Synopsis of the thesis

INVESTIGATIONS ON DNA BINDING AND CLEAVAGE ACTIVITY OF MONO & DINUCLEAR TRANSITION METAL COMPLEXES

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The design of small molecules that bind and react at specific sequences of DNA under physiological conditions via oxidative and hydrolytic mechanisms has been attracting great interest in the field of Bioinorganic Chemistry. One of the principal avenues of bioinorganic chemistry is the study of metal complexes which have the ability to cleave DNA under suitable conditions. In recent years a great deal of effort has been directed towards the study of the metal complexes as artificial nucleases.

Over the past decades transition metal complexes have attracted considerable interest due to their efficient interactions with DNA under various physiological conditions for different applications of metal complexes in nucleic acid chemistry. It has been demonstrated that inorganic complexes can be used in foot printing studies, as sequence specific DNA binding agents, as diagnostic agents in medicinal applications, and for genomic research. Among different modes of DNA cleavage, oxidative cleavage of DNA is of main interest due to the potential applications.

Biomimetic hydrolysis of nucleic acids is of increasing importance in biotechnology and medicine. The ability of mono and multinuclear metal complexes to cleave nucleic acids efficiently, in a non-degradative manner and with high levels of selectivity for a site, sequence or structure will offer many applications for the manipulation of genes, the design of structural probes and the development of novel therapeutics. Modeling the reactivity of such metallonuclease enzymes is therefore an area of intense research interest. In addition, the metal complex-mediated hydrolysis of phosphate esters is providing valuable information for modeling and elucidating the reactivity of metal containing nuclease molecule.

Schiff bases are considered as a very important class of organic compounds which have wide applications in many biological aspects. Transition metal complexes of Schiff
bases are one of the most adaptable and thoroughly studied systems. These complexes have clinical, analytical and industrial applications in addition to their important roles in catalysis and organic synthesis.

Transition metal complexes synthesized from unsymmetrical Schiff bases serve as models of relevance to bio-inorganic chemistry such as metalloproteins and metalloenzymes in which transition metal ions are bound to a macrocycle, such as a heme ring or to donor atoms of peptide chains, usually in a distorted environment. Unsymmetrical Schiff base complexes have shown a wide spectrum of applications such as biochemical, analytical, industrial and antimicrobial agents. This leads to the interest for preparing unsymmetrical Schiff base ligands and their metal complexes.

Transition metal complexes containing salen type of ligands derived from o-hydroxy aromatic carbonyl compound and various aliphatic/aromatic diamines have been the subject of a number of investigations. The so called salen type of ligands are very much like porphyrins, and unlike porphyrins these ligands can be easily prepared. Metal complexes of these ligands have been developed as catalysts for the epoxidation of olefins and as multidimensional magnetic compounds. In addition, these complexes have been designed as nucleic acid reagents to induce damages in DNA and RNA. DNA cleavage can be induced by salen-Cu (II) complexes. Here the focus is on those derived from polymethylene diamines with a regular progression of chain lengths.

Schiff bases derived from the mono-condensation of diamines with o-hydroxy aromatic carbonyl compounds are a group of mono-negative NNO donor ligands which readily react with transition metal ions. These ligands allow a good control over the stereochemistry of the metallic centers in homo and hetero polynuclear complexes. It is of interest to synthesize metal complexes with mono– condensed imino- Schiff base ligands.
The design of dinucleating ligands, with an additional donor atom that can bridge two metals in a more or less fixed geometry has rapidly developed in recent years. Part of the interest stems from the fact that the corresponding complexes are often studied as enzyme mimics. Hence the interest focused on studying the metal complexes of quinque dentate Schiff base ligands.

The thesis has been divided into nine chapters dealing with introduction & objectives, description of the experimental methods, results & discussion and conclusions/ summary.

Chapter I gives brief introduction to the present work. Objectives of the present work are also given in this chapter. In the light of the objectives following ligands have been selected for studies.

