CODEN [USA]: IAJPBB ISSN: 2349-7750

INDO AMERICAN JOURNAL OF
PHARMACEUTICAL SCIENCES

http://doi.org/10.5281/zenodo.1470665

Available online at: http://www.iajps.com Research Article

METAL BASED PHARMACOLOGICALLY DYNAMIC MIXED LIGAND CU (II), NI (II) AND ZN (II) COMPLEXES: SYNTHESIS, SPECTRAL AND DNA INTERACTION INVESTIGATION

S. Iyyam Pillai[1*], C. Joel[2], K. VIjayaraghavan[1] and S. Subramanian[3]

[1] P.G and Research Department of Chemistry, Pachaiyappa's College, Chennai, Tamilnadu, India

[2] P.G. Department of Chemistry, St. John's College, Tirunelveli, Tamilnadu, India

[3] Department of Biochemistry, University of Madras, Guindy Campus, Chennai, Tamilnadu, India

Abstract:

Small inorganic complexes that can interact with DNA or other biological molecules have tremendous conservation. The Schiff base mixed ligand L and its complexes have been characterized with the aid of elemental analysis, FT-IR, NMR, Mass, elemental analysis and electronic studies. Binding interaction of this complex with calf thymus (CT-DNA) has been investigated by emission, absorption, circular dichroism, and viscosity techniques. The complex displays significant binding properties to the CT-DNA. In fluorimeteric analysis, the binding mode of the complex with CT-DNA was investigated using Ethidium bromide as a fluorescence probe. The EB displacement assay also reveals the same trend as observed in UV–Vis spectral titration. The addition of the synthesized complexes to the DNA bound ethidium bromide (EB) solutions causes an obvious reduction in emission intensities indicating that these complexes competitively bind to DNA with EB. The CD spectra of these complexes show a red shift of 3–10 nm in the positive band with increase in intensities. The experimental results from all techniques i.e. binding constant (K_b), apparent binding constant (K_{app}) were in good conformity and inferred spontaneous DNA- complexes formation via intercalation. Among all the compounds Schiff base Cu(II) showed comparatively greater binding at pH 4.7 as evident from its greater K_b values 6.03 x 10^4 M^{-1}, (UV); 6.63 x 10^5 M^{-1} respectively (fluorescence). The data obtained suggests that the complexes interact with DNA by intercalative binding.

Key words: *Curcumin, Isatin, DNA binding, Intercalation, Mixed ligand complex, Schiff base.*

***Corresponding author:**

Dr. S. Iyyam Pillai,

Assistant Professor,
P.G and Research Department of Chemistry,
Pachaiyappa's College,
Chennai, Tamilnadu,
India
Mobile: 72990 36480
E-mail: iyyampillaichemistry@gmail.com

QR code

Please cite this article in press S. Iyyam Pillai et al., ***Metal Based Pharmacologically Dynamic Mixed Ligand Cu (II), Ni (II) and Zn (II) Complexes: Synthesis, Spectral and DNA Interaction Investigation.,*** *Indo Am. J. P. Sci, 2018; 05(10).*

1. INTRODUCTION:

Inorganic elements play crucial role in biological and biological medical processes, and it is evident that many organic compounds used in medicine do not have a purely organic mode of action, some are activated or bio transformed by metal ions metabolism . Many drugs possess modified toxicological and pharmacological properties in the form of metal complex and probably Schiff bases are versatile C=N (Imine) containing compounds possessing broad spectrum of biological activity and incorporation of metals in form of complexes showed some degree of antibacterial, antifungal , antitumor and anti-inflammatory activity[1]. Transition metal complexes containing Schiff base ligands have been of specific interest for many years. Synthesis and characterization of such complexes of the first row transition metals including copper and cobalt have received overwhelming attention in recent times due to their important catalytic, magnetic and biological properties in addition to the structural aspects [2].

DNA is the pharmacologic target of many drugs currently in clinical use or in advanced clinical trials. The interaction of Schiff base metal complexes with DNA has been widely studied in the past decades. In fact, the activity of numerous anticancer, antimalarial and antibacterial agents finds its origin in intercalative interactions with DNA. The identification of metal complex–DNA interaction is a fundamental importance to the understanding of the molecular basis of therapeutic activity. The consequence of DNA cleavage in pharmaceutical and biotechnological applications has enthused researchers to keep developing compounds that efficiently cleave nucleic acid molecule [3]. Interaction between small molecules and DNA provides a structural guideline in rational drug design administration for the synthesis of new, improved chemical drug entities with enhanced or more selective activity, thereby greater clinical efficacy and lower toxicity. Metal complexes such as cobalt, nickel, copper and zinc with N-containing ligands have shown astonishing binding and cleavage activities [4].

