



Synthesis, Spectral, Electrochemical, Antimicrobial and DNA Binding/Cleavage Studies of Metal Complexes with Schiff Base of Fluoro Substituted Aniline

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Received: 27 June 2020;

Accepted: 22 August 2020;

Published online: 28 October 2020;

AJC-20116

A new series of Schiff base complexes of transition (Fe^{3+} , Co^{2+} , Ni^{2+} , Cu^{2+} and Zn^{2+}) metal were synthesized from 4-fluoroaniline and 2-thiophene carboxaldehyde and structurally characterized by spectroscopic techniques. The Schiff base is found to be a bidentate ligand and coordinates to the metal ion through azomethine nitrogen and sulphur atom of the thiophene ring. In fluorescence studies, an interaction of Ca^{2+} and Mg^{2+} ions with the ligand was also studied. The Schiff base and its transition metal complexes showed inhibition activity against Gram-positive bacteria (*Staphylococcus aureus*, *Escherichia coli*), Gram-negative bacteria (*Pseudomonas aeruginosa*) and antifungal activity against *Candida albicans*. Electrochemical redox reactions of the metal complexes were analyzed by cyclic voltammetry. The DNA binding properties of the complexes with HS-DNA have been explored by electron absorption spectroscopy. The cleavage reaction of the synthesized ligand and its metal complexes was monitored by gel-electrophoresis method. The nuclease activity of the above metal(II) complexes shows that the Cu(II) complex can cleave DNA effectively than ligand and other metal complexes.

Keywords: 2-Thiophene carboxaldehyde, 4-Fluoroaniline, Schiff base, DNA Binding studies, Antimicrobial activity.

INTRODUCTION

Schiff base complexes are considered to be the most important stereochemical models in transition metal coordination chemistry due to their preparative accessibility and structural variety. Moreover, these Schiff base metal complexes possess potential biological applications like antibacterial, antifungal, antioxidant and antitumor activities [1-3].

The Schiff base of 2-thiophene carboxaldehyde have evoked great interest due its applicability as potential ligand for a large number of metal ions and demonstrate biological applications [4]. Metalloorganic chemistry is one of the materializing areas of research due to the demand for new metal based antibacterial and antifungal compound. It is known that 4-fluoroaniline has been used as an intermediate in the manufacture of pharmaceuticals, herbicides and plant growth regulators. Moreover, the Schiff base of 4-fluoroaniline has been recognized as molecule with potential antimicrobial utility [5,6]. It is notable that the therapeutic inorganic science is a multidisciplinary field with

components of science, pharmacology, toxicology and biochemistry. Deoxyribonucleic acid (DNA) assumes an important role in the existence procedure, since it conveys the genetic information and leads to organic synthesis of proteins and compounds through the replication and translation of hereditary data in living cells. DNA is particularly a good target for metal complexes as it gives a wide variety of possible metal intercalation sites [7,8]. Thus, studying the interaction of molecules to DNA is possibly helpful in synthesizing potential anti-cancer medicines, which can recognize the specific location or conformation of DNA. Ability of transition metal complexes to cleave DNA under physiological conditions initiated us to plan for focusing the research on developing the more efficient metal-based antitumor drugs.

Reports have been documented for the interaction of coordination compounds with DNA. The study shows that these metal complexes can bind in a non-covalent style with DNA by intercalation, groove-face binding or outside electrostatic interaction. The nature and efficacy of stacking interactions

can be analyzed by noting the changes in the absorbances in the UV-visible spectrum. In present work, the first row *d*-block metal complexes of the Schiff base obtained from 2-thiophene carboxaldehyde and 4-fluoroaniline have been studied for binding ability with DNA through electronic spectroscopy and cleavage studies by gel-electrophoresis

Schiff bases play a significant role as sensors for tracking cellular processes in living organisms [9]. Fluorescent sensors based on Schiff bases were applied to monitor the changes in the concentrations of calcium(II) and magnesium(II) ions. In this study, we have attempted to determine the role of Schiff base as a sensor for Ca^{2+} and Mg^{2+} ions through electronic spectroscopy. In continuation to our interest in the design of bioactive heterocycles [10], we aimed to synthesize the *N*-[4-fluoroaniline]thiophene carboxaldimine Schiff base and its first row transition metal complexes. The synthesis of Schiff base ligand and its metal complexes were confirmed by analytical and spectroscopic (UV-visible, IR and NMR) techniques. Furthermore, antimicrobial, DNA binding and cleavage efficiency of synthesized compounds were investigated.