The ligands selected for study are as follows:

I. Tridentate ligands

i. 2-[1-(Methylamino-ethylimino)-methyl]-phenol (SAMEN)

\[
\begin{align*}
&\text{OH} \\
&\text{H} \\
&\text{NH} \text{CH}_3 \\
&\text{N} \\
&\text{H} \\
&\text{CH}_3
\end{align*}
\]

ii. 2-[1-(Methylamino-ethylimino)-ethyl]-phenol (HAPMEN)

\[
\begin{align*}
&\text{OH} \\
&\text{CH}_3 \\
&\text{N} \\
&\text{NH} \text{CH}_3 \\
&\text{H} \\
&\text{CH}_3
\end{align*}
\]
iii. 2-[1-(propylamino-ethylimino)-methyl]-phenol (SAPEN)

![Structure of SAPEN](image)

iv. 2-[1-(propylamino-ethylimino)-ethyl]-phenol (HAPPEN)

![Structure of HAPPEN](image)

II. Symmetrical tetradentate Ligands containing polymethylene backbone

v. 2,2'(1,8-diiminoctamethylene)methyl bis phenol (SALOCMN)

![Structure of SALOCMN](image)

vi. 2,2'(1,7-diiminoheptamethylene)methyl bis phenol (SALHPMN)

![Structure of SALHPMN](image)
vii. 2,2'(1,6-diiminohexamethylene)methyl bis phenol (SALHXMN)

viii. 2,2'(1,8-diiminooctamethylene)ethyl bis phenol (HAPOCMN)

ix. 2,2'(1,7-diiminoheptamethylene)ethyl bis phenol (HAPHPMN)

x. 2,2'(1,6-diiminohexamethylene)ethyl bis phenol (HAPHXMN)
III. Unsymmetrical tetradebate Schiff base ligands:

xi. 2\{(1-(4-hydroxy-2-iminopentenyl)ethyl)imino)methyl\}-phenol (AAenSA)

\[
\text{H}_3\text{C} \quad \text{H}_3\text{C} \\
\text{OH} \quad \text{OH} \\
\text{N} \quad \text{N} \\
\text{CH}_2\text{C} \quad \text{CH}_2\text{C} \\
\text{H} \quad \text{H} \\
\text{N} \quad \text{N} \\
(\text{C}_\text{H}_\text{2})_2 \quad (\text{C}_\text{H}_\text{2})_2 \\
\text{H}_3\text{C} \quad \text{H}_3\text{C}
\]

xii. 2\{(1-(4-hydroxy-2-iminopentenyl)ethyl)imino)ethyl\}-phenol (AAenHAP)

\[
\text{H}_3\text{C} \quad \text{H}_3\text{C} \\
\text{OH} \quad \text{OH} \\
\text{N} \quad \text{N} \\
\text{CH}_2\text{C} \quad \text{CH}_2\text{C} \\
\text{H} \quad \text{H} \\
\text{N} \quad \text{N} \\
(\text{C}_\text{H}_\text{2})_2 \quad (\text{CH}_3)_2 \\
\text{H}_3\text{C} \quad \text{H}_3\text{C}
\]

xiii. 2\{(1-(4-hydroxy-3-iminopentenyl)propylimino)methyl\}-phenol (AApnSA)

\[
\text{H}_3\text{C} \quad \text{H}_3\text{C} \\
\text{OH} \quad \text{OH} \\
\text{N} \quad \text{N} \\
\text{CH}_2\text{C} \quad \text{CH}_2\text{C} \\
\text{H} \quad \text{H} \\
\text{N} \quad \text{N} \\
(\text{CH}_2)_3 \quad (\text{CH}_2)_3 \\
\text{H}_3\text{C} \quad \text{H}_3\text{C}
\]

xiv. 2\{(1-(4-hydroxy-3-iminopentenyl)propylimino)ethyl\}-phenol (AApnHAP)