Isatin and its derivatives are unique members in the Schiff base family. The simple isatin based Schiff base compounds having, acyl, aroyl and heteroacroyl Schiff bases have additional donor sites >C=O, >C=N-, etc. These donor sites make them more flexible and versatile. This versatility has made them excellent chelating agents that can form a variety of complexes with various transition and inner transition metals and has attracted the attention of many researchers [5]. Further, Isatin has also been found in mammalin tissues [6]. Schiff bases derived from isatin exhibit many neurophysiological and neuropharmacological effects like antimicrobial, antiviral, anticonvulsant, anticancer, antimycobacterial, antimalarial, cysticidal, herbicidal and anti-inflammatory activity [7-9]. Curcumin (1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6 heptadiene -3,5-dione) is an important natural phytochemical compound found in the rhizomes of Curcuma longa or turmeric, which has been used in China to treat digestive and neuropsychiatric disorders [10]. The derivatives along with synthetic analogs of curcumin and its various type of in-house synthesized metal complexes are mainly the substances which have attracted scientists and are still receiving paramount attention in that era due to their dramatic and very special properties [11,12]. In Ayurveda curcumin is known as a blood purifier and recently, its chelating ability with different metal ions have been explored which is responsible for reduction of the deposition of metal ions in the human body [13].The study of Schiff base hydrazones has been growing interest because of their antimicrobial, anti-tuberculosis, and anti-tumor activity [14,15]. Hydrazone compounds have played an important role in the development of coordination chemistry, due to their ability to form a range of stable complexes which have applications in different fields. These include their role in biological, medical and environmental sciences [16].

The coordination chemistry of transition metal complexes with mixed ligands are of current interest because they can provide new materials with useful properties such as magnetic exchange, electrical conductivity, photoluminescence, nonlinear optical property, and antimicrobial activity. The biological importance of mixed ligand complexes is that they are sometimes more effective than the free ligands. Mixed-ligand complexes containing nitrogen and oxygen donors are important owing to their antifungal, antibacterial, and anticancer activities[17]. In view of diversified roles of Schiff base transition metal complexes, in this paper we have synthesized and characterized the Cu(II), Ni(II) and Zn(II) mixed ligand metal complexes with primary ligand derived from curcumin and Isatin and secondary ligand as hydrazine hydrate. The Schiff base and their metal complexes were screened for their DNA binding activities.