EXPERIMENTAL

All the chemicals used *viz.* 2-thiophene carboxaldehyde, 4-fluoroaniline, iron(III), cobalt(II), nickel(II) and copper(II) and zinc(II) chlorides were of AR grade. Commercial solvents were distilled and then used for the synthesis of the ligand and metal(II) complexes. Percentage of metal was determined using Atomic Absorption Spectrophotometer SYS-813. Molar conductivities in DMSO (10^{-3} mol/dm³) at room temperature were measured on an ELICO CM-180 digital conductivity meter. The IR spectra were recorded on Shimadzu FT-IR IR-Affinity-1 spectrometer in the 4000-400 cm⁻¹ range using KBr pellets. The NMR spectra were recorded on a Bruker DRX-300 MHz spectrometer in DMSO (*d*₆) with tetramethylsilane (TMS) as the internal reference. The absorption spectra were recorded using a Shimadzu UV-1800 spectrophotometer between 200-1100 nm by using suitable solvent. Different solvents such as acetonitrile, chloroform, dimethylformamide and ethanol were used to study the solvatochromatism of the ligand. Cyclic voltammetry measurements (CH instruments 608 C voltammograph, USA) were carried out in DMSO at room temperature using a three-electrode cell containing a reference Ag/AgCl electrode, Pt wire auxiliary electrode and glassy carbon working electrode with tetrabutylammonium perchlorate (TBAP) as supporting electrolyte. Herring Sperm Deoxyribonucleic acid (HS-DNA) was purchased from Sigma-Aldrich and used for DNA binding

and cleavage studies gel electrophoresis experiment was carried with 50 V and documented with UV lamp.

Synthesis of *N*-(4-fluoroaniline)thiophene carboxaldimine (L): A solution of 2-thiophene carboxaldehyde (5 mmol) in 20 mL ethanol was added to a solution of 4-fluoroaniline (5 mmol) in 20 mL ethanol and the mixture was stirred for 3-5 h. The resulting precipitate was then filtered, washed with ethanol and dried. The purity of prepared ligand was checked by TLC and recrystallized from the ethanol to obtain pale yellow crystalline solids. m.f. C₁₁H₈NSF (m.w.: 205.31); IR (KBr, ν_{max} , cm⁻¹): 1634 (CH=N), 786 (C-S), 1155 (C-F); ¹H NMR (δ ppm, DMSO-*d*₆): 8.5 (s, 1H, azomethine proton), 7.25-7.67 (4H, benzene ring protons), 6.63-7.01 (3H, thiophene ring protons); ¹³C NMR (75 MHz, DMSO-*d*₆, δ ppm): 142 (-N=CH-), 115-129.7 (-C₆H₄), 127-137 (-C₄H₃S).

Synthesis of metal complexes: All the metal complexes were synthesized from hydrated metal salts. To a Schiff base (6 mmol) dissolved in 20 mL of ethanol was added 3 mmol of MCl₂·*x*H₂O (M = Cu, Zn; *x* = 2), (M = Ni, Co; *x* = 6) and MCl₃ [M = Fe]. The solution was stirred for 3 h and allowed to settle. The precipitated complexes were filtered and washed with ethanol and dried. The purity of complexes was checked by TLC technique. The physical characterization of the ligand and its metal complexes are listed in Table-1.

[Fe(L)₂Cl₂]Cl: m.f. C₂₂H₁₆S₂N₂Cl₃F₂Fe, brown solid. IR (KBr, ν_{max} , cm⁻¹): 1592 (CH=N), 723 (C-S), 626 (M-N).

[Co(L)₂Cl₂]: m.f. C₂₂H₁₆S₂N₂Cl₂F₂Co, green solid. IR (KBr, ν_{max} , cm⁻¹): 1614 (CH=N), 768 (C-S), 612 (M-N).

[Ni(L)₂Cl₂]: m.f. C₂₂H₁₆S₂N₂Cl₂F₂Ni, light yellow solid, IR (KBr, ν_{max} , cm⁻¹): 1607 (CH=N), 764 (C-S), 620 (M-N).

[Cu(L)₂Cl₂]: m.f. C₂₂H₁₆S₂N₂Cl₂F₂Cu, dark brown solid, IR (KBr, ν_{max} , cm⁻¹): 1610 (CH=N), 774 (C-S), 634(M-N).

