1.1. Introduction

Bioinorganic chemistry is a field that examines the role of metals in biology. It includes the study of both natural phenomena such as the behaviour of metalloproteins as well artificially introduced metals, including those that are non-essential, in medicine and toxicology. Many biological processes including respiration, much of metabolism, nitrogen fixation, photosynthesis, development, nerve transmission, muscle contraction, signal transduction and protection against toxic, mutagenic agents etc., fall into the realm of bioinorganic chemistry. Also, important in elucidating the implications of electron-transfer proteins, substrate bindings and activation, atom and group transfer chemistry as well as metal properties in biological chemistry. The discipline includes the study of inorganic models or mimics that imitate the behavior of metalloproteins [1].

The two major components of bioinorganic chemistry are: i) the study of naturally occurring inorganic elements in bio-systems and ii) introduction of these

Synthesis, spectral characterization and biological evaluation of new binuclear metal(II) complexes of ONNO chelating tetradentate Schiff base derived from 3, 3'-dihydroxybenzidine
elements as probes or drugs into biological systems and studying inorganic models that mimic the behaviour of various metallo-proteins. It also investigates the nutritional aspects, toxicity, therapeutic action, transport and storage of metals and non metals in plants and animals including microorganisms.

**Inorganic Elements and Their Biological Functions**

The inorganic elements, especially metal ion play an important role in biology which has lead to the development of huge number of metal complexes with diverse therapeutic activity. Based on the relative concentrations in the biological systems, the metals are divided into:

Bulk metals - C, H, N, O

Macronutrients (relatively large amounts) - Na, K, Mg, Ca, S, P, Cl & Fe

Micronutrients (trace elements) - Mn, Co, Ni, Cu, Zn, V, Cr, Mo, Se, F, I, Al, Pb, Sn, Si etc., which are present in low concentration and are used for biocatalysts.

The advances in the field of Inorganic chemistry provide better opportunities to use metal complexes as potential drugs. Cisplatin, carboplatin and oxaliplatin are the well known metal-based drugs widely used in treatment of cancer. Besides these complexes various metal complexes have shown promising results in the treatment of diseases like diabetes, ulcer, rheumatoid arthritis, inflammatory and cardiovascular diseases etc. This hypothesis includes the application of some potential metal complexes in the treatment of various diseases/ disorders to improve the therapeutic efficacy of the pharmaceuticals [2]
1.1.2. Schiff Base Transition Metal Complexes

Schiff base ligands have been in chemistry catalogue for over 150 years. Schiff bases are generally regarded as excellent ligands. Their instant and enduring popularity undeniably stem from the ease of their synthesis, bewildering versatility and wide ranging complexing ability once formed [3]. Hugo Schiff, a German chemist developed a new class of organic compounds in 1864 [4]. They are prepared by condensing a carbonyl compound with an amine, generally in refluxing alcohol. The dynamic and well designed Schiff base ligands are considered as “privileged ligands”. In actual fact, Schiff bases are able to stabilize many different metals in various oxidation states, controlling the performance of metals in a large variety of useful catalytic transformations. The common structural feature of these compounds is the azomethine group with a general formula RHC=N-R', where R and R' are alkyl, aryl, cyclo alkyl or heterocyclic groups which may be variously substituted. Several studies showed that the presence of a lone pair of electrons in a sp² hybridised orbital of nitrogen atom of the azomethine group is of substantial chemical and biological importance. Examples of a few Schiff bases are given in Fig 1.1.1. Schiff bases have been often used as chelating ligands in coordination chemistry [5], in catalysis, anti-oxidative activity, anti-bacterial activity and medicine as antibiotics, anti-inflammatory agents, in industry for anti-corrosion properties [6]. On the industrial scale, they have wide range of applications such as dyes and pigments [7]. The Schiff base metal complexes are also very useful model systems. Similarly, Schiff base have extended their applications in numerous fields.
Fig 1.1.1. Diverse examples of Schiff bases

Transition metal complexes have vast application in various fields it is because it has many good characteristics and excellent biological activity in medicinal fields especially. Transition metal complexes of Schiff base compounds are used as growth inhibiting agents for most of bacteria and fungi also they are widely used as potential therapeutics they are useful in health and skin care. The complexes of this type are also used as catalysts in organic redox and electrochemical reduction reactions for various chemical reactions. Transition metal complexes possess many
advantages such as facile approach for synthesis, relative tolerance, readily adjusted ancillary ligands, and tunable steric and electronic coordination environments on the metal centre.

1.1.3. ONNO Chelating Schiff Base

Schiff bases can be considered as useful chelating agents due to their ability to encapsulate into their coordinating moiety metal ions when a suitable function group, e.g. OH etc., is present sufficiently close to the azomethine group [8].

The architectural beauty of these coordination complexes arises due to the interesting ligand systems containing different donor sites in heterocyclic rings, e.g., N and O in Fig 1.1.2. A great deal of attention in this area has been focused on the complexes formed by transition metal ions with Schiff bases because of the presence of both nitrogen and oxygen donor atoms in the backbones of these ligands. For several reasons, Schiff bases have been found to be the most convenient and attractive ligands for forming complexes. One of which is, the two donor atoms,
N and O, of the chelated Schiff base exert two opposite electronic effects; the phenolate oxygen is a hard donor and stabilizes the higher oxidation state of the metal atom; whereas the imine nitrogen is a borderline donor and stabilizes the lower oxidation state of the metal ion.

