical chemistry [13, 14]. Applications of many new analytical devices require the presence of organic reagents as
essential compounds of the measuring system. Schiff bases are used, e.g., in optical and electrochemical sensors, as well as in various chromatographic methods, to enable detection of enhanced selectivity and sensitivity [15-17]. Among the organic reagents actually used, Schiff bases possess excellent characteristics, structural similarities with natural biological substances, relatively simple preparation procedures and the synthetic flexibility that enables design of suitable structural properties [18, 19]. Schiff bases are widely applicable in analytical determination, using reactions of condensation of primary amines and carbonyl compounds in which the azomethine bond is formed (determination of compounds with an amino or carbonyl group);

$$\text{C} = \text{N} \quad \text{OH}$$

Bidentate (1)

$$\text{R}_1$$

$$\text{R}$$

$$\text{R}$$

$$\text{R}_1$$

$$\text{R}_2$$

$$\text{NR}_1\text{R}_2$$

Tridentate (2)
Fig.1.1 Some Classes of Schiff base ligands

using complex formation reactions (determination of amines, carbonyl compounds and metal ions); or utilizing the variation in their spectroscopic characteristics following changes in pH and solvent [20]. Schiff bases play important roles in coordination chemistry as they easily form stable complexes with most transition metal ions [21, 22]. In organic synthesis, Schiff base reactions are useful in making carbon-nitrogen bonds.
Lanthanide Schiff base complexes have been known since the middle of nine tenth century. However, there was no comprehensive and systematic study until the preparative work of Pfeiffer and associates. This work still serves as a source of chemical and preparative detail for the contemporary chemists and its importance may gauged by the relatively large number of citations of these papers in the modern literature. Since Pfeiffer initial contributions the interest in schiff base complexes has increased significantly. However, only over the last two decades with the advent of suitable techniques i.e. structural electronic, infrared and magnetic properties etc. have been measured and explains in some depths. Recently a good deal of work has been done on the structural aspect of schiff base complexes. In order to provide further insight in their physico-chemicals behavior the nature of chemical bonding, coordination number, oxidation state of lanthanide complexes and stereo chemistry.

Schiff bases condensation product of an active carbonyl compound and a primary amine are known to process bacteriostatic, tuberculostatic and fungicidal activities. They can be easily prepared because of their systamic flexibility by selecting
the appropriate reactants with different substituents. By Changing
the nature and positions of donor atoms and groups. It is possible
to control the size of the ring formed during complexation and the
explain the effect of substitution. phenolic compound in which the
azomethine group is situated in the ortho positions of hydroxy
group are known to form complexes exhibit antibacterial and
fungicidal properties.

Schiff bases appear to be important intermediates in a
number of enzymatic reactions involving interaction of the amino
group of an enzyme, usually that of a lysine residue, with a
carbonyl group of the substrate [23]. Stereochemical investigations
[24] carried out with the aid of molecular models showed that
Schiff bases formed between methylglyoxal and the amino group
of the lysine side chains of proteins can bend back in such a way
towards the N atom of peptide groups that a charge transfer can
occur between these groups and the oxygen atoms of the Schiff
bases. Schiff bases derived from pyridoxal (the active form of
vitamin B6) and amino acids are considered as very important
ligands from biological point of view. Schiff bases are involved as
intermediates in the processes of non-enzymatic glycosylations.
These processes are normal during aging but they are remarkably accelerated in pathogeneses caused by stress, excess of metal ions or diseases such as diabetes, Alzheimer’s disease, and atherosclerosis. Non-enzymatic glycosylation begins with an attack of sugar carbonyls or lipid peroxidation fragments on amino groups of proteins, amino-phospholipids and nucleic acid, causing tissue damages by numerous oxidative rearrangements. One of the consequences is cataract of lens proteins [25]. Many biologically important Schiff bases have been reported in the literature possessing, antimicrobial, antibacterial, antifungal, anti-inflammatory, anticonvulsant, antitumor and anti HIV activities [26-31]. Another important role of Schiff base structure is in transamination [32]. Transamination reactions are catalyzed by a class of enzymes called transaminases. Transaminases are found in mitochondria and cytosol of eukaryotic cells. All the transaminases appear to have the same prosthetic group, i.e., pyridoxal phosphate, which is covalently attached to them via an imino group. Schiff base formation is also involved in the chemistry of vision, where the reaction occurs between the aldehyde function of 11-cis-retinal and amino group of the protein (opsin) [33]. The biosynthesis of porphyrin, for which glycine is a precursor, is
another important pathway, which involves the intermediate formation of Schiff base between keto group of one molecule of δ-amino levulinic acid and ε-amino group of lysine residue of an enzyme.

Some Schiff bases derived from S-methyldithiocarbazate and S-benzyldithiocarbazate exhibit potential antiamoebic activity [34]. The properties of benzimidazole and its derivatives have been studied over more than one hundred years [35]. However, a special interest of researches towards this class of compounds was stimulated by the fact that 5,6-dimethyl benzimidazole is a component of vitamin B₁₂ [36]. Shortly after, it was established that, albeit vitamin B₁₂ is capable of stimulating the growth of bacteria, the benzimidazole fragment and some of its derivative suppress the bacterial growth. This discovery was followed by attempts to create new antibacterial preparations based on the benzimidazole derivatives. A series of halogen substituted 5,6-dimethylbenzimidazole 5 derivatives exhibit pronounced antibacterial and antiviral properties. This was explained by their competition with purines resulting in inhibition of the synthesis of microbial nucleic acid and proteins [37-42].
Derivatives of pyrimidobenzimidazole 6 represent a new class of DNA gyrase inhibitors, which are effective antibacterial agents [43].

Antimicrobial and antifungal properties were reported for 3-alkylthiomethyl-1-ethyl, 3-alkoxymethyl-1-butyl and 3-alkylthiomethyl-1-butylbenzimidazolium chloride, as well as for 2-chloromethyl-5H-methyl-benzimidazoles [44,45]. Some 5-nitrobenzimidazole derivatives also exhibit fungicidal activity [46], certain antimicrobial and antifungal potential was observed in heterocyclic benzimidazole derivatives[47]. Antiparasitic activity of 2-(trifluoromethyl) benzimidazole 7 was reported by Gabriel
against Giardia lamblia, Entamoeba histolytica and the helminth Trichinella spiralis [48].

\[ \text{2-(trifluoromethyl)benzimidazole(7)} \]

The class of benzimidazole derivatives contains compounds possessing affinity to DNA and produces non-specific inhibition of the catalytic activity of many enzymes involved in DNA synthesis including DNA polymerase [49]. Another promising group of antitumour compounds is represented by benzimidazoquinazolines 8 and thiazolebenzimidazole [50,51].

\[ \text{Benzimidazoquinazolines(8)} \]

Some bimethyl (2 ethylthiobenzimidazole hydrobromide) are capable of correcting the mutagenic action of xenobiotics [52,53]. At present, more than twenty benzimidazole derivatives are used
as antihelminth preparations in the world of veterinary and medical practice, including thiabendazole and parbendazole [54], oxofendazole and flubendazole [55], fenbendazole [56], triclabendazole [57], oxibendazole and cambendazole [58], luxabendazole 9[59], mebendazole 10 and albendazole 11[60].