[Zn(L)₂Cl₂]: m.f. C₂₂H₁₆S₂N₂Cl₂F₂Zn, yellow solid. IR (KBr, ν_{max} , cm⁻¹): 1616 (CH=N), 738 (C-S), 628 (M-N).

UV-visible spectra : The Schiff base (L) to Ca^{2+} / Mg^{2+} ions fluorescence behaviour was investigated by the absorbance spectra. A solution of Schiff base (1×10^{-4} M) and Ca^{2+} / Mg^{2+} (1×10^{-3} M) ions were prepared. The Schiff base solution was added with metal ions and the complexation was recorded by UV-visible spectrophotometry.

DNA binding studies

Electronic absorption studies: The interaction of Fe(III), Co(II), Ni(II), Cu(II) and Zn(II) complexes with HS-DNA was examined using UV-visible spectroscopy. The electronic absorption spectrum of metal complexes (1.0×10^{-3} M) in presence of increasing amounts of HS-DNA in 5 mM Tris-HCl and

TABLE-1
PHYSICAL CHARACTERIZATION OF THE LIGAND AND ITS METAL COMPLEXES

Compound	Mol. mass	% of metal calculated	% of metal estimated from AAS	Λ_m (mho cm ² mol ⁻¹)	Melting point (°C)
C ₁₁ H ₈ NSF	205	–	–	–	192
C ₂₂ H ₁₆ S ₂ N ₂ F ₂ FeCl ₃	570	9.79	9.01	41	210
C ₂₂ H ₁₆ S ₂ N ₂ F ₂ CoCl ₂	538	10.95	9.90	03	202
C ₂₂ H ₁₆ S ₂ N ₂ F ₂ NiCl ₂	538	10.90	9.94	04	200
C ₂₂ H ₁₆ S ₂ N ₂ F ₂ CuCl ₂	543	11.70	10.95	02	220
C ₂₂ H ₁₆ S ₂ N ₂ F ₂ ZnCl ₂	545	11.99	11.05	08	270

50 mM NaCl buffer (pH 7.4) was measured. The concentration of metal(II) complexes was kept constant. The concentration of DNA solution was calculated from the molar extinction coefficient ($6600 \text{ M}^{-1} \text{ cm}^{-1}$) and the intensity at 260 nm. The concentration of DNA was found to be $3.09 \times 10^{-4} \text{ M}$. Stock solutions were kept at 4°C and used after not more than 4 days.

DNA cleavage studies by gel electrophoresis: Agarose gel electrophoresis technique was used to predict the cleavage of HS-DNA. The gel electrophoresis experiment was done by incubation of the samples containing $30 \mu\text{M L}^{-1}$ HS-DNA, $50 \mu\text{M L}^{-1}$ metal(II) complexes and $50 \mu\text{M L}^{-1} \text{H}_2\text{O}_2$ in tris-HCl/NaCl buffer (pH 7.2) at 37°C for 2 h. After incubation, the samples were electrophoresed for 2 h at 50 V on 1% agarose gel using tris acetic acid-EDTA buffer (pH 7.0). The gel was then stained using $1 \mu\text{g cm}^{-3}$ ethidium bromide and the image was captured under ultraviolet light at 360 nm.

Antimicrobial activity: The Schiff base (L) and its metal complexes were tested for their inhibitory effects on the growth of bacteria (*S. aureus*, *E. coli* and *P. aeruginosa*) and fungus (*C. albicans*) using the well diffusion method [11]. The stock solutions were prepared by dissolving the compounds in DMSO with the concentration $0.001 \mu\text{L}$. In a typical procedure, a well was made on the agar medium inoculated with microorganisms. The well was filled with test solutions using a micropipette and the plates were incubated at 37°C for 24 h. During this period, the test solution diffuses and affects the growth of the inoculated bacteria/fungus. The inhibition zone was developed and measured in mm.