![Chemical structures](image)

Fig 1.1.2. N$_2$O$_2$ Schiff bases

Schiff base ligands can also accommodate different metal centers involving various coordination modes thereby allowing successful synthesis of homo and hetero metallic complexes with varied stereochemistry. Tetradeinate Schiff base ligands have wide applications in biological field, as antidepressants, antimicrobial, antitumor, antiphlogogistic, nematocide, and other medicinal agents have been
reported based on these compounds. Furthermore Schiff bases have good catalytic role in many reactions [9].

1.1.4. Binucleating Schiff Base Ligands

The term “dinucleating ligands” was first introduced in 1970 by Robson [10] to describe the class of polydentate chelating ligands which able to bind simultaneously two metal ions. The study of binuclear transition metal complexes forms an extremely active area of research [11].

Fig 1.1.3. Examples of Binucleating Ligands

In general, the preparation of binuclear complexes can be approached from two different directions. Firstly, binuclear complexes may be formed through the aggregation of two metal centres with bridging and capping ligands via “self-assembly”. By employing multidentate capping ligands and taking into account the geometric preferences of the metal ions, binuclear arrays may be designed and synthesised in which one, two or three group’s bridge the metal ions. A second route involves the use of preformed ligands (“binucleating ligands”) that
are capable of simultaneously binding to two metal centres. This synthetic strategy has an obvious advantage over the self-assembly route in that it offers greater control over the products which form. By using ligands with predetermined binding characteristics, complexes may be generated with prescribed coordination geometries, metal–metal separations and donor atom types [12].

Binuclear Metal Complexes

A bimetallic core is versatile at the active site of many metalloenzymes and plays an essential role in biological systems by the interplay of a pair of metal ions [13]. In the last decade, a large number of bimetallic Schiff base complexes of different structural types have been researched [14]. These complexes span the gamut in their new applications, donating types, structures and biological activities [15]. The binuclear complexes have greater cleaving efficiency than mononuclear complexes. The Schiff base complexes are able to inhibit the growth of several animal tumors, and some metals have shown good antitumor activity against animal tumors [16]. A rational control of the nuclearity of transition metal complexes is important to design systems with the desired properties, as some of these applications require the presence of more than one metal center in the particular complex. Indeed, binuclear complexes may have very different reactivity than mononuclear counterparts, thereby enabling transformations inaccessible to single metal ions [17]. Furthermore, bi- or oligonuclear complexes containing transition metals may be formed by bridging ligands that can mediate magnetic interactions between paramagnetic metal ions. Binuclear complexes of Schiff-base ligands have been proven to be useful for the design of molecular ferromagnets, catalysts for many organic reactions, models for the active sites in metalloenzymes, optical
and luminescent materials, or DNA cleavage reagents [18]. Also, metal complexes of these bases have numerous applications including antibacterial, antifungal and antiviral activities as well as other biological applications. Several applications have been related for these complexes in chemical analysis, absorption and transport of oxygen, in pesticides and heterogeneous and homogeneous catalysis for oxidation and polymerization of organic compounds [19].

1.1.5. Role of Metal Complexes in Chemotherapy

Transition metals have a vital place within medicinal chemistry. Research has shown significant progress in utilization of transition metal complexes as in the treatment of cancer, as antibactericide agents, as fungicide agents, as antivirus agents, as antimalarial agent, as antidiabetic, neurological disorder and slow release long acting drugs in nutrition and in the study of metabolism and for other biological properties [20]. Transition metals exhibit different oxidation states and can interact with a number of negatively charged molecules. This activity of transition metals

Synthesis, spectral characterization and biological evaluation of new binuclear metal(II) complexes of ONNO chelating tetridente Schiiff base derived from 3, 3′-dihydroxybenzidine
has started the development of metal based drugs with promising pharmacological applications. Versatility of Schiff base ligands and the biological applications of their complexes make further investigations in this area highly desirable.

Although various Schiff base metal complexes exhibiting promising biological activity we are especially interested in the coordination chemistry of electron rich first row transition metals such as Copper, Nickel and Vanadium, as our work is focused on complexes of these metal ions. The significance of these Schiff-Base Complexes is as follows:

1.1.6. Why Copper Complexes?

Copper is a late first row transition metal with an electronic configuration of [Ar]3d^{10}4s^{1}. Copper coordination complexes generally have the copper ion in the +1 or, more commonly, in +2 oxidation state, which is the most stable.

Copper(III) are also known but are rare in comparison. Copper is one of those metals that help the world go around. Without copper, so many of the
conveniences we are used to would probably be impossible. Importance of copper [21] complexes is depicted in the figure.