**Luxabendazole (9)**

**Mebendazole (10)**

**Albendazole (11)**
The helminthic activity was also found in some derivatives of 3-(benzimidazole-2-yl)-2-propanoic acid and 2-methoxycarbonyl amino-5-(4-salicyloylpiprazin-1yl) benzimidazole [61,62]. 2-Methylthiobenzimidazole-4, 7-dione 12 showed potent antiproliferative activity [63].

\[
\begin{align*}
R & \quad \text{SCH}_3 \\
\text{O} & \\
\text{N} & \\
\text{O} & \\
\text{O} & 
\end{align*}
\]

2-Methylthiobenzimidazole-4,7-dione (12)

Thiosemicarbazones (TSCN) and semicarbazones (SCN) is a class of compounds obtained by condensing thiosemicarbazide or semicarbazide with suitable aldehydes or ketones. The chemistry of thiosemicarbazides, thiocarbohydrazides, thiosemicarbazones, thiocarbohydrazones and their cyclised products and related compounds has evinced considerable interest due to their biological activity and industrial importance. A large number of thiocarbohydrazones and thiosemicarbazones substituted with sulphur and nitrogen are generally more versatile intermediates with respect to the oxygenated ones [64,65].
Thiosemicarbazones and thiocarbohydrazones are expected to be facile intermediate in the preparation of individual sulphur derivatives of steroids as well as non-steroids. The derivatives of thiosemicarbazones and thiocarbohydrazones have been reported as antibacterial and herbicidal agents [66-69]. Some of the thiosemicarbazone derivatives were also tested for potential antitumor [70, 71] and antithyroid activity [72] including their antiviral activity [73,74]. Various thiocarbohydrazones derivatives have also been investigated as radioprotector [75], anticonvulsant [76], ulcer inhibitor and anticancer agents [77-79]. Recently, thiocarbohydrazones derived from 2-acetyl pyridine have been found potential antimalarial [80], antitrypanosomal [81] and antifilarial [82] agents. Semicarbazones of aromatic and unsaturated carbonyl compounds have anticonvulsant properties and their advantage over the analogous thiosemicarbazones is their lower neurotoxicity. Vanadium(V) complexes with salisilaldehyde semicarbazones derivatives show in vitro antitumor activity towards kidney tumor cells. Leovac et al. reported the physicochemical and structural characteristics of Ni(II) complexes with pyridoxal semicarbazones. Dimmock and Baker reported the anticonvulsant activities of 4-bromo benzaldehyde
Jagst et al. studied the synthesis and structural characterization of lanthanide complexes with pentadentate asymmetric ligands system derived from 2,6-di-acetylpyridine or 2,6-diformyl pyridine with mixed semicarbazone /benzoyl hydrazone and semicarbazone/ thiosemicarbazone coordination sites. Semicarbazones and thiosemicarbazones are biologically important nitrogen and oxygen/sulphur donor ligands. Computational method is at present widely used for simulating IR spectrum. Such simulations are indispensable tools to perform normal coordinate analysis so that modern vibrational spectroscopy is unimaginable without involving them. In the present study, the IR, Raman and theoretical calculations of the wavenumbers of the title compound are reported. Nonlinear optics deals with the interaction of applied electromagnetic fields in various materials to generate new electromagnetic fields, altered in wavenumber, phase, or other physical properties. Organic molecules able to manipulate photonic signals efficiently are of importance in technologies such as optical communication, optical computing, and dynamic image processing [83].

Thiosemicarbazones derived from arly / alkyl ketones /
aldehydes and related compounds showed neurotoxic activity in mice and rats. The antiviral activity of thiocarbohydrazones was reported first in 1950 by Hamre et al. [84] who found that derivatives of benzaldehyde thiocarbohydrazones were active against neurovaccinal infection in mice when given orally. The thiosemicarbazone of isatins was found to be one of the most active compounds against \textit{ectromelia} infection [85] and a clinical trial of the N-methyl derivative of isatin-\(\beta\)-thiosemicarbazone (methisazone) was carried out in India [86,87]. The studies indicated that the drug was effective in the prevention of smallpox. Although these studies have been widely accepted as evidence of the effective antiviral activity of methisazone in humans, a subsequent field trial study demonstrated little efficacy [88]. The drug has been also used to treat patient with genital lesion caused by \textit{Herpes simplex} virus, but it had little effect on the severity or duration of the lesion [89].

Easman et al. [90] prepared thiosemicarbazone 14 from corresponding alkyl pyridazinyl kethones 13 by the process of condensation with thiosemicarbazone.
Salman et al. [91] prepared 4-acetylantripyrine-4-substituted thiosemicarbazone derivatives 16 by the reaction of antipyrine 15 with thiosemicarbazide.

Leng et al. [92] converted 2-acetylbenzimidazole 17 to thiosemicarbazone 18 by reacting with thiosemicarbazide.

A series of 3-and 5-alkyl amino derivatives of 2-pyridine carboxaldehyde 19 thiosemicarbazones were synthesized and evaluated as inhibitors of CDP reductase activity and cytotoxicity in
*vitro* and antineoplastic activity *in vivo* against the L 1210 leukemia. The most compounds were 20 [92].

Bernstein et al. [93] reported the preparation of arylketone thiosemicarbazone 22 by the reaction of arylketone 21 with thiosemicarbazide.

Islam et al. [94] reported the synthesis of aldehyde thiosemicarbazones 25 by the reaction of aromatic aldehyde 23 with thiosemicarbazide 24.
Dimmock et al. [95] subjected arylmethylketone 26 to react with thiosemicarbazide, which afforded thiosemicarbazone 27.

Cveary and coworkers [96] have reported the preparation of a-N-heterocyclic carboxaldehyde thiosemicarbazone 28.

David and Tony [97] prepared heterocyclic thiosemicarbazone 30 by the reaction of heterocyclic aldehyde 29 with thiosemicarbazide.
2- Acetylpyridine thiosemicarbazones were synthesized by Klayman et al. [99] and have been found to exhibit antimalarial activity in mice infected with *Plasmodium berghei*.

![Chemical structure](image)

In these studies, it was noted that such activity was limited to those compounds in which the alkylidene group was attached to the 2-position rather than the 3 or 4-position of the pyridine ring and also to those in which a thiocarbonyl group rather than a carbonyl group is present [100]. Klayman et al. [101] reported selenosemicarbazones of 2-acetylpyridine and related thiosemicarbazides, which possess antimalarial activity.