\[
\text{H}_3\text{C} \quad \text{H}_3\text{C} \\
\text{OH} \quad \text{OH} \\
\text{N} \quad \text{N} \\
\text{CH}_2\text{C} \quad \text{CH}_2\text{C} \\
\text{H} \quad \text{H} \\
\text{N} \quad \text{N} \\
(\text{CH}_2)_3 \quad (\text{CH}_3)_2 \\
\text{H}_3\text{C} \quad \text{H}_3\text{C}
\]
IV. Quinquedentate ligands

xv. {2-Hydroxy-(1,3-diiminopropyl)2,2'-methyl}bis phenol (HDMPH$_3$)

![Image of HDMPH$_3$ structure]

xvi. {2-Hydroxy-(1,3-diiminopropyl)2,2'-methyl}bis phenol (HDEPH$_3$)

![Image of HDEPH$_3$ structure]

Copper(II) complexes along with some nickel(II) complexes have been synthesized from above listed ligands and characterized based on physico-chemical, spectral and structural analyses.

Chapter II, which is further, divided into five sections deals with the review of literature. Section 2(i) gives a review on di and polynuclear metal complexes with monocondensed tridentate imino-Schiff base ligands. Section 2(ii) provides a review on mononuclear metal complexes of unsymmetrical tetradentate Schiff base ligands, whereas the section 2(iii) reviews the literature on metal complexes of with symmetrical tetradentate Schiff base ligands containing polymethylene diamines of varying chain length. Section 2(iv) gives brief review on metal complexes of quinquedentate Schiff bases and section.
Finally section 2(v) gives review on nuclease activity metal complexes of the ligands discussed in the previous sections.

**Chapter III** comprises of four sections in which the materials and methods used in the present study are described. In section 3(i), details of chemicals, reagents and solvents used in the present study are given. Syntheses of organic ligands and metal complexes are given in section 3(ii) and section 3(iii). Section 3(iv) provides a brief description of equipment used in the present study.

**Chapter IV** deals with the studies on mononuclear copper (II) complexes of monocondensed tridentate Schiff base ligands derived from salicylaldehyde / o-hydroxyacetophenone and N-methylendiamine/ N-propylethylenediamine (ligands i to iv in the above said list) and Cu(NO\(_3\))\(_2\). 3H\(_2\)O. These complexes were characterized by IR, ESR, magnetic moments and conductivity measurements.

Molecular structure of copper(II) complex with 2-[1-(methylamino-ethylimino)-ethyl]-phenol (HAPMEN) was determined by single crystal X-ray diffraction technique. Electrochemical behavior of these complexes was investigated by cyclic voltammetry.
Binding interactions of metal complexes with calf-thymus DNA are carried out using absorption spectrophotometry. Cleavage activities of these complexes are investigated on a double stranded pBR plasmid DNA by using gel electrophoresis experiments in the absence and in the presence of an oxidant, a complexing agent, a free radical scavenger and a reducing agent.

Ortep diagram of [(2-[1-(methylamino-ethylimino)-ethyl]-phenol)copper(II)] nitrate

Chapter V deals with the study of dinuclear copper(II) complexes derived from the same ligands used to prepared mononuclear complexes in Chapter IV using Cu (ClO$_4$)$_2$.6H$_2$O in stead of Cu(NO$_3$)$_2$. 3H$_2$O. These complexes were characterized by IR, ESR, magnetic moments and conductivity measurements. Molecular structure of dinuclear copper(II) complex with 2-[1-(propylamino-ethylimino)-methyl]-phenol (SAPEN) was determined by single crystal X-ray diffraction technique. Electrochemical behavior of these complexes has been investigated by cyclic voltammetric studies. Binding interactions of metal complexes
with calf-thymus DNA are carried out using absorption spectrophotometry. Cleavage activities of these complexes are investigated on a double stranded pBR plasmid DNA by using gel electrophoresis experiments in the absence and in the presence of an oxidant, a complexing agent, a free radical scavenger and a reducing agent.