1.1.7. Why Nickel Complexes?

Nickel has a function that is related to changes caused by deprivation of folic acid, pyridoxine or vitamin B12. Essential role for nickel in protein synthesis in animals. Functioning of various plant enzymes such as urease and hydrogenase. Nickel-depletion only low levels of nitrogen fixation occurred, which resulted in slow plant growth.
Nickel is the 24th most abundant element in the Earth’s crust, comprising about 3% of the composition of the earth. Nickel is known to be an essential micronutrient in animals and plants and is therefore also likely to be essential for humans. The importance of nickel has been well documented in many cases [22]. These nickel(II) complexes function by exploiting the coordination chemistry of the nickel(II) ion and its preference for adopting a square planar geometry.

1.1.8. Why Vanadyl Complexes?

Vanadium is the 21st abundant element in the earth’s crust, the average concentration being at 35 ppm, and is contained at 2 ppm in sea water, was discovered by Sefstrom in 1831, owing its name to Vanadis – Norse goddess of beauty and fertility. Vanadium chemistry has attracted attention due to its interesting structural features and biological relevance [23].
Pharmacological uses of vanadium include lowering of cholesterol, triglycerides and glucose levels, diuretic and natriuretic effects, anti-carcinogenic effect, contraction of blood vessels, enhancement of oxygen-affinity of haemoglobin and myoglobin. These complexes have multiple biological and pharmacological activities, including antimicrobial, anti-leukemia, antitumor, photodynamic therapy and as an insulin mimetic. Applications of vanadyl complexes in medicine have focused in the treatment of many diseases.

1.1.9. DNA Cleavage

Deoxyribonucleic acid (DNA) is the primary target molecule for most anticancer and antiviral therapies according to cell biologists. Investigations on the interaction of DNA with small molecules are important in the design of new types of pharmaceutical molecules.

Since the chemical nuclease activity of transition metal complexes was discovered in the 1980s, there has been a great interest in studying the interaction model and the mechanism of transition metal complexes with DNA. There are metal complexes which interact with DNA and induce the breakage of DNA strands by appropriate methods. The structure of DNA is shown in Fig 1.1.4. The importance of certain compounds in medical diagnosis and genomic research is based on the ability of such compounds to bind and cleave double stranded DNA under physiological conditions [24]. The hydrolytic and oxidative cleavage pathways are involved in DNA cleavage reactions. The formation of fragments may be considered to take place through enzymatic processes which occurs due to hydrolysis of phosphodiester. The nucleobase oxidation and/ or degradation of sugar by abstraction of sugar hydrogen atom(s) take place during oxidative process.
The binding ability of DNA is the main source for making comparison in cleavage efficiency of the complexes to that of the control. The open circular DNA is obtained from supercoiled DNA (Fig 1.1.5) by complexes.

The account of DNA cleavage by hydroxyl radicals via abstraction of a hydrogen atom from sugar units and proposed general mechanisms that predicts the
release of specific residues which arise from transformation of sugars, which also
depends on the position of hydrogen atom removal [25]. The hydroxyl radical
mediated cleavage reactions and cleavage of peroxo derivatives is inhibited by free
radical scavengers. The hydroxyl radical or a peroxo moiety generated from the
co-reactant \( \text{H}_2\text{O}_2 \) is bound by the metallo complexes which modulate the cleavage
reactions as well.

The outstanding criteria for the development of metallodrugs as
chemotherapeutic agents are the ability of the metallodrug to provoke DNA
cleavage. A large number of transition metal complexes because of their redox
properties, have been found to enhance DNA cleavage. A huge number of transition
metal complexes have been shown to promote DNA cleavage in the presence of
co-reagents [26]. The cleavage efficacy of metal complexes depends on the nature of
the metal ions and the ligands.

1.1.10. DNA-Binding Studies

A variety of small molecules, such as drugs, dyes, metals and some other
components, can bind to nucleic acids. This binding will affect the normal function
of nucleic acids, and different type of binding will result in different type of
processes. Since DNA is a target of anticancer treatment as well as other diseases, it
is very important to characterize the binding affinity and to understand the
mechanism of DNA-drug interactions.

The Importance of DNA-Binding Study

The binding between DNA and metal complexes has attracted interest over
the past ten years. DNA intercalating agents disrupt the normal function of cellular
DNA and can lead to interference with gene expression, gene transcription,
mutagenesis, carcinogenesis, and cell death. Hence, a precise understanding of the DNA-binding properties of metal complexes is very important in medicinal and pharmaceutical fields. Most of the studies are directed toward the design of site and conformation specific reagents to provide routes toward rational drug design as well as a means to develop sensitive chemical probes of DNA [27].

Binding Modes

The transition metal complex can interact non-covalently with DNA by intercalation, groove-face binding or external electrostatic binding. Different types of binding will bring different effects. For example, it may induce intercalation between stacked base pairs, distorting the DNA backbone conformation and therefore, affect the DNA-metal complex interaction [28]. The interaction can be studied by using various spectrometric techniques, such as NMR, ESR, CV, Circular dichroism (CD), Fluorescence, Resonance Raman, UV-visible and Fourier transform infrared spectroscopy, as well as electrochemical method. Among these spectra, the results of UV-visible, Cyclic voltammetry and electrochemical method of small molecules bound to nucleic acid provide conveniently a signature for the binding mode to DNA [29].