RESULTS AND DISCUSSION

Synthesis of Schiff base ligand was done by the condensation reaction of 2-thiophene carboxaldehyde and 4-fluoroaniline. The obtained yellow coloured ligand was stable at room temperature. The Schiff base (L) forms stable complexes in ethanol medium. The metal complexes are partially soluble in ethanol, but soluble in DMF and DMSO solvents. The low values of conductance of Co(II), Ni(II), Cu(II) and Zn(II) in DMSO indicate that all the metal complexes except Fe(III) complex are non-electrolytes (Table-1). The molar conductance of Fe(III) complex is $41 \text{ mho cm}^2 \text{ mol}^{-1}$ indicating the 1:1 electrolytic nature [12]. In $^1\text{H NMR}$ of zinc(II) complex, the signal due to azomethine proton appears at 8.65 ppm and shows downfield shift compared to the free ligand, suggesting coordination of azomethine nitrogen. The azomethine nitrogen gets deshielded due to coordination to the metal ion. Atomic absorption spectroscopy results also confirmed the stoichiometric ratio of metal ion to ligand. The percentage on metal(II) ion in the complexes shows the stoichiometric ratio of M:L as 1:2. Based on spectral, conductivity and AAS data, the molecular formula $[\text{ML}_2\text{Cl}_2]$ is assigned for Co(II), Ni(II), Cu(II) and Zn(II) complexes and $[\text{ML}_2\text{Cl}]\text{Cl}$ is assigned for Fe(III) complex.

FT-IR studies: In IR spectrum of the free ligand, the bands at 1634 and 786 cm^{-1} are assigned to azomethine group $\nu(\text{CH}=\text{N})$ and $\nu_{\text{ring}}(\text{C}-\text{S})$ [13], respectively. In the spectra of the metal complexes, the frequencies at 1634 and 786 cm^{-1} are moved to lower values 1610 - 1592 and 774 - 723 cm^{-1} , respectively confir-

ming the coordination of the azomethine nitrogen and thiophene ring sulphur to the metal ion, respectively. Appearance of medium intensity band in the region 634 - 612 cm^{-1} indicates the $\nu(\text{M}-\text{N})$ vibration [14]. Thus the donor atoms of the ligand in the present complexation process are nitrogen and sulphur.

Electronic spectral studies: The electronic spectral data of the metal complexes along with the assignment of transition are shown in Table-2. The electronic spectrum of ligand shows the bands at 347 and 424 nm due to $\pi-\pi^*$ and $n-\pi^*$ transitions of intra ligand charge transitions (ILCT), respectively. The UV-visible spectrum of Fe(III) complex exhibits bands at 259 and 707 nm which was attributed to the $^1\text{A}_{1g} \rightarrow ^1\text{T}_{1g}$ and $^1\text{A}_{1g} \rightarrow ^1\text{T}_{2g}$ transitions and suggested an octahedral geometry around the central metal ion. The Co(II) complex shows bands at 614 and 676 nm which were assigned to the $^4\text{T}_{1g(\text{F})} \rightarrow ^4\text{A}_{2g(\text{F})}$ and $^2\text{T}_{2g(\text{F})} \rightarrow ^2\text{E}_{g(\text{F})}$ transitions, respectively. In the electronic spectrum of Ni(II) complex two bands at 343 and 772 nm which were dispensed to the $^3\text{A}_{2g} \rightarrow ^3\text{T}_{1g(\text{p})}$ and $^3\text{A}_{2g} \rightarrow ^3\text{T}_{1g}$ transitions. The spectrum of Cu(II) complex display bands at 349 and 549 nm which were attributed to the $^2\text{B}_{1g} \rightarrow ^2\text{B}_{2g}$ and $^2\text{B}_{1g} \rightarrow ^2\text{E}_g$ transitions. Zn(II) complex has d^{10} configuration and does not exhibit $d-d$ transitions. So the ILCT band of Zn(II) complex observed at 305 and 370 nm confirming the formation of Zn(II) complex. The absorptions in the zinc(II) complex are shifted to lower values compared to the ligand indicating the complexation process. The transitions are characteristic of octahedral coordination for Fe(III), Co(II), Ni(II), Cu(II) and Zn(II) complexes [15,16]. Fe(III) complex is a cationic one with electrolytic nature. Based on the spectral data, octahedral geometry may be proposed for the metal(II) complexes as depicted in Fig. 1.