![Chemical structure](image)

Scovill et al. [102] synthesized the derivatives of 2-
acetylpyridine 1-oxide thiosemicarbazone 34 from 2-acetylpyridine 1-oxide with methyl hydrazine-carbodithioate and observed their activity as antimalarial agents.

\[
\text{N} \quad \text{C} \quad \text{H} \quad \text{N} \quad \text{C} \quad \text{N} \quad \text{R}_1 \text{R}_2 \\
\text{O} \quad \text{S} \quad \text{C} = \text{N} \text{NCN} \\
\text{CH}_3 \\
(34)
\]

Several investigations on pyruvic acid (HPy), as well as compounds derivatives of HPy, have been carried out in biological science research. However little works on salts of pyruvic acid (H3C-CO-COOH) have been described in the literature. In aqueous solutions the formation of some metal ions complexes with pyruvic acid in the ratio of metal: ligand 1:1 and 1:2 have been established by the spectroscopic method. The stability constants and thermodynamic functions of complexes formation of lanthanides with pyruvic acid: \( \Delta G, \Delta H, \Delta S \), have also been determined. In the solid state the preparation of europium pyruvate, as well as the preparation and properties of lanthanides and yttrium pyruvates have also been described. The aim of this work has been to obtain light trivalent lanthanide pyruvates in solid state and to investigate
by means of complexometry, elemental analysis, X-Ray powder
diffractometry, infrared spectroscopy, thermogravimetry (TG) and
differential scanning calorimetry (DSC). metal chloride or nitrate.
The solutions were maintained in water bath until total precipitation
of the metal pyruvates and the precipitates washed with hot
distilled water to eliminate the chloride (or nitrate ions), then
filtered through and dried on Whatman nº 42 filter paper, and kept
in a desiccator over anhydrous calcium chloride. After igniting the
compounds to the respective oxides (CeO₂, Pr₆O₁₁ and Ln₂O₃,
Ln = La, Nd, Gd) the residues were dissolved in a hot solution of
concentrated hydrochloric acid or a hot solution comprising a
mixture of concentrated hydrochloric acid and hydrogen peroxide
for cerium and praseodymium oxides, and their lanthanides
contents were determined by complexometric titration with
standard EDTA solution, using xylenol orange as indicator. The
lanthanides contents were also estimated from their
responding TG curves. The dehydration of the compounds was
firstly pointed out by their DTG curves and subsequently confirmed
by the broad endothermic peaks centered at 75–175°C in the
respective DSC curves. The water contents were then determined
from the corresponding mass losses observed in the TG curves.
Next, the ligand content was also assessed from the TG curves. X-Ray powder patterns were obtained by using a Siemens D-5000 X-Ray diffractometer with CuKα radiation (λ = 1.541 Å) and under 40 kV and 20 mA settings. Infrared spectra for sodium pyruvate as well as for its metal-ion compounds were recorded on a Nicolet model Impact 400 FTIR Instrument in 4000-400 cm\(^{-1}\) range. The solid samples were pressed into KBr pellets[103].

### 1.3.2 Lanthanides

Lanthanides are members of a large family of elements located between barium and hafnium in the 6\(^{th}\) row of the periodic table. The name of rare earths covers lanthanides and some other elements (yttrium, scandium) usually mixed together in the ores. Lanthanides are no more considered as rare, since industrial processes allow to prepare them in high degree of purity. There is an increasing demand of some lanthanides for modern technology. Rhone Poluenc Co. is the world leader for production of purified lanthanides. Cerium is the most abundant element in the lanthanide series, followed by lanthanum and samarium. Thullium is the rarest lanthanide but it still remains four times more abundant than silver on the earth crust. Apart from cerium (ceric
oxidations) lanthanides have been for a long time neglected by organic chemists. However in the last decade there has been a progressive change in the attitude, a recent review [103] summarizes the use of lanthanides in organic synthesis.

Z  57  58  59  60  61  62  63  64  65  66  67  68  69  70  71
Ln La Ce Pr Nd Pm Sm Eu Gd Tb Dy Ho Er Tm Yb Lu

The Lanthanum Series

Several features concerning lanthanides are of potential interest in organic chemistry:

Main oxidation states : 3.

Oxidations possible by tetravalent lanthanides (mainly carium).

Reductions possible by divalent lanthanides (Eu, Yb, Sm, Tm).

Rich coordination chemisty with high coordination numbers.

Lewis acidity, especially for Ln (III) derivatives, with application
to catalysis.

Oxophilicity.

The 4-f orbitals do not much participate to bonding, most of Ln derivatives have a pronounced ionic character.

Redox chemistry with lanthanides involves monelectronic transfers (Ce(IV) → Ce(III) and Ln(II) → Ln(III). In the present article wish to discuss the use of divalent lanthanides in organic chemistry and then to present some results we have recently obtained[104].

Lanthanides, the fifteen chemically similar elements, as such occur only in traces in whole body assay analyses. The amount of lanthanides occurring in different organs shows significant accumulation of these metals, reported in kidney, liver, bones and spleen; however, the remaining organs contain only much smaller concentrations of lanthanides. The amount of lanthanides in eyes has been found to vary in a wide range. The most important and strikingly noticeable part of lanthanide biochemistry is the observation made in a number of studies that the concentration of lanthanides accumulated in different organs varies widely with the progress of different stages in diseases [3].
Webster [105] reported much higher lanthanide accumulation in infarcted cardiac tissues, as compared to the normal ones, as early as in 1965. Esposito and coworkers could locate dramatic upward changes of lanthanide level in the synovial fluid of patients suffering from rheumatoid arthritis of the joints[106,107] This observation led these workers to propose that lanthanides are "excellent markets" for the "diagnosis and prognosis" of cancer in bones. The lanthanide accumulation has been systematically examined in spleen where it is found that lanthanide level showed regular variation with different degree of infection of the organ in alcoholic persons. Their findings were later on extended to the investigations of liver occurrence of lanthanide, because liver is among the organs which shows great preference for lanthanide accumulation. Their findings suggested that liver which showed great propensity for lanthanide accumulation is damaged and hence becomes less elective in sequestering lanthanides in humans with prolonged alcoholic addiction. This therefore led to significant spilling of lanthanide over spleen, the organ which has a large accumulation of reticuloendothelial cells like liver and hence one could explain higher levels of lanthanides in the spleen of alcoholic persons. The concentration of lanthanides in malignant
laryngeal tissue was found to be significantly lower than the normal ones; however, no noticeable lanthanide could be detected in erythrocytic lysate from patients suffering from malignancy of laryngeal tissues [106]. The concentration of lanthanide showed a dramatic spurt (up to 12 fold) in patients with laryngeal carcinoma than the found in normal persons [108,109]