A. Electrostatic Binding

![Fig 1.1.6. Electrostatic binding mode](image-url)
Complexes are positively charged and the DNA phosphate sugar backbone is negatively charged and their interaction is known as electrostatic [30].

B. Groove Binding

In the Groove-binding mode, molecules sit in either the minor or major groove with close contact to both the functional groups of the sugar-phosphate backbone lining the groove as well as the functional groups from the nucleobases exposed at its bottom. High binding constants can be achieved with linear, extended molecules which wrap around the double helix in one of the grooves and are able to make multiple attractive interactions [31].

Fig 1.1.7. Groove-binding mode

Hydrogen bonding to the nucleobase donor/ acceptor groups exposed at the bottom of the groove leads to especially strong binding and is also the basis of a base sequence-specific recognition which can be achieved with specially-designed groove binders. This is largely the realm of modular extended organic molecules.

C. Intercalative Binding

Intercalative binding is one of the ways of molecules to interact with DNA. Intercalation occurs when ligands of an appropriate size and chemical nature fit themselves in between base pairs of DNA. In this case, the planar part of the molecule inserts between two Watson–Crick base pairs with the rest of the
compound positioned in either the major or minor groove. The backbone of the DNA is expanded to accommodate the intercalator, which acts like an additional base pair inserted into the DNA sequence. Stacking interactions between the π-systems of the intercalator and the nucleobases give rise to strong binding, which can further be increased by additional weak interactions between the rest of the molecule positioned in the groove and the DNA backbone lining it. These interactions can also enable recognition of certain base sequences, but since the number of characteristic contacts is usually smaller than with extended groove binders, the specificity is lower and recognition sequences are much shorter [32].

![Intercalative binding](image.png)

Fig 1.1.8. Intercalative binding mode

1.1.11. Antibacterial Activity

Microbial infections are a growing problem in contemporary medicine, and the use of antibiotics is inevitable. The global sales of antibiotics are generally higher when compared to other drugs which are prescribed. Antibiotic resistance is a major problem in hospitals as well as in community settings. A continuous increase in the number of infections caused by bacterial resistance to one or multiple class of antibiotics possess a significant threat as it may lead to treatment failures and associated complications. Thus the treatment of bacterial infections remains a challenging therapeutic problem. Despite the many antibiotics and chemotherapeutics
available, the emergence of old and new antibiotic resistant bacterial strains in the last decades constitutes a substantial need for new classes of antibacterial agents.

An antibacterial is a substance may either kill microorganisms outright or simply prevent their growth. Danish Physician Christian Grams, discovered the differential staining technique known as Gram staining, which differentiates the bacteria into two groups “Gram positive” and “Gram negative”, Gram positive bacteria retain the crystal violet and resist decolorization with acetone or alcohol and hence appear deep violet in colour; while Gram negative bacteria, which loose the crystal violet, are counter-stained by safranin and hence appear red in colour.

Gram positive and Gram negative bacteria have similar internal, but very different external structures. A Gram positive bacterium has a thick, multilayered cell wall (Fig 1.1.9) consisting mainly of peptidoglycan surrounding the cytoplasmic membrane. Gram negative cell walls are more complex, contains two layers external to the cytoplasmic membrane.

![Gram Positive and Gram Negative Bacteria](image)

Fig 1.1.9. Cell wall structure of Gram positive and Gram negative bacteria
Antimicrobial Drugs

Antimicrobial drugs either kill microbes or prevent the growth of microbes. Generally transition metal complexes have higher antimicrobial activity. Such increased activity of the metal chelates can be explained on the basis of chelation theory [33]. The transition metal complexes also disturb the respiration process of the cell and thus block the synthesis of proteins, which restricts further growth of the organism.

In general, when the antimicrobial activity of metal complexes is concerned, the following five principal factors may be considered:

The chelating effect
The nature of the ligands
The total charge of the complex; generally the antimicrobial efficiency decreases in the order cationic > neutral > anionic complex
The nature of the counter ion in the case of the ionic complexes
The nuclearity of the metal center in the complex; dinuclear centers are more active than mononuclear ones

Mode of Action of Antibacterial Drugs

The mode of action for the antimicrobials involves various targets in the organisms but the exact biochemical mechanisms are not known.

Following are Included in the Targets:

Antimicrobial drugs may inhibit

Cell-wall synthesis
Protein synthesis

Nucleicacidsynthesis

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Enzymatic activity
Folate metabolism or
Damage cytoplasmic membrane

Fig 1.1.10. Mechanisms of action of antibacterial drugs

Effect of azomethine (-C=N-) group

The mechanism of working of such compounds may be on the basis of hydrogen bond formation by the azomethine group (-C=N-) at the active centers of cellular entities, which cause the interferences in normal cellular phenomenon.

Four different microbial species were used to screen the possible antimicrobial activity of the synthesized metal complexes. They are as follows,
Gram Positive Bacteria

Staphylococcus Aureus

Staphylococcus was first identified in 1880 in Aberdeen. The word staphylococcus is derived from the Greek language (Gr. Staphylo = bunch of grapes; Gr. Coccus = a grain or berry), while the species name is derived from Latin language (L. aureus = golden).