TABLE-2
ELECTRONIC SPECTRAL DATA OF METAL COMPLEXES

Compounds	λ_{max} , cm^{-1} (nm)	Transition
Ligand (L)	23584 (424), 28818 (347)	—
$[\text{Fe}(\text{L})_2\text{Cl}_2]\text{Cl}$	14144 (707), 38610 (259)	$^1\text{A}_{1g} \rightarrow ^1\text{T}_{1g}$, $^1\text{A}_{1g} \rightarrow ^1\text{T}_{2g}$
$[\text{Co}(\text{L})_2\text{Cl}_2]$	14761 (676), 16286 (614)	$^4\text{T}_{1g(\text{F})} \rightarrow ^4\text{A}_{2g(\text{F})}$, $^2\text{T}_{2g(\text{F})} \rightarrow ^2\text{E}_{g(\text{F})}$
$[\text{Ni}(\text{L})_2\text{Cl}_2]$	12953 (772), 29154 (343)	$^3\text{A}_{2g} \rightarrow ^3\text{T}_{1g(\text{p})}$, $^3\text{A}_{2g} \rightarrow ^3\text{T}_{1g}$
$[\text{Cu}(\text{L})_2\text{Cl}_2]$	18214 (549), 28653 (349)	$^2\text{B}_{1g} \rightarrow ^2\text{B}_{2g}$, $^2\text{B}_{1g} \rightarrow ^2\text{E}_g$
$[\text{Zn}(\text{L})_2\text{Cl}_2]$	32786 (305), 27027(370)	ILCT

Electrochemical studies: Cyclic voltammetric studies of Fe(III), Co(II) and Cu(II) complexes were performed in DMSO solution. The copper(II) complex shows a quasi reversible peak in cathodic direction at $E_{\text{pc}} = 0.18 \text{ V}$, may be due to direct reduction $[\text{Cu}(\text{II})/\text{Cu}(\text{I})]$ that takes place in the complex. The cobalt(II) complex with scan rate 0.10 Vs^{-1} shows the formation of Co(II)/Co(III) couple at $E_{\text{pa}} = 0.410 \text{ V}$ and $E_{\text{pc}} = 0.190 \text{ V}$ (Table-3). The couple is quarsireversible with $\Delta E_{\text{p}} = 0.22 \text{ V}$ and the ratio of anodic peak to cathodic peaks currents ($I_{\text{pa}}/I_{\text{pc}} \approx 1$) corresponding to a simple one electron process [17,18]. Redox curves of Fe(III) complex with scan rate 0.1 V s^{-1} corresponds to the formation of Fe(III)/Fe(II) couple at $E_{\text{pa}} = 0.205 \text{ V}$ and $E_{\text{pc}} = 0.063 \text{ V}$. The cyclic voltammograms of Cu(II) and Co(II) complexes are shown in Fig. 2.

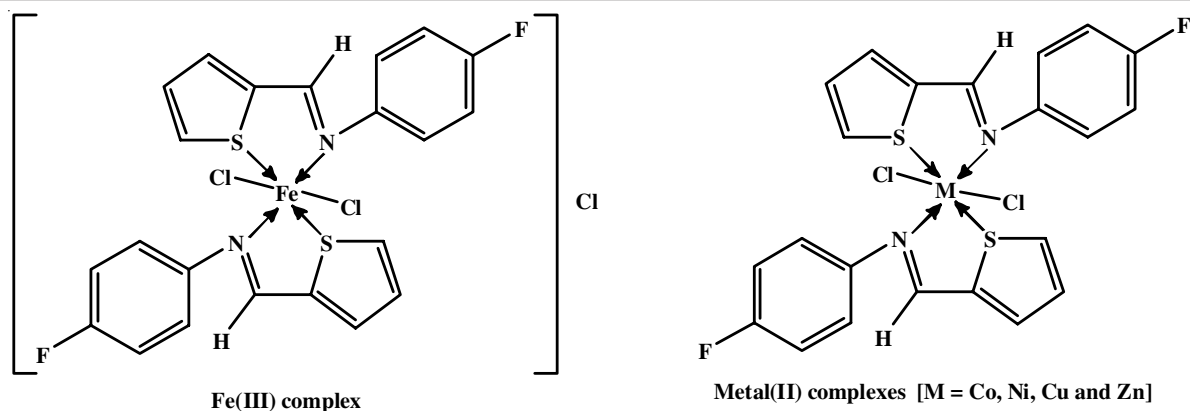
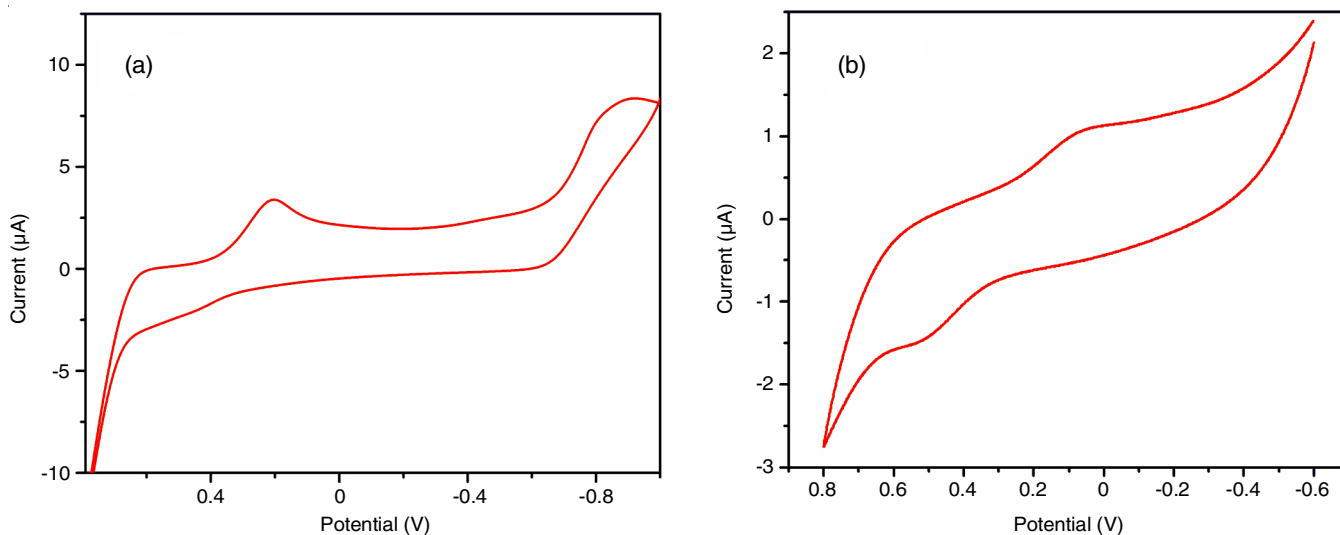