2. EXPERIMENTAL PROTOCOLS:

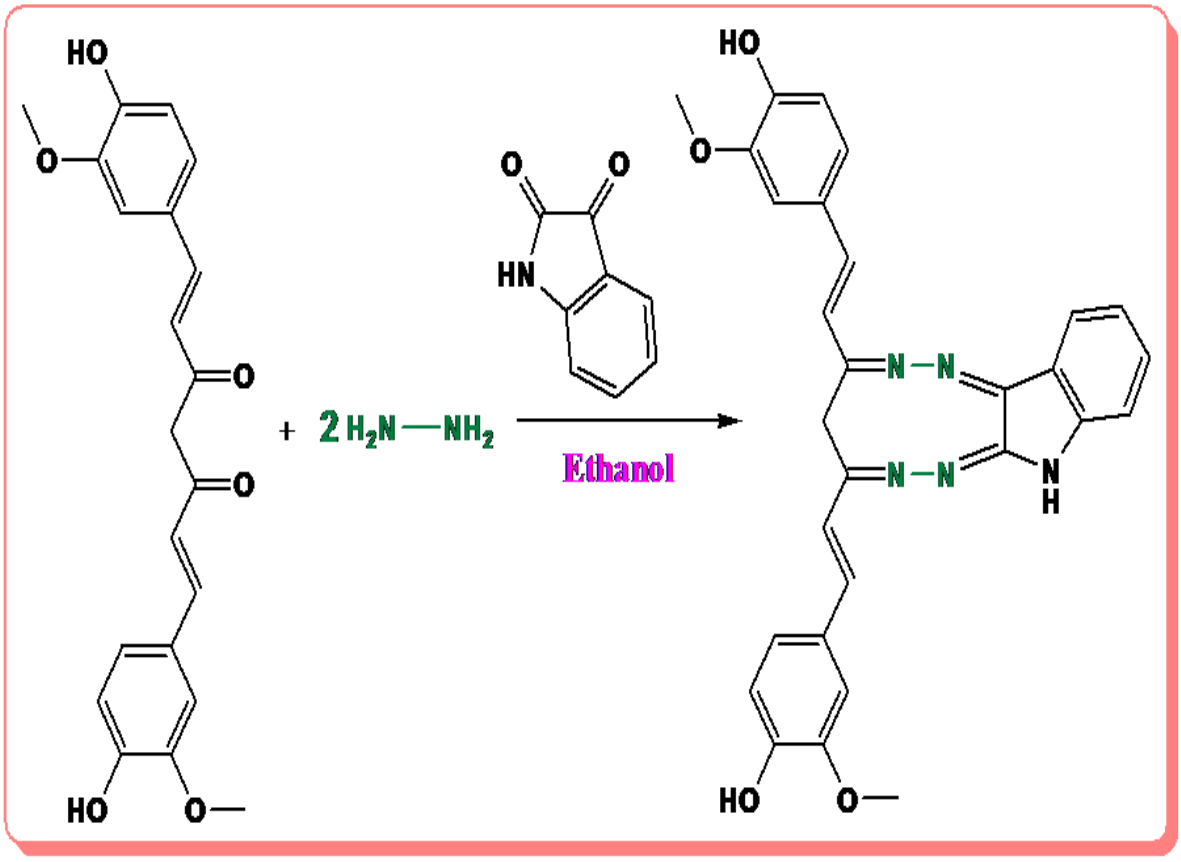
2.1 Reagents and instruments

All the chemicals used in the current work *viz*, 1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6 heptadiene -3,5-dione (Curcumin), hydrazine hydrate, (1H-Indole-2,3-dione) Isatin and metal(II) acetates were

of analytical reagent grade (produced by were obtained from Fluka and Sisco Chemicals. Commercial solvents were distilled and then used for the preparation of ligands and their complexes. DNA was purchased from Bangalore Genei (India). Tris(hydroxymethyl)aminomethane–HCl (Tris–HCl) buffer solution was prepared by means of deionized and sonicated triple distilled water. The IR spectral analysis was carried out in the solid state as pressed KBr pellets by means of a Perkin-Elmer FT-IR spectrophotometer in the range of 400-4000 cm^{-1}. The electro spray mass spectra were establlished on a Q-TOF micro mass spectrometer. The emission spectra were recorded on Perkin Elmer LS-45 fluorescence spectrometer. The electronic spectra in diverse solvents were recorded on using a Perkin Elmer Lambda 35 spectrophotometer working in the range of 200-900 nm with quartz cells and ε are given in $M^{-1}cm^{-1}$. Circular dichoric spectra of CT-DNA were obtained using a JASCO J-715 spectropolarimeter equipped with a Peltier temperature control device at 25 ± 0.1°C with 0.1 cm path length cuvette. Viscosity measurements were verified using a Brookfield Programmable LV DVII+ viscometer. The entire glassware used was made of Pyrex or corning glass. The burettes, pipettes, and standard flasks were calibrated by the technique described by Vogel[18]. An analytical balance of 100µg sensitivity was used for weighing samples.