![Microbial structure of Staphylococcus aureus](image_url)

Fig 1.1.11. Microbial structure of Staphylococcus aureus

S. aureus has long been recognized as one of the most important bacteria that cause disease in humans. It is the leading cause of skin and soft tissue infections such as abscesses (boils), furuncles, and cellulitis. Although most staph infections are not serious, S. aureus can cause serious infections such as bloodstream infections, pneumonia, or bone and joint infections [34]. Microbial structure of Staphylococcus Aureus is shown in Fig 1.1.11.

Bacillus Subtilis

Bacillus subtilis named by Ferdinand Cohn in 1872. Bacillus subtilis (Fig 1.1.12). It is a ubiquitous bacterium commonly recovered from water, soil, air, and decomposing plant residue. B. subtilis produces a variety of proteases and other
enzymes that enable it to degrade a variety of natural substrates and contribute to nutrient cycling. B. subtilis is only known to cause disease in severely immunocompromised patients, and can conversely be used as a probiotic in healthy individuals [35]. It rarely causes food poisoning. The potential risk associated with the use of this bacterium in fermentation facilities is low.

Fig 1.1.12. Microbial structure of Bacillus subtilis

Gram Negative Bacteria

Escherichia Coli

In 1885, a German pediatrician, Theodor Escherich, discovered this organism in the feces of healthy individuals and called it Bacterium coli commune.

Fig 1.1.13. Microbial structure of Escherichia coli
Escherichia coli (Fig 1.1.13) is a Gram-negative, facultative anaerobic, rod-shaped bacterium that is commonly found in the lower intestine of warm-blooded organisms. E. coli infection by coming into contact with the feces, or stool, of humans or animals. This can happen when you drink water or eat food that has been contaminated by feces. These problems can cause kidney failure and sometimes long-term disability or death in some children and older adults [36].

Klebsiella Pneumoniae

Klebsiella was named after the German bacteriologist Edwin Klebs (1834–1913). Klebsiella pneumoniae is a Gram-negative, non motile, encapsulated, lactose fermenting, facultative anaerobic, rod shaped bacterium. Although found in the normal flora of the mouth, skin, and intestines [37] it can cause destructive changes to human lungs if aspirated. They cause destructive changes to human lungs inflammation and hemorrhage with cell death (necrosis) that sometimes produces a thick, bloody, mucoid sputum (currant jelly sputum). Typically these bacteria gain access after a person aspirates colonizing oropharyngeal microbes into the lower respiratory tract.

Fig 1.1.14. Microbial structure of Klebsiella pneumoniae
1.1.12. Antioxidant Assay

Free radicals contain one or more unpaired electrons, produced in normal or pathological cell metabolism. Reactive oxygen species (ROS) react easily with these free radicals to become radicals themselves. ROS are various forms of activated oxygen, which include free radicals such as superoxide anion radicals (O$_2^-$) and hydroxyl radicals (OH$^-$), as well as non-free radical species (H$_2$O$_2$) and the singlet oxygen (O$_2$) [38]. They are formed in living organisms in different ways, including normal aerobic respiration, peroxisomes etc., they are natural by-products of our body’s metabolism.

Fig 1.1.15. Formation and prevention of Free radical

They are dangerous; however, when present in excess, they can attack biological molecules such as lipids, proteins, enzymes, DNA and RNA, leading to cell or tissue injury. The cells experience an oxidative stress which contributes in a various clinical disorders such as cancer, heart diseases, neurogenerative diseases like multiple sclerosis, Parkinson’s disease, autoimmune disease, stroke, arthritis, ischemia, reperfusion injury, acute hypertension, haem-orrhagic shock, emphysema, cirrhosis, diabetes mellitus, hepatitis, cancer, atherosclerosis as well as other ailments [39]. The formation and prevention of free radical is as shown in Fig 1.1.15.
DPPH Radical Scavenging Activity

Fig 1.1.16. DPPH free-radical mechanism

DPPH (2, 2-diphenyl-1-picrylhydrazyl) is a dark-colored crystalline powder composed of stable free-radical molecules. DPPH is a well-known radical and a trap ("scavenger") for other radicals. Therefore, rate reduction of a chemical reaction upon addition of DPPH is used as an indicator of the radical nature of that reaction. Because of a strong absorption band centered at about 517 nm, the DPPH radical has a deep violet color in solution, and it becomes colorless or pale yellow when neutralized. This property allows visual monitoring of the reaction, and the number of initial radicals can be counted from the change in the optical absorption at 517 nm [40]. DPPH free radical mechanism is shown in Fig 1.1.16.