Fig. 1. Proposed structure of metal complexes

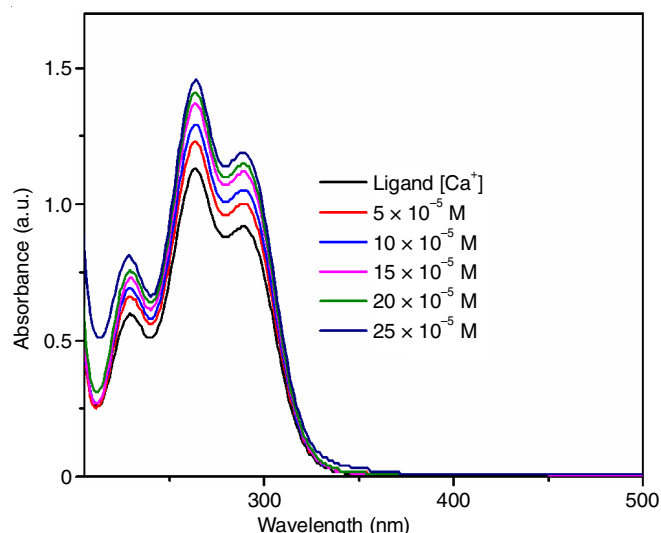
Fig. 2. Cyclic voltammogram of synthesized complexes in DMSO, (a) $[\text{Cu}(\text{L})_2\text{Cl}_2]$ and (b) $[\text{Co}(\text{L})_2\text{Cl}_2]$ TABLE-3
REDOX POTENTIAL OF THE COMPLEXES

Compounds	E_{pa} (V)	E_{pc} (V)
$[\text{Fe}(\text{L})_2\text{Cl}_2]$	0.205	0.063
$[\text{Co}(\text{L})_2\text{Cl}_2]$	0.410	0.220
$[\text{Cu}(\text{L})_2\text{Cl}_2]$	–	0.181

Solvent effect on the UV-visible spectrum of ligand:

Intensity and absorption maximum for the Schiff base in the electronic spectrum exhibited changes due to polar nature of the different solvents. The UV-visible spectrum of Schiff base *N*-[4-fluoroaniline]thiophene carboxaldimine in DMF shows a band at $\lambda_{\text{max}} = 264$ nm. In DMSO, the absorption maximum shifted towards longer wavelength (280 nm). Increase in polarity of the solvent [19] leads to red shift or also known as positive solvatochromism. This confirms that the molecules are more polarized in the excited state *i.e.* the molecule is stabilized by the polar solvents in the excited state. The spectrum was also recorded in the solvents chloroform, acetonitrile and ethanol. The intensity of absorption is maximum for highly polar solvent DMSO and it decreases when the polarity of the solvent is reduced. The maximum absorbance was noted for DMSO and minimum was noted for ethanol.