The toxicity of a non-essential metal ion such as Ln$^{+3}$ can be determined by its degree of deviation from relevant essential metal ion, such as Ca$^{++}$ as reference. The deviation spans the whole range of similarity with respect to Ca$^{++}$. Among the factors determining, how far these metal (Ln) ions deviate from Ca$^{++}$, softness, covalency and redox tendency are the most decisive. The high toxicity of other heavy metal ions or even metals (Hg, Au, Pt, Pb etc) is due to strong deviations in these aspects. Their toxicity is therefore obvious and cannot be avoided. Contrary to this, lanthanides are very similar to Ca$^{++}$ in these aspects. The deviation, however, originates due to deviation in charge, ionic radii and the presence and involvement of inner lying 4f orbitals, and these deviations lead to minor adverse effects, which are related to the level of lanthanides in vivo. The high spin
paramagnetism and long electron relaxation times of Gd(III) have made it preeminent among the contrast enhancing agent for "Magnetic Resonance Imaging" (MRI) [110,111]. Related complexes of Dy(III) and Tm(III) with much shorter electronic relaxation times are very effective NMR shift reagents [112]. The controlled modulation of Lewis acidity across the lanthanide series allows the development of the lanthanide complexes exhibiting phosphatase activity, while the redox activity of cerium, samarium, europium and ytterbium may be expected to allow the development of selective oxidants and reductants [113]. Lanthanide complexes in solution exhibit a well defined luminescence which is characterized by narrow emission bands, large stokes shifts and long excited state lifetimes in aqueous solution up to 5 minutes which emit in red and green. This has been used in fluoroimmunoassays [114-117] and shows considerable promise for being used, in luminescence imaging and as sensors for certain bioactive ions [118-120]. Lanthanide (III) also gives characteristic 4f-4f transition bands, which are sharp, narrow and Lappporate forbidden in nature. Under certain conditions created by coordination of certain types of chelating ligands including biomolecular ligands, some of the 4f-4f intra
configurational transitions undergo substantial intensification and high sensitivity, towards even minor coordination changes. Such coordinational changes are the outcome of the conformational changes of structural changes which occur during complexation with paramagnetic Ln (III) ions [121-123]. Comparative absorption spectroscopy like luminescence spectroscopy can also be used in certain biological systems to probe the structural, conformational and even changes in the biological activities of biomolecules when these are coordinatively bonded to paramagnetic lanthanides (III) ion [122-125].

The above changes are necessary for the entry of Ln(III) inside the cell. The metal uptake on the cell membrane and strong attachment to the external surface of the bilayer cell membrane can be due to phosphate, which forms strong Ln(III) complex through polar phosphate end as given in Fig. 1.2 Lecithin (phosphatidyl choline) being the most prominent phospholipid biomembrane, forms 1:2 complex with Ln(III).
The binding of lanthanide ion occurs at PO$_2$ groups, while the binding of different lanthanide ions for vesicle bilayer is different, most probably due to the presence of interconvertible high affinity sites which are also known as Relaxed (R) sites, low affinity sites and Tense (T) sites. The R/T ratio is fixed and stands around 0.14. The addition of Ln(III) ion increases R/T ration significantly as a result of conversion of T sites to R sites. This interconversion is mainly due to conformational changes induced by different Ln(III) ion. Paramagnetic Ln(III), due to their NMR characteristics, help in distinguishing outer and inner polar heads of bilayer vesicles. Bentz and coworkers [127] have reported that in the presence of larger concentrations of Ln(III) in, their role changed and this initiated the disruption of fusion of unilamellar phosphatidyl serine liposome, most probably by altering the overall charge on the...
surface of the vesicle. During the process of fusion of vesicles, a leakage of intracellular contents took place. No doubt the role of Ln(III) is much faster and stronger than the role played by Ca(II) in the fusion of vesicle where the leakage of intracellular contents took place [127]. El-Fakahany et al. used EM (electron microscopy) EDAX (energy disperse analysis of X-rays) studies in explaining Ln(III) binding to biological membranes and their findings conclusively remarked that the distribution of Ln(III) was basically irregular and was in the form of "clusters" named as "Hot Spots". These hot spots or confined areas of lanthanide accumulation most probably play the role of Ca(II) channel around receptor [128].

The above findings have been found very effective in the exploring lanthanide compounds (salts, complexes as well as coordinated chelates) in drug development as well as during diagnosis and prognosis of diseases like multiple sclerosis, atherosclerosis, cerebrospinal and cardiovascular, and oncological diseases [126, 127, 129, 130]. Very recently immensely useful investigations have been carried out, involving lanthanide induced perforation of cell membrane. In gene recombination, the critical
step that is necessary is to promote transformation of plasmid in bacteria by incubation with CaCl$_2$. The elevation in the permeability induced by Ca(II) was the main factor in perforation mechanism [131, 132]-. Want et al. [133,134]. reported that the perforation of membranes was caused by Ln(III) even when these are administered in low concentration. Huang et.al. [135] found Ln(III) ions like Ca(II), when applied Ln(III) enhanced the transformation of plasmid pBR 322 and PUC 18 in E.coli and highest transformation required a dose as low as 10$^{-5}$M of Ln(III). No doubt higher concentration of lanthanide application inhibited plasmid transformation. Canada et. al. [136] reported earlier that Ln(III) binding to cell surface is always accompanied by significant physiological changes like rapid increase in membrane potential especially in Ehrlich ascite tumor cells and this type of lanthanide (Tb$^{3+}$) binding also led to substantial changes in electrophoretic behaviour. However, in their latter publication Canada et. al. [136]found tthat Tb(III) increases significantly the intracellular accumulation of cisplatin. Their observation was soon supported, though indirectly, by permeability increase induced by Ln(III) reported by Wang et. al., who proposed the mechanism for perforation induced by metal ion like Ln(III). Hence this has been
used in the leaking of hemoglobin from erythrocytes by the presence of lanthanide(III) salts or compounds [135]. Their findings [135-137] showed that lanthanide aquo complexes were quite effective in perforation of erythrocytes. Complexation interaction is a triphasic process, with perforation occurring in the second stage. This stage was characterized by sustainable and recoverable hemolysis. EDTA washing could lead to resealing of the membrane of erythrocytes. These findings could prove that Ln(III) complexation with cell surface-active centre involves a predominantly electrostatic interaction [136-138]. There are two types of pores, domain and crater shaped pores, which result due to different concentrations of Ln(III) used during investigations. Ln(III) binding incurs conformational changes followed by aggregation of membrane proteins [139-141]. Since lanthanide(III) ions in aqueous medium are always in the form of a non-coordinated stereochemistry and in biofluids these undergo multimetal multiligand complexation due to the presence of endogenous metal ions and physiological ligands. Therefore even when Ln(III) ions are used, to alter the biological properties in Ln(III) and biological substrate interaction, complexation is undergone with the binding sites of the biological substrate by
partial substitution of coordinated water molecules surrounding tervalent lanthanide (III) in [Ln(H$_2$O)$_9$]$^{3+}$. The extent of substitution depends on several factors like pH, composition and in vivo chemical environment. The binding sites of the biological substrate which are preferred by Ln(III) for complexation involve donor sites O>F>Cl>N<S. The inherent strong oxyphilicity of lanthanide causes the interaction binding sites being COOH, OH (phenolic) OH (hydroxylic), O (carbonyl), N (amino, imido, imino), S (sulphhydryl). Nitrogen sulphur donor sites biomolecules also enter into complexation when Ln(III) undergoes chelation.