Reducing power assay

Reducing power is associated with antioxidant activity and may serve as a significant reflection of the antioxidant activity. Reducing power assay method is based on the principle that substances, which have reduction potential, react with...
potassium ferricyanide (Fe$^{3+}$) to form potassium ferrocyanide (Fe$^{2+}$), which then reacts with ferric chloride to form ferric ferrous complex. In this assay, the yellow colour of the test solution changes to various shades of green and blue depending on the reducing power of each compound. By measuring the formation of Pearl’s Prussian blue at 700 nm [41], it is possible to determine the concentration of Fe$^{3+}$ ion.

\[
\begin{align*}
\text{Antioxidant} \\
\text{Potassium ferricyanide + Ferric chloride} & \rightarrow \\
\text{Potassium ferrocyanide + Ferric chloride}
\end{align*}
\]

Scientists in many different disciplines become more interested in the thrust of new compounds, either synthesized or obtained from natural sources that could provide active components to prevent or reduce the impact of oxidative stress on cell [42]. Moreover, recent reports have also suggested that many transition metal complexes have exhibited interesting antioxidant activity.
References


1.2. Literature Review

Introduction

The chelating Schiff base ligands derived from 3, 3’-dihydroxybenzidine, Glyoxal/ Diacetyl/ Benzil/ 2, 3- pentanedione/ o- phthalaldehyde and 2-aminophenol encompass a highly remarkable class of compounds. In the past two decades, the properties of Schiff base metal complexes stimulated much interest for their noteworthy contributions to our society in many ways. These complexes find a wide range of applications in catalytic, synthetic, analytical, clinical and biochemical areas and in addition they possess considerable physiological activities. Various reports about the Schiff base complexes of first transition series metals like, vanadium, chromium, manganese, iron, cobalt, nickel, copper and zinc are available in the literature. Few of the examples are as follows:

Previous Report on Binuclear Schiff Base Metal Complexes

Mohamed et al\textsuperscript{1} have synthesized a new tetradeinate imine ligands are derived from Schiff base condensation in a 1:2 molar ratio of the 1, 2, 4, 5-tetra-amino benzene with 2-hydroxy benzaldehyde, (L\textsuperscript{1}), 2, 4-dihydroxy benzaldehyde (L\textsuperscript{2}) and 2-hydroxy naphthaldehyde (L\textsuperscript{3}). These ligands react with CoCl\textsubscript{2} and CuCl\textsubscript{2} in refluxing ethanol to yield a series of cobalt(II) and copper(II) complexes of the type [M\textsubscript{2} \textsuperscript{II}L\textsuperscript{n}] nH\textsubscript{2}O. The structure of the obtained ligands and their metal(II)
complexes were characterized by various physicochemical techniques. The probable mechanistic implications of the catalytic oxidation reactions are discussed.

Bhattacharjee et al\textsuperscript{2} have reported that a novel photoluminescent salicylaldimine ligands condensed from 3', 3', 4', 4' -tetraminobiphenyl and 4- substituted long alkoxy salicylaldehyde possessing two sets of tetradeutate [N\textsubscript{2}O\textsubscript{2}] donor site and their binuclear zinc(II) complexes have been synthesized.

The mesogenic, photophysical properties and DFT calculations of the synthesized complexes were investigated.
Rajavel et al\textsuperscript{3} have reported synthesis and characterization of Cu(II), Ni(II) and VO(II) complexes of Schiff base ligands prepared by condensation of 2-aminobenzaldehyde with aromatic diamine such as, 1,4- diaminobenzene, 4,4′- diaminophenyl, bis(4 aminophenyl)ether, 4,4′- diaminodiphenylmethane and N,N′-bis-(4-aminophenyl)thiourea. The ligands and its complexes were characterized by elemental analysis, molecular weight determination, molar conductance, IR, UV-Visible, magnetic moment measurement, TGA/DTA, electrochemical and EPR studies.

\[
\begin{array}{c}
\text{\includegraphics[width=0.5\textwidth]{image}}
\end{array}
\]

Uma et al\textsuperscript{4} have prepared a novel complexes was carried out by the reaction of di(2-pyridyl)amine ,4, 4′-dibromobiphenyl and Cu(CIO\textsubscript{4})\textsubscript{2}.6H\textsubscript{2}O. The EPR spectrum of the complex is consistent with coordination of each copper ion to two nitrogens of the binuclear ligand. Cyclic voltammogram of the complex also reveals that the two copper(II) centres have identical ligating environment. DNA cleavage studies carried out with the complex demonstrate that the complex is very efficient in promoting the cleavage of plasmid DNA under hydrolytic conditions, in the absence of any added cofactors. The maximum rate of degradation of supercoiled form to nicked form was found to be \(1.8 \times 10^{-3} \text{ s}^{-1}\).
Kasumov et al\textsuperscript{5} have synthesized that a new salen type ligands, N, N'-bis (X-3-tert-butylsalicylidene)-4,4'-ethylenedianiline [(X = H (1), 5-tertbutyl (2)] and N,N'-bis(X-3-tert-butylsalicylidene)-4,4'-amidedianiline [X = H (3), 5-tert (4)] and their copper(II) complexes. Their spectroscopic properties, as well as magnetic and redox-reactivity behavior are reported. In IR spectra, comparison of the free ligands IR spectra with those for their complexes suggests pentacoordinate geometry (trigonal bipyramidal or square pyramidal) with CuN\textsubscript{2}O\textsubscript{3} coordination chromophore.

The electrochemical behaviour of copper complexes exhibited the presence of non-interacting copper(II) centers where the redox processes occur. Thus, according to electrochemical and chemical study results, metal centered oxidation can be proposed for these complexes. On the basis of IR, UV/vis and solid state ESR spectroscopic results for amide linkage complexes a square based pyramidal geometry is proposed.