UV-visible studies: A gradual decrease in the absorbance at 260/265 nm (hypochromic effect) was observed by the incremental addition of $\text{CaCl}_2/\text{MgCl}_2$ solution (Fig. 3). The intensity

Fig. 3. Electronic absorbance spectrum of ligand in DMSO-water solution in presence of Ca^{2+} ion

variations of the bands on increasing the concentration of $\text{Ca}^{2+}/\text{Mg}^{2+}$ ions may lead to study of concentration of $\text{Ca}^{2+}/\text{Mg}^{2+}$ ions in aqueous solution [20].

Antimicrobial activity: The Schiff base and its metal complexes were tested for their inhibitory effects on the growth of bacteria and fungus. Table-4 suggested that the synthesized metal complexes are more effective in inhibiting the growth of microorganisms than the Schiff base against some microorganisms under identical experimental conditions. The complex ion polarity is lowered [21] mainly due to the partial sharing of its positive charge with the donor atoms and possibly the π -electron delocalization within the whole chelate ring system, thus formed during coordination. The lipophilic nature of the central metal atom is increased due to the chelation process, which in turn initiates its permeation through the lipid layer of the membrane. This would propose that the chelation could encourage the capacity of a complex to cross a cell film and can be clarified by Tweedy's chelation hypothesis. The comparative results of Schiff base and complexes shows that Cu (II) and Zn (II) complexes have effective inhibition activity for *E. coli* than the other bacterial strains [22-24].

TABLE-4
ANTIMICROBIAL ACTIVITY OF
LIGAND & ITS METAL COMPLEXES

Compound	Zone of inhibition (mm)* against			
	<i>E. coli</i>	<i>P. aeruginosa</i>	<i>S. aureus</i>	<i>C. albicans</i>
Ligand	9	9	10	8
Fe(III) complex	9	9	7	7
Co(II) complex	11	10	11	9
Ni(II) complex	10	11	10	7
Cu(II) complex	13	10	11	10
Zn(II) complex	15	10	14	7
Standard	23	26	21	16

*Each value observed are the mean (n=3) within the error limits of ± 2 .

DNA binding studies

Electronic absorption studies: The concentration of DNA solutions varied from $1.14-1.01 \times 10^{-4}$ M for Zn(II) with absorption band 270 nm. In the UV region, intra ligand $\pi-\pi^*$ transition was attributed to the intense absorption band of zinc(II) complex observed at 270 nm. On addition of HS-DNA to Zn(II) complex, a significant hypochromic effect in the absorption bands with blue shift of 5-10 nm (hypsochromism) was observed. For other complexes, Fe(III): $2.86 - 2.69 \times 10^{-4}$, absorption band at 707 nm; Ni(II): $1.59-1.63 \times 10^{-4}$, absorption band at 772 nm; Cu(II): $1.74-1.07 \times 10^{-4}$, absorption band at 549 nm (Fig. 4), Co(II): $3.50-3.01 \times 10^{-4}$, absorption band at 676 nm. On expanding the convergence of DNA arrangement, all the metal complexes show hypochromism and bind with HS-DNA in an intercalative mode with electrostatic authoritative. In this way, HS-DNA has a two-fold helix structure and bind with metal ions in electrostatic way.

The association constants of complex with DNA were calculated using Benesi-Hildebrand equation [17].

$$\frac{A_0}{A - A_0} = \left(\frac{\epsilon_G}{\epsilon_{H-G}} - \epsilon_G \right) + \left(\frac{\epsilon_G}{\epsilon_{H-G}} - \epsilon_G \right) \times \frac{1}{K_b [DNA]}$$

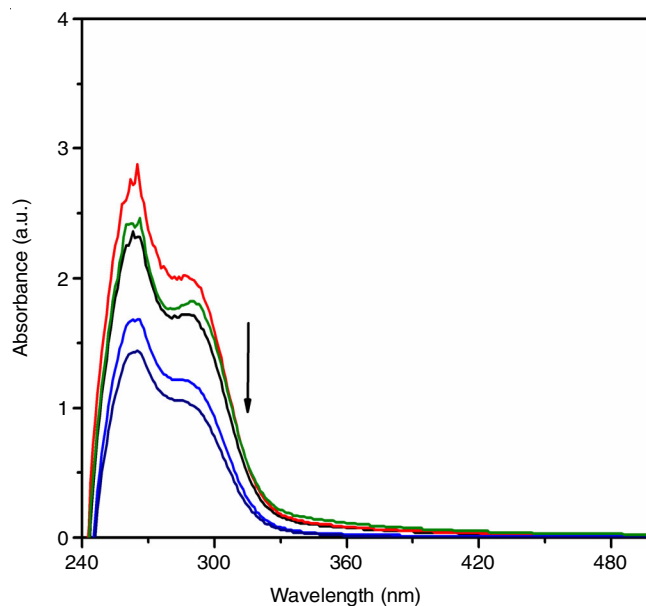


Fig. 4. Electronic spectra of copper complex with increasing DNA concentration in Tris-HCl/NaCl buffer solution