A number of techniques like Atomic Force Microscopy (AFM), $^1$H, $^{13}$C, $^{31}$P NMR, FT-IR luminescence and absorption spectroscopy have been found very useful in such studies. Lanthanides consuming, Reactive Oxygen Species (ROS), which are mainly the oxygen derived free radicals and peroxides, are the mediators of a number of degenerative diseases. The 'antioxidants' are excellent substances used as drugs for the treatment of degenerative diseases. Tocopherol, ascorbates and a number of other organic compounds are considered as components of such drugs due to their antioxidant property shown
towards ROS induced degenerative diseases. Lanthanides are considered of high potential because of their inherent antioxidant properties [125]. Wu et al. [142] found LaCl$_3$ effective in inhibiting silica induced lipid peroxidation of lung macrophages and smaller doses of some other lanthanide chlorides inhibited lipid peroxidation in rat lung, Wang et al. found almost all lanthanide compounds, especially chlorides, quite effective in inhibiting H$_2$O$_2$ mediated peroxidation of liposomes; however, when terbutyl hydroperoxide was used to mediate the peroxidation, lower lanthanides inhibited while the higher lanthanides promoted peroxidation. Interestingly Ln(III) lost reactivity of peroxides when they were bound to membrane. The prior lanthanide (III) became more sensitive to oxidation attack. The lanthanide inhibiting ROS involves strong oxyphilicity inherent in lanthanides, because of the availability of oxygen sites on these free radicals, makes them excellent targets for Ln(III) coordination (attack). This causes lanthanide to play the role of scavenger of reactive oxygen species, therefore presenting good potential for lanthanide as a future drug for a number of degenerative diseases due to ROS. No doubt the involvement of lanthanide in ROS removal is quite different from the inhibition of ROS by organic compounds like
Tocopherol, Ascorbate etc. Most of the organic antioxidants scavenge free radicals by single electron exchange with radicals and thus transform themselves into radicals, hence acting as "pro-oxidants". Ln$^{3+}$ very easily interacts with either free radicals or peroxides but is not transformed as radicals. However the mechanistic understanding about the role of Ln(III) as scavenger of antioxidant is very meager.

The liability of lanthanide complexes, strong oxyphilicity, very fast water exchange reaction, non-directionality of lanthanide ligand bond and varying coordination number, all contribute towards lanthanide interaction with bio-molecules. The ionic size of Ln(III) varies from one lanthanide to another lanthanide; in addition, the ionic size of a particular lanthanide also varies significantly with the coordination number.

Smaller size of chelating biomolecular ligand can even suit larger lanthanides with lowered coordination number. Similarly small lanthanides can expand their coordination number and can form stable chelates with larges biomolecules. This explain the different coordinating power (also biological behaviour) or different lanthanides under different physiological conditions.
Significant disorganization in cytoskeleton including microtubules and microfilaments is a phenomenon of common occurrence in tumor cells and apoptotic sized cells. Microtubules undergo stabilization and repair and this occurs during the action of some anticancer drugs like TAXOL. Depolymerization of cytoskeleton is one of the most relevant steps in the apoptosis process. Lanthanide compounds have been shown to influence the stability of microtubules. Xiao et al. [143] found mixed lanthanide compounds, increasing the amount of orderliness of microtubules in PAMC82 cells. Soto et al. [144] found different lanthanides showing different behavior and this was ascribed to similarity of Ln(III) predominantly with Ca$^{++}$ or similarity of Ln(III) with Mg$^{2+}$, because Ca$^{++}$ ion was found to destabilize microtubules contrary to the strengthening of microtubule activity induced by Mg$^{2+}$. The lower lanthanides behaved more like Ca$^{++}$ because of their larger size and hence propensity of exhibiting relatively higher coordination number. Heavier lanthanides like Tb, Dy, Ho being smaller in size behaved more like Mg$^{2+}$ having more potential for strengthening the microtubules. Soto et al. attributed the role of Ln(III) and different behavior of different lanthanides to their effect of GTPase activity of tubulin. The microtubules formation tubulin-
GTP-Mg$^{++}$ system is a multiple step process, where the role of Mg$^{2+}$ is very important because it binds to tubulin and thus modulating the conformation to favour self association of tubulin as shown in Fig. 2.

![Diagram](image)

**Fig.1.3 : Probable steps involved in the formation of microtubule.**

As we have explored the complexation of mononucleotides with paramagnetic lanthanide using nucleotide, mono, di, and triphosphates, in aqueous and in aquated organic mediums at pH as low as 1.00 to as high as 6.5, using 4f-4f transition spectroscopy $^1$H NMR and $^{31}$P NMR spectroscopy. Sudhran Misra has found that different lanthanides showed different stability of Ln-monomononucleotide complexes. The nature of the nucleoside moiety and the number of phosphate groups significantly affected the degree of complexation. The size of Ln(III) ion also played an important role.

Comparative absorption spectroscopy, involving electric
dipole Laporte forbidden 4f-4f transitions, have shown that different mononucleotides showed different affinities towards Ln(III). In general the binding of Ln(III) with nucleotides derived from pyrimidine bases is weaker than the binding of Ln(III) with nucleotides derived from purine bases, irrespective of the nature of the experimental conditions. Sudhram Misra has also observed that in aqueous medium lanthanide interaction with monomucleotides shows the presence of both syn and anti conformation. The increase in the organic solvent percentage resulted in the shift in the equilibrium towards anticonformations of the Ln-nucleotid complex [145-148].

Studies also showed that in aqueous solutions, Ln(III) initiates the hydrolysis of NTP (nucleotide triphosphate) and this reaction was appeared to be dependent on H⁺ ion concentration and the nature of lanthanides [148-151].

The important steps in the association of tubulin are GTP hydrolysis and binding to GTP, and both of these activities are governed by Mg²⁺. With association process, setting in the size of regulatory effect proceeds and also consequently controls the shape and size of microtubule. Ln³⁺ when administered in small
doses behaves like Mg$^{2+}$ supporting the association of tubulin. However when administered in high doses, Ln$^{3+}$ interferes with the assembly, by distorting the protein conformation, altering crosslinking and consequently destabilizing the polymers.