Venkatachalam et al\textsuperscript{6} have synthesized binuclear ruthenium(III) Schiff base complexes bearing bis-naphophen units. The binucleating Schiff base ligands containing two N\textsubscript{2}O\textsubscript{2} type tetradentate compartments. Ruthenium complexes used
for oxidation of organic substrates generally show mechanisms that involve a higher oxidation state of ruthenium. The formation of high-valent Ru$^V$ =O species as a catalytic active intermediate is proposed for the catalytic processes. The spectral data results suggest that the presence of an octahedral geometry around ruthenium metal in binuclear metal complexes.

Achar et al.$^7$ have synthesized that hexasulphonated binuclear complexes of copper, nickel and cobalt phthalocyanines. The remarkable increase in the electrical conductivities $10^6$-$10^7$ times after iodine doping has been identified.
Patel et al. have reported that a novel complex which is derived from anisidine, 2, 3-butanedione, piperazine ring of ciprofloxacin with Co(NO₃)₂.6H₂O. Synthesized compounds were found to be more potent compare to drugs, ligands and metal salt against selective gram(+) and gram(-) organisms.

Interaction of the complexes with nucleic acid (DNA) were investigated using spectroscopic technique, viscosity measurement and gel electrophoresis and it was found that the complexes bind to DNA via intercalative mode. Supercoiled pBR322 DNA (0.12 μg) in TE buffer was treated with different Co(II) complexes(10 μM); and efficacy of DNA cleavage was investigated by gel electrophoresis method.

Motswainyana et al. have prepared two new phenylene bridged binuclear bis (imino-quinolyl) palladium(II) complexes were prepared from the reaction of the ligand 1,4-bis (imino-quinolyl) benzene, L₁ with either Pd (cod) Cl₂ or Pd (cod)
ClMe. The molecular structure of L1 was confirmed by single crystal X-ray diffraction analysis. The complexes were evaluated for the first time as catalysts for Heck reaction of iodobenzene with methyl and butyl acrylate under mild reaction conditions and low catalyst loading. The preliminary results showed that the complexes were highly active, with good selectivity towards formation of the trans-isomers.

Yang Sun et al.\textsuperscript{10} have reported novel chiral binuclear Mn(III)-Schiff-base complexes was carried out by the asymmetric epoxidation of trans-stilbene with pyridine N-oxide in 2 mL CH\textsubscript{2}Cl\textsubscript{2} followed by Mn(III) salts and the application of these complexes in the asymmetric epoxidation of trans-stilbene is described, catalytic mechanism is also discussed briefly. The catalytic results indicate the utilization of these binuclear Mn(III)-complex catalysts result in high to moderate yields.

Xiaohong Chang et al.\textsuperscript{11} have worked on reactions of \(\mu_2\)-Chloro bridged cyclometallated Pd(II) complexes. All these complexes were fully characterized by FT-IR, NMR spectroscopy, elemental analysis and/or X-ray crystallography. All calculations were carried out with the SHELX-97 programs. All structures of Suitable crystals for X-ray analysis were solved by direct methods. All
non-hydrogen atoms were refined with anisotropic thermal parameters by using full-matrix least-squares methods

Takeshi Fujinami et al\textsuperscript{12} have designed two binuclear iron(III) complexes, \([L^1\text{Fe}^{III}(bpy)\text{Fe}^{II}L^1](\text{BPh}_4)_2\) and \([L^2\text{Fe}^{III}(bpy)\text{Fe}^{III}L^2](\text{BPh}_4)_2\), were synthesized and characterized. The different SCO behaviours of compounds were ascribed to one and two crystallographically unique Fe sites and the order/disorder at the saturated six-membered chelate rings of aminopropyl moieties. These data demonstrate that more functional SCO compounds could be achieved, if we could use disorder efficiently. The magnetic susceptibility measurements of two complexes showed one-step and two-step SCO behaviors, respectively. Complexes had a similar 4, 4'-bipyridine bridged binuclear structure.

Bulent Dede et al\textsuperscript{13} have designed a new tetradentate diimine–dioxime ligand containing a donor set of N\textsubscript{4}, and its homo, heterodinuclear and homotrinuclear copper(II) complexes were prepared and characterized on the basis of their elemental analysis, FT-IR, \(^1\text{H}\) and \(^{13}\text{C}\) NMR spectra, molar conductivity and magnetic moment measurements. In addition they have tested the catalytic activity of the complexes toward the disproportionation of hydrogen peroxide.
The catalytic results indicated that the complexes have good catalase activity and may be suitable and functional as a model for the pseudocatalase enzyme. The DNA cleavage results showed that the homo- and heterodinuclear copper complexes can effectively cleave supercoiled DNA to form nicked or linear DNA by performing single strand and double strand scissions under aerobic conditions. The complexes cleaved the supercoiled pBR322 DNA into much smaller fragments in the presence of hydrogen peroxide as a co-oxidant.

