A new series of transition metal complexes of Cu(II), Ni(II), Zn(II) and VO(IV), were synthesized from the Schiff base (L) derived from 4-aminoantipyrine, 3-hydroxy-4-nitrobenzaldehyde and acetylacetone. The structural features were arrived from their elemental analyses, magnetic susceptibility, molar conductance, Mass, IR, UV-Vis., ¹H NMR and ESR spectral studies. The data show that the complexes have composition of [ML]X type. The UV-Vis., magnetic susceptibility and ESR spectral data of the complexes suggest a square-planar geometry around the central metal ion except for VO(IV) complex which has square-pyramidal geometry. The redox behavior of copper and vanadyl complexes were studied by cyclic voltammetry. The antimicrobial screening tests were also recorded and gave good results in the presence of metal ion in the ligand system. The nuclease activity of the above metal complexes shows that the copper and nickel complexes cleave DNA through redox chemistry, whereas other complexes are not effective.

Keywords: Square-planar geometry, Calf Thymus DNA, Nuclease activity, Antimicrobial activity

INTRODUCTION

Compounds containing an azomethine group are known as imines (Schiff bases). The chelating abilities and analytical and biological applications of these compounds have attracted remarkable attention [1-3]. These compounds are readily hydrolyzed under acidic conditions leading to active aldehydes which can act as alkylating agents [4]. Besides, several azomethines have been reported to possess remarkable antibacterial [5-9], antifungal [10-12], anticancer [13-16] and diuretic activities [17]. Antibiotics such as Streptomycin, Aspergillus acid and Tetracycline are known to have chelating properties. Presumably, some antibiotics are delicately balanced so as to be able to compete successfully with the metal-binding agents of bacteria while not disturbing the metal processing by the host. There is evidence that at least some bacteria have developed resistance to antibiotics through the development of altered enzyme systems that can compete successfully with antibiotics. The action of the antibiotic need not be a simple competitive one. The chelating properties of antibiotics may be used in metal transport across membranes or used to attach the antibiotic to a specific site from which it can interfere with the growth of bacteria [18]. Pyrazolone and its derivatives are a group of antibiotics that have been extensively used in treating several bacterial diseases [19-21]. They have antibacterial activity against gram-negative and gram-positive strains. Transition metal complexes of these antibiotics with enhanced potentiality against bacterial strains have been reported elsewhere [22].

Moreover, mimicking the activities of nuclease is currently
an attractive research area in molecular biology since artificial nucleases have potential applications as novel restriction enzymes and anticancer therapeutic agents [23]. Among the different therapeutic strategies to eradicate cancer cells through DNA damage, the view of using small water soluble transition metal complexes, capable of oxidative or hydrolytic DNA cleavage as anticancer drugs is a challenging issue in bioinorganic chemistry [24,25]. Many transition metal complexes with vanadium [26], iron [27], copper [28,29], cobalt [30], lanthanides [31,32] and also actinides [33] have been reported as efficient DNA cleavage agents with or without sequence specificity, moreover the ligand or the metal in these complexes can be varied in an easily controlled manner to facilitate the individual applications [34].

In continuation of our series of investigations, we attempted to widen the scope of derivatization by providing more flexibility through Schiff base formation with 4-aminoantipyrine containing keto group, >C=N and complexation with metal ions. The Schiff base structure affords a greater choice and flexibility, and complexation with a metal ion adds to the stability and versatility of the compounds. The novel investigated compounds and their metal complexes were also evaluated for their antimicrobial activity against several bacterial strains and nuclease activity against calf thymus (CT) DNA.

EXPERIMENTAL

All reagents, 4-aminoantipyrine, 3-hydroxy-4-nitro-benzaldehyde and acetylacetone and various metal chlorides and vanadyl sulphate were Merck products, CT DNA from GENEI and used as supplied. For the voltammetric experiments, tetrabutylammonium perchlorate (TBAP) used as supporting electrolyte, was purchased from Sigma. Anhydrous grade ethanol, DMF and DMSO were purified according to standard procedures. Microanalytical data of the compounds were recorded at the Sophisticated Analytical Instrument Facility, Central Drug Research Institute (SAIF, CDRI), Lucknow. The mass spectra of the ligand and its complexes were recorded at the Sophisticated Analytical Instrument Facility (SAIF), Indian Institute of Technology, Mumbai. 1H NMR spectra (300 MHz) of the samples were recorded in CDCl₃ and DMSO-d₆ by employing TMS as internal standard at Madurai Kamaraj University, Madurai. The IR spectra of the samples were recorded on a Perkin-Elmer 783 spectrophotometer in 4000-400 cm⁻¹ range using KBr pellet. The UV-Vis. spectra were recorded on a Shimadzu UV-1601 spectrophotometer using DMF as solvent. The X-band ESR spectra of the copper and vanadyl complexes were recorded at 300 and 77 K on a Varian ESR spectrophotometer using diphenylpicrylhydrazyl (DPPH) as internal standard at RSIC, IIT, Chennai. Magnetic susceptibility measurements of the complexes were carried out by Gouy balance using copper sulphate as the calibrant. Electrochemical studies were carried out using EG&G Princeton Applied Research Potentiostat/Galvanostat Model 273A, controlled by M270 software. CV measurements were performed using a glassy carbon working electrode, platinum wire auxiliary electrode and an Ag/AgCl reference electrode. All solutions were purged with N₂ for 30 min prior to each set of experiments. The molar conductance of the complexes was measured using a Systronic conductivity bridge at room temperature in DMSO solution. Solutions of CT DNA (calf thymus DNA) in 50 mM NaCl/50 mM tris-HCl (pH = 7.2) gave a ratio of UV absorbance at 260 and 280 nm, A₂₆₀/A₂₈₀ of ca. 1.8-1.9, indicating that the DNA was sufficiently free of protein contamination [35]. The DNA concentration was determined by the UV absorbance at 260 nm after 1:100 dilutions. The molar absorption coefficient was taken as 6600 M⁻¹ cm⁻¹. Stock solutions were kept at 4 °C and used within 4 days. Doubly distilled H₂O was used to prepare the buffer. The antimicrobial activities of the ligand and its complexes were carried out by well diffusion method.

**Synthesis of Knoevenagel Condensate β-Diketone (I)**

Condensation of acetylacetone with 3-hydroxy-4-nitro-benzaldehyde was performed by heating equimolar amounts (10 mmol) under reflux in 50 ml ethanol, in the presence of 5 drops of piperidine as a catalyst for not less than 10 h. The solution was then cooled and the condensed solid product of Knoevenagel condensate (I) β-diketone was isolated by filtration, washed and recrystallised from ethanol.

**Synthesis of Schiff Base**

An ethanolic solution (50 ml) of 3(3'-hydroxy-4'-nitro-benzalidene)-2,4-pentanedione (I) (5 mmol) and 4-aminoantipyrine (10 mmol) was boiled under reflux for ca. 3 h. The