In 1931 Maxwell and coworkers [152] used in aqueous solution of lanthanum chloride for treating cancer by administering LaCl$_3$ solution intraperitoneally. However, it was only the work of Anghileri and coworkers which could successfully demonstrate the strong inhibitory effects of LaCl$_3$ and other lanthanide compounds on the growth of sarcoma tumors in rats. Excellent work as come out of Anghileri's laboratory, on lanthanide compounds and complexes in cancer research as a diagnostic and prognostic probe [153-156]. These workers used Ln(III) as an adjunct to the distraction of tumors by using a combination of the complexes of two different lanthanides specially derived from hydroxy carboxylic acids for treating animals and also in some cases involving humans suffering from Yoshida Sarcoma. The results were found astounding [156].

In preliminary studies S. Mishra used a combination of two or even three different lanthanide complexes derived from citric
and mandelic acids on rats with Yoshida Sarcoma and found encouraging results [157, 158]. The synthesis and reactivity of these citrates, mandelates and tertrates have been reported from laboratories in 1966. Though study is quite preliminary, the results are very promising. Lanthanide citrates and mandelates, when administered along with drug hematoporphyrins, showed drastic reduction of growth of Ehrlich ascite cells, much better than that obtained by using hematoporphyrin alone. We attributed this to much improved absorption of the drug due to the presence of lanthanide coordination complexes [159].

The complexes of lanthanides are getting more and more applications in cancer therapy and the most important of these are those derived from poly (amino-carboxylic) acids. The formation constants of the lanthanide chelates with these acids are of the order of $10^{20}$ to $10^{25}$, which enables them to remain intact, while diffusing into extracellular spaces with rapid clearance through kidneys. Due to the high thermodynamic stability and extreme kinetic inertness of thee poly (amino-carboxylates) of Ln(III), the intact excretion enhances thereby lowering considerably the body retention of chelated Ln(III) complexes.
These days diagnostic imaging procedures [160, 161] are a routine part of modern medicine and are useful in performing the initial diagnosis, the planning of the treatment and post treatment evaluation.

Even with the recent phenomenal growth of magnetic resonance imaging (MRI) and ultrasound procedures, X-ray imaging studies remain even today the workhorse of modern radiology. Currently 75-80% of all diagnostic imaging procedures, as listed in Table 1, are X-ray related [161-166].
<table>
<thead>
<tr>
<th>Body System</th>
<th>Diagnostic Procedures</th>
<th>Techniques</th>
<th>Share %</th>
</tr>
</thead>
</table>
| Vasculature | Angiography, arteriography (arteries), venography (veins), ventriculography and interventional angiography | (a) intravenous or intra-arterial administration of soluble contrast media during imaging.  
(b) patient is catheterized by administering CA near the region of interest.  
(c) interventional procedures include vessel remodeling procedure such as angioplasty, atherectomy during angiographic visualization  
(d) angiographic procedure | 17%     |
<p>| Organs      | Brain CT, abdominal CT, liver CT, hepatosphenography, pyelography, cholecystography     | General i.v. administration of contrast agent prior to image acquisition, often before and after the CA administration                                                                                     | 54%     |</p>
<table>
<thead>
<tr>
<th>Region</th>
<th>Procedure</th>
<th>Method</th>
<th>Percentage</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spinal canal</td>
<td>Myelography (spine) and cisternography (brain)</td>
<td>Direct injection of CA into spinal canal or subarachnoid space</td>
<td>2%</td>
</tr>
<tr>
<td>Urinary tract and bladder</td>
<td>Retrograde pylegraphy and urethrography</td>
<td>Contrast administration through cathether placed in bladder</td>
<td>3%</td>
</tr>
<tr>
<td>Joints</td>
<td>Arthrography and discography</td>
<td>CA is administered directly into joints</td>
<td>2%</td>
</tr>
<tr>
<td>Uterine cavity and fallopian tubes</td>
<td>Hysterosalpingography</td>
<td>Contrast agents administered to uterus and/or to fallopian tubes</td>
<td>2%</td>
</tr>
<tr>
<td>Rest</td>
<td></td>
<td></td>
<td>20%</td>
</tr>
</tbody>
</table>

To better delineate soft tissues regions, such as cardiovascular system and cerebrospinal systems, safe and efficient X-ray contrast agents (also called radiographic contrast agents, radiopaque agents or roentgneographic agents) were long sought after. Contrast agents are a class of pharmaceuticals that, when administered to a patient, enter and pass through anatomic regions of interest to provide transient contrast enhancement. These contrast enhancing agents are then completely excreted.
renally from patients without being metabolized.

1.3.3 Infrared Spectral Studies

Infrared studies of organic compounds used as ligands in preparing complexes have been made to identify various modes of vibrations and hence their structures. This techniques had provided a powerful and useful unambiguous method of structural determination for complexes of transition metals. Based on these studies many conclusions of the general nature and feasible with respect of ligand chain strength, presence of absence of certain functional groups, multiple bonding, hydrogen bonding isomerism, bound state of ligands, degree of symmetry, but lately it has been also possible to decide alternative stereo-chemistries [167-169].

In general the vibrations originating in the ligand appear in the high frequency region, and those originating in the coordinate bonds appear in the far infrared region. The analysis of low frequency spectra provide direct information about the coordination bonds, where as high frequency spectra reveal the secondary effect of coordination on the ligands. These secondary effects on coordination on ligand vibrations is the key to elicit whether coordination has taken place or not and if so then at what points.
The most conclusive proof of coordination taking place is the appearance of several new modes of vibrations eg. \( v(M-N) \); \( v(M-S) \); \( v(M-O) \) or metal halogen in the far I.R. region. The frequencies observed in infrared spectra are the functions of the mass as well as the force constant and on this account the band shift are the function of metals. As a rule the direction of the shift depends upon relative electro-negativities of ligand and X-halogen atoms in the ligand LX\(_n\).

The data on metal-halogen frequencies can be used to ascertain the dependence of \( v(M-X) \) vibration on the oxidation number, mass and coordination number and on the stereochemistries of complexes. Making use of symmetry arguments, relationship between \( v(M-X) \) vibrations and stereo chemistry has been established. Such studies are of particular importance where the metal atom has a closed shell of valence electrons and hence the electronic absorption spectra and magnetism yield no conclusive information regarding the stereochemistry.