Raisanen et al\textsuperscript{14} have reported that a novel, bridged bis(salen-type) ligand precursors, 1,1,3,3-tetrakis(salicylidene-3-iminopropyl)butylenediamine and 1,1,3,3-tetra(salicylideneiminomethyl)propane, were prepared by Schiff base condensation of salicylaldehyde with appropriate tetraamines. The complexes were characterized in detail. Magnetic measurements showed that the complex is high-spin complexes at 300 K with $\mu_{\text{eff}}$ values of 6.35 B.M., respectively.
The values suggest square-pyramidal geometry around the Co(II) centers respectively, which are in accordance with the modelled structures.

Mohammad Shakir et al\textsuperscript{15} have synthesized a novel Schiff base ligand, N, N′-bis-(2 thiophenecarboxaldimine)-3, 3′-diaminobenzidine (L) obtained from condensation of 2-thiophenecarboxaldehyde and 3, 3′-diaminobenzidine. All the complexes were behaved as 1:2 electrolytic nature. Absorption and fluorescence spectroscopic studies were studied to find the DNA binding ability of the complexes. Comparative DNA binding studies of L with its complexes revealed that the complexes exhibited higher binding affinity toward DNA as compared to ligand. The Cu(II) complex showed the highest affinity.
Kandaswamy et al\textsuperscript{16} have synthesized a new class of binuclear copper(II), nickel(II) and zinc(II) complexes are synthesized and characterized. The redox, phosphate hydrolysis, DNA binding and cleavage properties of complexes were studied. The complexes show good binding propensity to calf thymus DNA. The binding site size and viscosity data suggest the DNA groove binding of the complexes. The complexes display significant hydrolytic cleavage of supercoiled pBR322 DNA at pH 7.2 and 37 °C.

Magdy Shebl\textsuperscript{17} has designed a tetradeutate N\textsubscript{2}O\textsubscript{2} donor Schiff base ligand, H\textsubscript{2}L, was synthesized by the condensation of 4, 6-diacetylresorcinol with benzylamine. The structure of the ligand was elucidated by elemental analyses, IR, \textsuperscript{1}H-NMR, electronic and mass spectra. Reaction of the Schiff base ligand with vanadyl(IV) ion in 1:2 molar ratio afforded binuclear metal complexes. The spectroscopic data showed that, the Schiff base ligand acts in all metal complexes as
neutral or dianionic tetradsentate ligand through the two azomethine nitrogen atoms and the two oxygen atoms of the two phenolic groups.

Tahir Ali Khan et al\(^{18}\) have reported a new series of binuclear decaazamacrocyclic complexes: dichloro/ nitrate [1-phenyl bis(8,9-diphenyl-1,3,7,10,14 pentaazacyclo -pentadeca-7,9-diene) metal(II)], \([M_2LX_4]\) (\(M = \text{Mn}^{II}, \text{Co}^{II}, \text{Ni}^{II}, \text{Cu}^{II}\) and \(\text{Zn}^{II}\), \(X = \text{Cl}\) or \(\text{NO}_3\)).

Mosae Selvakumar et al\(^{19}\) have worked on imine based bis-bidentate ligands with copper(II) acetate in 2:2 equivalent of L: M ratio, resulted in a series of binuclear \([\text{Cu}_2(\text{m-xysal})_2]\) neutral complexes.
An appropriate catalytic study converting 4-nitrobenzaldehyde to corresponding nitroaldol was carried out.

Pati\textsuperscript{20} have investigated that the complexes of Co(II), Ni(II) and Cu(II) with N,N'-bis-[5-X-salicylidene]-4,4'-diaminodibenzyl, abbreviated as H\textsubscript{2}nXS\textsubscript{al}PDADB (X = -H, -CH\textsubscript{3}, -Br). Various ESR parameters for copper complexes have been calculated. The compounds have been screened for their biological activities.
Mahalakshmi et al\textsuperscript{21} have reported the binuclear Schiff base Cu(II), Ni(II) and VO(II) complexes derived from 3,3', 4,4'-tetraminobiphenyl and 2-aminobenzaldehyde. The ligand and complexes have been established by analytical, spectral and electrochemical data. The interaction studies of the complexes with CT-DNA were carried out cyclic voltammetry, viscosity and fluorescence spectroscopy. The free ligand and their complexes were screened for their antimicrobial activities against pathogenic bacteria. The metal complexes exhibit higher antibacterial activity than the free ligand.

Ashok Kumar Singh et al\textsuperscript{22} have worked on a template condensation of benzidine, formaldehyde ethylenediamine or 1, 3-diaminopropane, metal salt and 1-phenyl-1, 3-butanedione or 2, 3-butanedione in a 1:4:2:2 molar ratio results in the formation of two new series of binuclear pentaaza macrocyclic complexes.
Emara et al\textsuperscript{23} have investigated two series of new binuclear complexes
with Schiff base ligands, H\textsubscript{4}L\textsubscript{a} or H\textsubscript{2}L\textsubscript{b}, derived from the reaction of
4,6-diacetylresorcinol and ethylenediamine, in the molar ratio 1:1 and 1:2 have been
prepared, respectively.

\[
\begin{align*}
\text{Cu} - \text{O} - \text{O} - \text{Cu} \\
\text{H}_{2} \text{O}
\end{align*}
\]
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