Ortep view of $\mu$-oxo-$\{2-[1-(propylamino-ethylmino)-methyl]-phenol\}$dicopper(II)perchlorate
Chapter VI goes with the physico-chemical and spectral characterization of acetato bridged dinuclear copper(II) and nickel(II) complexes derived from symmetrical tetradeate ligands containing polymethylene groups (6, 7 and 8). Electrochemical behavior of these complexes has been investigated by cyclic voltammetry. Binding interactions of metal complexes with calf-thymus DNA are carried out using absorption spectrophotometry. Cleavage activities of these complexes are investigated on a double stranded pBR plasmid DNA by using gel electrophoresis experiments in the absence and in the presence of an oxidant, a complexing agent, a free radical scavenger and a reducing agent.

**Diagram**

<table>
<thead>
<tr>
<th>R</th>
<th>n</th>
<th>Formula</th>
</tr>
</thead>
<tbody>
<tr>
<td>H</td>
<td>8</td>
<td>SALOCMN</td>
</tr>
<tr>
<td>H</td>
<td>7</td>
<td>SALHFMN</td>
</tr>
<tr>
<td>H</td>
<td>6</td>
<td>SALHEXMN</td>
</tr>
<tr>
<td>CH₃</td>
<td>8</td>
<td>HAPOCMN</td>
</tr>
<tr>
<td>CH₃</td>
<td>7</td>
<td>HAPHEMN</td>
</tr>
<tr>
<td>CH₃</td>
<td>6</td>
<td>HAPHEXMN</td>
</tr>
</tbody>
</table>

Chapter VII describes the spectral characterization along with DNA binding and cleavage studies on Mononuclear copper(II) and nickel(II) complexes derived from unsymmetrical tetradeate ligands. Mononuclear copper(II) and nickel(II) complexes with these ligands have been synthesized characterized based on molar conductance, magnetic moment, mass, electronic, IR and ESR spectral data. Electrochemical behavior of these complexes is investigated by cyclic voltammetric studies. Binding interactions of metal complexes with calf-thymus DNA are carried out using absorption spectrophotometry. Cleavage activities of these complexes are investigated on a double stranded pBR plasmid DNA.
DNA by using gel electrophoresis experiments in the absence and in the presence of an oxidant, a complexing agent, a free radical scavenger and a reducing agent.

\[ R = H, \quad n = 2 \quad \text{AAenSA}; \quad R = H, \quad n = 3 \quad \text{AAPnSA} \]
\[ R = \text{CH}_3, \quad n = 2 \quad \text{AAenHAP}; \quad R = \text{CH}_3, \quad n = 3 \quad \text{AAPnHAP} \]

**Chapter VIII** describes the physic-chemical and spectral characterization of acetato bridged dinuclear copper(II) complexes of quinquedentate ligands derived from salicylaldehyde/ o-hydroxyacetophenone and 1,3-diaminopropan-2ol. Electrochemical behavior of these complexes has been investigated by cyclic voltammetry. Binding interactions of metal complexes with calf-thymus DNA are carried out using absorption spectrophotometry. Cleavage activities of these complexes are investigated on a double stranded pBR plasmid DNA by using gel electrophoresis experiments in the absence and in the presence of an oxidant, a complexing agent, a free radical scavenger and a reducing agent.
Where $R = \text{H} ; \{2\text{-Hydroxy-(1,3-diiminopropyl)2,2’-methyl}\} \text{bis phenol (HDMPH}_3\text{)}$

$R = \text{CH}_3; \{2\text{-Hydroxy-(1,3-diiminopropyl)2,2’-methyl}\} \text{bis phenol (HDEPH}_3\text{)}$

Finally, summary and conclusions of the present work based on the results incorporated in the previous chapter are presented in chapter IX.