The ligand dithio-oxamide show a medium intensity band at 3225 cm\(^{-1}\); the position of which shift to lower frequency. This may be taken as an evidence for the coordination of secondary nitrogen
to vanadium. The ligand also show four thioamide bands at 1550-1520 cm\(^{-1}\), 1280-1260 cm\(^{-1}\), 1100-1080 cm\(^{-1}\) and 780-760 cm\(^{-1}\). These bands have contributions from \(\delta(C-H)\), + \(\delta(N-H)\), \(\nu(C=S) + \nu(C-N) + \delta(C-H)\), \(\nu(C-N) + \nu(C-S)\) and \(\nu(C=S)\) modes of vibrations, respectively. The lowering of first three bands in the complexes further supports the coordination through \(-NH\) group. The ligands show a band at 1640 cm\(^{-1}\) characteristic of \(\nu(C=N)\). This band is found to occur at lower frequency (15-20 cm\(^{-1}\)) in the complexes. The lowering of the frequency of this band is an indication of the coordination of unsaturated nitrogen of the azomethine linkage. In other complex however a strong bond at 1090 cm\(^{-1}\) are due to ionic perchlorate[170] overlaps the \(\nu(C-N)\) vibrations. The perchlorate vibration between 1095-1090 cm\(^{-1}\) and 620-625 cm\(^{-1}\) and due to \(\nu_3\) and \(\nu_4\) modes respectively. A weak band \(\nu_1\) is observed at 940 cm\(^{-1}\) in the complexes and it is due to crystal field effect. The absence of splitting of these bands indicate tetrahedral symmetry of the perchlorate and hence its ionic nature. The coordinated water absorbs between 3400-3500 cm\(^{-1}\) \(\nu(O-H)\) in transition and inner transition metal complexes. Bending vibration of \(H_2O\) overlaps with the carbonyl frequency. The \(H_2O\) rocking frequencies occur between 8325-8454 cm\(^{-1}\) indicating that water is
coordinated to the metal ion[171]. The non ligand v (M-O) and v(M-N) vibrations are observed at 450-400 cm\(^{-1}\) and 330-300 cm\(^{-1}\) respectively.

The I.R. spectrum of the phenolic ligand shows band at 3458 and 1358 cm\(^{-1}\) which are assigned to the stretching and in plane bending modes of the OH- group. These modes absorb in 2262-3477 and 1360-1400 cm\(^{-1}\) regions respectively in the complexes indicating that the OH group of the ligand is not involved in bond formation. The spectra of stretching all vanadyl sulphate complexes show new bands around 970 cm\(^{-1}\) due to v(V=O) vibration. The presence of an ionic sulphate group in the complexes has been confirmed by the appearance of three bands at 1140 (v\(_3\)), 960 (v\(_1\)) and 660 cm\(^{-1}\) (v\(_4\)). The absence of v\(_2\)- band and non splitting of the v\(_3\) band indicate that Td symmetry still holds. Thus the infrared spectral data show that the ligands act as N\(_4\)-chelating agents, bonding through all four nitrogen atoms to the oxovandadium ion [172-174].

1.3.4 \(^1\text{H NMR}\)

NMR is one of the most powerful tools for the study of the environment about paramagnetic metal ions, such as Fe\(^{2+}\), CO\(^{2+}\),
Ni\(^{2+}\) and some Ln\(^{3+}\) ions, in metal complexes and metallo proteins via the assignment of the isotropically shifted \(^1\)HNMR signal. Paramagnetic Ln\(^{3+}\) complexes have been used as sift reagents for structural determinations of diamagnetic compound.

Further, NMR has been utilized for the structural and mechanistic studies of several Ca\(^{2+}\) binding proteins such as parvalbumin, calmodulin and \(\alpha\)-lactabumin by using paramagnetic Ln\(^{3+}\) as probes [179]. The isotropic shift in paramagnetic Ln\(^{3+}\) complexes is mainly attributable to the distance and geometry dependent diploar shift mechanism [176].

The proton NMR spectra of the ligand and lanthanum (III) complex were recorded in DMSO-d6. The characteristic signal appearing at 12.96 ppm in the free ligand and at 12.71 ppm in the complex can be assigned to the phenolic protein [176]. Peak position of the OH proton in the free ligand and in the lanthanum (III) complex indicated that the OH group coordinated to the metal ion without deprotonation. This observation furthermore confirmed. The peak position of the OH proton in the free ligand as well as in the complexes indicated that the interaction between the phenolic oxygen atom and the lanthanide (III) ion, does not
increase the acidity sufficiently for ionization of the proton [180].

The $^1$HNMR spectra of [La(HSAT)$_2$(NO$_3$)$_3$] and [Lu(HSAT)$_2$(NO$_3$)$_3$] recorded in DMSO-d$_6$ also exhibited signals for the OH proton at 12.64 ppm, indication that the OH group the coordinated to the metal ion without deprotonation. The signal for the $-$CH=N- proton are resonating at 7.98 ppm in metal complexes. The signals due to other protons are found in the expected region and resonating by about 0.10 - 0.20 in the spectra of the metal complexes. [181].

1.3.5 UV-Visible Spectrophotometry:-

The UV–Vis absorption spectra were recorded with a Perkin–Elmer UV–Vis spectrophotometer Lambda 6 Model 2688–002, using 10 - 4 M ethanolic solutions of the complexes. The bands observed in the absorption spectra of the complexes are ascribed to ligand-centered transitions. UV absorption spectra of the fluorinated complexes were also obtained in the solid state from thin films of thickness ranging from 400 to 800 nm. The solid state photophysical properties of the complexes (viz. Eu$^3+$ quantum yields, luminescence and excited state lifetimes at 300, 77, and in some cases, 4.2 K) were investigated for all the
The excitation of a molecule from its electronic ground state to an electronic excited state corresponds to absorption of light in the near-infrared, visible or ultraviolet regions of the spectrum. For lanthanide complexes, the absorption bands in the first two or these regions (infrared and visible) are relatively weak and are associated with transitions largely localized on the lanthanide atom. The ultraviolet bands are intense and they are associated with the transfer of an electron from one atom to another and so are called charge-transfer bands.

The spectra lanthanide complexes depend on the transition of unpaired electrons from the ground state to an excited state. Transitions may occur between the split levels of the central atom, giving rise to f-f ligand field spectra. The spectra region where these bands occur spans the near infra-red, visible and UV. Most of the lanthanide complexes are colored due to f-f transitions in the visible region. The atomic overlap in lanthanide ligand bonds allows f.

The spectral data for the solution of some representative lanthanide (iii) i.e. 4F. metal complexes investigated in CH₃CN are
recorded. Lanthanide (iii) has no significant absorption in the visible region. The absorption band of Pr$^{3+}$, Nd$^{3+}$, Gd$^{3+}$, Tb$^{3+}$ and Dy$^{3+}$ in the visible and near infrared region appear due to transition from the ground levels $^3\!H_4$, $^4\!H_{9/2}$, $^6\!H_{5/2}$, $^8\!S_{7/2}$, and $^6\!H_{15/2}$, to the excited J-levels of 4f, configuration, respectively some red shift is observed in CH$_3$CN solution[184].