2.2 Synthesis of Schiff base ligand L

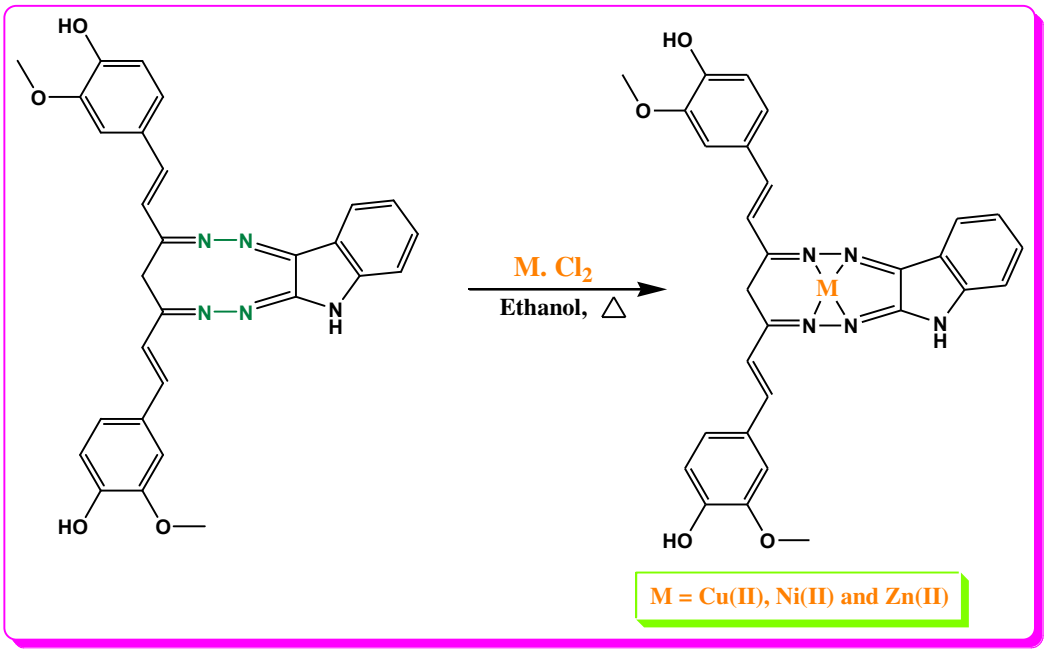
The Schiff base ligand L used in current research was prepared by reacting hot ethanolic solution (0.001 mol, 20 mL) 1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6 heptadiene -3,5-dione with hydrazine hydrate (0.002 mol) with methanolic solution (25 mL). To this hot stirring solution 1H-Indole-2, 3-dione (0.001 mol) is added with constant stirring. The mixture was refluxed for 6-7 h on water bath and allowed to cool at room temperature. The coloured compound formed was filtered, washed, dried and recrystallized from ethanol.



Scheme 1. Synthesis of Schiff base ligand L.

2.3 Synthesis of Schiff base ligand L and their respective complexes:

All the complexes were prepared by the following general method. Hot ethanolic solution of metal chloride salts (0.001 mol, 25 mL) were added to hot ethanolic solution of the ligand (0.001 mol, 25 mL) in 1:1 ratio. The reaction mixture was heated under reflux for about 6 hours. The resulting solution was concentrated to half its volume and allowed to cool. On cooling the complexes separated out which were filtered and washed several times with aqueous ethanol, diethyl either and finally dried over in vacuo.



Scheme 2. Synthesis of Schiff base metal complexes.

2.4 DNA binding and cleavage activity

2.4.1 Electronic absorption titration

All spectroscopic titrations were carried out in 5 mmol L–1 Tris–HCl buffer (pH 7.1) containing 50 mmol L^{-1} NaCl at room temperature. A solution of CT−DNA in the buffer gave a ratio of V absorbance at 260 and at 280 nm of 1.84:1, indicating that the DNA was sufficiently free of protein[19]. The concentration of DNA was estimated from its absorption intensity at 260 nm with a known molar absorption coefficient value of 6600 M^{-1} cm^{-1}. Stock solutions were stored at 4 °C and used within 4 days [20]. Before adding CT-DNA to complexes, complexes were dissolved in less than 4 % DMSO: H_2O. Titration experiments were performed varying the concentration of the CT DNA from (0, 40, 80, 120, 160, 200, 300 and 400 µM) while keeping the drug and its metal complexes concentration constant (40 µM). While measuring the absorption spectra, equal quantity of CT DNA was added to both the complex solution and the reference solution to eliminate the absorbance of CT DNA itself. The complex-DNA solutions were allowed to equilibrate for 10 min before spectra were recorded. The intrinsic binding constant (K_b) for the interaction of the complexes with CT DNA was determined from a plot of [DNA]/(εa - εf) versus [DNA] using absorption spectral titration data[21] and the following equation,

$$[DNA]/(\varepsilon_a - \varepsilon_f) = [DNA]/(\varepsilon_b - \varepsilon_f) + 1/K_b(\varepsilon_b - \varepsilon_f) \quad (1)$$

where [DNA] is the concentration of DNA, the apparent absorption coefficients ε_a, ε_f and ε_b correspond to A_{obsd}/[complex], the extinction coefficient for the free metal complex and the extinction coefficient for the metal complex in the fully bound form, respectively. The K_b value is given by the ratio of the slope to the intercept.