where K_b is the association constant, A_0 and A are the intensities of the free metal(II) complex and the complex bound with DNA, respectively, and ϵ_G and ϵ_{H-G} are the absorption coefficients of the complex and the complex bound with the DNA, respectively. K_b is calculated by taking the slope to the intercept ratio of $A_0/A - A_0$ versus $1/[DNA]$ plot. The values of association constant are presented in Table-5. It can be inferred that the copper(II) complex possess greater binding ability with the DNA than the other metal complexes [25,26].

TABLE-5
ASSOCIATION CONSTANT VALUES OF METAL COMPLEXES

Compounds	Association constant
Fe(III) complex	12.0×10^3
Co(II) complex	23.3×10^3
Ni(II) complex	12.5×10^3
Cu(II) complex	25.0×10^3
Zn(II) complex	15.0×10^3

DNA cleavage studies by gel electrophoresis: The studies clearly indicated that the Schiff base and its metal(II) complexes have performed on DNA as there was variation in the bands of lanes compared to the control DNA. The metal complexes are capable to change supercoiled DNA into open circular DNA [27]. The reaction is regulated by a bound hydroxyl radical or a peroxy species generated from H_2O_2 . From Fig. 5, it is noted that the control DNA does not exhibit any significant cleavage of HS-DNA even on longer exposure time. The Cu(II) complex cleaves DNA as compared to control DNA while other complexes do not split that much in the presence of H_2O_2 . The presence of a smear in the gel diagram indicates the presence of radical cleavage and the cleavage efficiency of the complexes is comparable to that of the control is due to their promising DNA-binding ability. As the Cu(II) complex was able to cleave the DNA very well, it may be reasoned that the compound restrains the development of the pathogenic life form by cleaving genome.

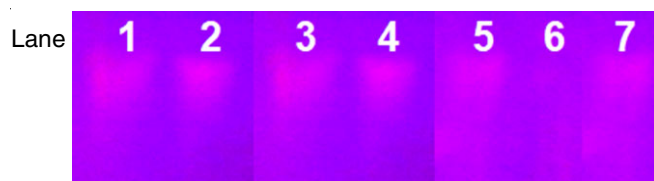


Fig. 5. DNA cleavage efficiency of ligand and its metal complexes. Lane 1: DNA + H₂O₂; Lane 2: DNA +H₂O₂ + ligand; Lane 3: DNA + H₂O₂ + Fe(III) complex; Lane 4: DNA + H₂O₂ + Co(II) complex; Lane 5: DNA + H₂O₂ + Ni(II) complex; Lane 6: DNA +H₂O₂ + Cu(II) complex and Lane 7: DNA +H₂O₂ + Zn(II) complex

Conclusion

The Schiff base complexes of Fe(III), Co(II), Ni(II), Cu(II) and Zn(II) derived from 2-thiophene carboxaldehyde and 4-fluoroaniline were synthesized and characterized. The analytical results support the stoichiometric ratio of ligand and its metal complexes are 1:2 ratio. The Schiff base acts as a bidentate ligand which coordinates through sulphur and nitrogen atoms. The results of this investigation support the suggested octahedral structure of all the metal complexes. In absorbance studies, the interaction of Ca²⁺ and Mg²⁺ ions with the Schiff base ligand, gives a way to determine the concentration of calcium and magnesium ions in physiological processes. The Schiff base ligand and its metal complexes were found to have maximum therapeutic activity against *E. coli* compared to other the species. The DNA binding studies suggested the intercalative mode with electrostatic binding and the higher value of association factor of Cu(II) complex may lead to design a creative anticancer drug. The interaction of these complexes with HS-DNA was investigated by gel electrophoresis and concluded that Cu(II) complex cleaves DNA in the presence of H₂O₂, whereas the control DNA, ligand and other complexes are not effective.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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