The luminescence spectra of europium and gadolinium complexes were obtained by scanning a double-grating Jobin-Yvon U-1000 monochromator. The excitation wavelengths were selected by a 0.25 m Jobin-Yvon H-10 monochromator, using a 150 W Xe–Hg lamp as the excitation source. The light detection was performed by a water-cooled RCA C31034 photomultiplier tube, the photocurrent signal being acquired through a EG&G discriminator model 1182 and digitally stored by a Jobin-Yvon Spectralink interface and a personal computer. This set-up allows for measurements at room temperature (298K) and 77 K. The excitation and luminescence spectra of some complexes were also obtained by using a SPEX Fluorolog DM3000F Spectrofluorometer with double-grating 0.22 m SPEX 1680 monochromators, and a 450 W Xe Lamp as the excitation source. This set-up is equipped
with an Oxford LF205 liquid Helium flow cryostat, allowing for measurements down to 4.2 K. The spectra are corrected for the instrumental response. Excited state decay time and rise time measurements were performed at 298 K using a pulsed N2 laser as the excitation source. The luminescence was detected with a modified 1P28 photomultiplier tube, after dispersion through a 0.25 m monochromator. The signal was then analyzed on a fast oscilloscope. The temporal resolution of the overall system is ca. 50 ns. The emission spectra and decay time measurements for the Gd 3+ complexes allowed the identification of the lowest ligand triplet state in the complexes. As a representative example, the emission spectrum of Gd(btfa)3phenNO at 77 K is The luminescence spectra of the europium complexes upon ligand excitation consist of Eu 3+ emission lines only. Apart from intensity differences, the spectra are essentially identical at low temperatures. As an example, Figs. 2 and 3 show the luminescence spectra of Eu(btfa)3phenNO at 300 K and at 4.2 K, respectively. The temperature dependence of the emission intensities can be quite large for some complexes. This will be discussed below, in conjunction with decay times and quantum yields for a few selected complexes. The excitation spectra of the
5D0 emission of the Eu3+ ion in the complexes indicate an efficient ligand-to-metal energy transfer, since the most intense feature in the spectrum is a broad band corresponding to transitions populating ligand-centered excited states (Due to the lanthanides’ electronic structure, their complexes have unique optical properties, including luminescence lifetimes that range from micro- to milliseconds, and sharp emission bands whose width at half-height (fwhm) rarely exceed 10 nm. This is much narrower than the typically broad fluorescence arising from organic molecule or Cd/Se nanoparticles. As a result of these unique properties, well-designed lanthanide complexes can be used as luminescent probes to analyze biological problems, where a targeted analyte may be present in a matrix that can also contain a large number of other fluorescent molecules (i.e., high background autofluorescence). Similarly, discrimination of the relevant fluorescent signal from that present in biological media (e.g., fluorescence from amino acids such as tyrosine or tryptophan) is readily achieved by time gating (a time-resolved measurement). Indeed, the combination of temporal and spectral discrimination properties have led to the development of time-resolved fluoroimmunoassays with very high sensitivities, without the need
for tedious prior purification of the sample which is useful when a very large number of samples must be screened. Because the lanthanide f-f transitions are Laporte forbidden, the molar absorptivity is very low. The typical strategy to circumvent this is to increase the luminescence excitation by coordinating the luminescent lanthanide ion to a chromophore, which acts as an antenna to effectively transfer light energy to the metal. For example, in biological systems, amino acids such as tryptophan have been utilized for this purpose, although in that case the binding environment is not optimized for the lanthanide cation. In order to form luminescent lanthanide complexes with CPL activity, chiral complexes have been developed where the coordination geometry around the lanthanide is well-defined and controllable, which enables finetuning of the photophysical properties.

Despite these attractive features, there is a limited number of lanthanide based luminescent probes suitable for practical CPL applications in solution. The reason is the weak CPL signal intensity for the complexes described in the literature, due to a combination of limitations imposed by key requirements which must be fulfilled in order to obtain efficient luminescent complexes
with detectable CPL signals in solution. First, the limited absorption of lanthanide cations must be overcome by coordinating a ligand that is able to harvest significant amounts of UV or visible light and then efficiently transfer this electronic energy to the lanthanide metal ion. Second, since the excited states of the lanthanide cations can be easily deactivated through nonradiative interaction with their chemical environment (coupling to vibrational modes of solvent molecules or of the ligand), the lanthanide must be protected from this quenching environment when bound to the ligand. This implies that the ligand must fulfill the high coordination number requirement of the lanthanide cation by providing an adequate number of donor atoms (typically between 8 and 12 in solution). Third, the ensuing complex must have sufficient thermodynamic stability and/or kinetic inertness at the appropriate concentration to interact with, and report on, the macromolecule of interest, since dissociation would lead to loss of the antenna effect and a subsequent decrease in luminescence. Fourth, to observe an unambiguous CPL signal it is preferable that only a single enantiomer of the chiral complex be present in solution or, more specifically, the existence of only one of these enantiomers must match the time scale of the experiment (microseconds to
milliseconds depending on the nature of the lanthanide cation). Hence, any type of exchange between the complex and the environment in the luminescence time scale should be avoided. Ideally, a rigid, nonfluxional complex, with a saturated coordination sphere, should best satisfy this latter condition. and reported the properties of a new family of lanthanide complexes formed with ligands incorporating 2-hydroxyisophthalamide as the chelating unit. Upon coordination, these complexes demonstrate several superior luminescence properties, including highly efficient transfer of electronic excitation from the ligand to metal center and sufficient protection of the lanthanide metal ion against nonradiative deactivation from the environment (e.g., solvent molecules). Indeed, the quantum yield of the Tb(III) complex in H$_2$O, 61%, is the highest value yet reported for a luminescent metal complex with high stability in aqueous solution. In addition, these newly developed ligands allow the sensitization of four different trivalent lanthanides emitting in the visible (Sm, Eu, Tb, and Dy) with the same ligand. Herein, we have extended the superior luminescence properties of these complexes to CPL activity by the addition of a chiral substituent to the open face of an octadentate, tetrapodal ligand framework, utilizing the 2-
hydroxyisophthalic acid chelating unit as the sensitizer. This strategy results in the formation of enantiomERICALLY pure complexes with strong circular dichroism (CD) activity. Significantly, these complexes are also strongly luminescent, emitting in the visible region, and display strong CPL activity. As a result of their brightness and improved signal-to-noise ratio, these complexes present new possibilities for increased sensitivity of assays based on CPL measurements. While the Tb(III) complex reported here is the most fluorescent, the strongest CPL effect has been observed for the Eu(III) complex, with one of the highest ΔI values reported to date. In addition, the Dy(III) and Sm(III) complexes are also luminescent and have significant CPL activity, a unique feature which enables us to have four different luminescent probes that can be easily discriminated from each other. Since the sharp emission bands do not appreciably overlap, multiplex detection is possible[227].