2.4.2 Fluorescence spectral study

The emission spectrum is obtained by setting the excitation monochromator at the maximum excitation wavelength and scanning with emission monochromator. Often an excitation spectrum is first made in order to confirm the identity of the substance and to select the optimum excitation wavelength. Further experiments were carried out to gain support for the mode of binding of complexes with CT-DNA. Non-fluorescent or weakly fluorescent compounds

can often be reacted with strong fluorophores enabling them to be determined quantitatively. On this basis molecular fluorophore Ethidium Bromide was used which emits fluorescence in presence of CT-DNA due to its strong intercalation. Quenching of the fluorescence of EthBr bound to DNA were measured with increasing amount of metal complexes as a second molecule and Stern–Volmer quenching constant K_{sv} was obtained from the following equation [22].

$$Io/I=1 + K_{svr}. \quad (2)$$

Where I_0, is the ratio of fluorescence intensities of the complex alone, I is the ratio of fluorescence intensities of the complex in the presence of CT-DNA. K_{sv} is a linear Stern – Volmer quenching constant and r is the ratio of the total concentration of quencher to that of DNA, [M] / [DNA]. A plot of I_0 / I_{vs}. [complex]/ [DNA], K_{sv} is given by the ratio of the slope to the intercept. The apparent binding constant (K_{app}) was calculated using the equation K_{EB}[EB] / K_{app}[complex], where the complex concentration was the value at a 50% reduction of the fluorescence intensity of EB and K_{EB} = 1.0 x 10^7 M^{-1} ([EB] = 3.3 µM).

2.4.3 Viscosity measurements

Viscosity titration analyses were carried out by means of a capillary viscometer at a unvarying temperature (25.0 ± 0.1 ^{0}C). Flow times were calculated with a digital stopwatch, and each sample was measured three times, and then a standard flow time was calculated. To get the binding mode of the complexes towards CT-DNA, viscosity dimensions were carried out on CT-DNA (0.5 mM) by varying the concentration of the complexes (0.01 mM, 0.02 mM, 0.03 mM, 0.04 mM, 0.05 mM). Data were accessible as (η/η_o) versus binding ratio of concentration of complex to that of concentration of CT-DNA , where η is the viscosity of DNA in the presence of complex and η_o is the viscosity of DNA alone.

2.4.4. CD spectral studies

The CD spectral analysis of CT-DNA in the presence or absence of complexes were collected in Tris–HCl buffer (pH=7.2) containing 50 mM NaCl at room temperature. The spectra were recorded in the region of 220–320 nm for 200 µM DNA in the presence of 100 µM of the complexes. Each CD spectrum was collected after averaging over at least three accumulations using a scan speed of 100 nm min^{-1} response time. Machine plus cuvette base lines, and CD contribution by the CT-DNA and Tris buffer were subtracted and the resultant spectrum zeroed 50 nm outside the absorption bands. Circular dichroic spectra of CT DNA in the presence and absence of metal complexes were obtained by using a JASCO J-715 spectropolarimeter equipped with a Peltier temperature control device at 25 ± 0.1 °C with a 0.1 cm path length cuvette.

RESULTS AND DISCUSSION:

3.1 Structural characterization of the Schiff base ligand (L) and their complexes.

3.1.1 FT-IR spectral analysis.

The IR spectrum of Schiff base ligand L, a sturdy band roughly around 1638 cm^{-1} was suitable to the azomethine linkage and absence of free NH_2 groups around 3250 cm^{-1} confirms the condensation between Curcumin, Isatin and Hydrazine hydrate as represented in Figure 1.The IR spectra of metal complexes are like to each other, except for slight shifts and intensity changes of few vibration peaks caused by different M(II) ions, which indicate that the complexes have similar structure as represented in Figure 2 and S1, S2. In order to study the bonding mode of Schiff base ligand L to the metal complexes, the IR spectrum of the free ligand is compared with the spectra of the complexes. A sharp band at 1638 cm^{-1} in the IR spectrum of the Schiff base ligand (CH=N) shifts downward by about 1620 - 1609 cm^{-1} in all the complexes indicating coordination through azomethine nitrogen. The bands around 1375–1390 cm^{-1} in all the complexes is due to the methyl (CH_3) frequency, is not affected upon complexation. Additional sustain for the formation of the complexes were provided by the existence of medium intense bands in the region 460 - 480 cm^{-1} assigned to the M-N [23].

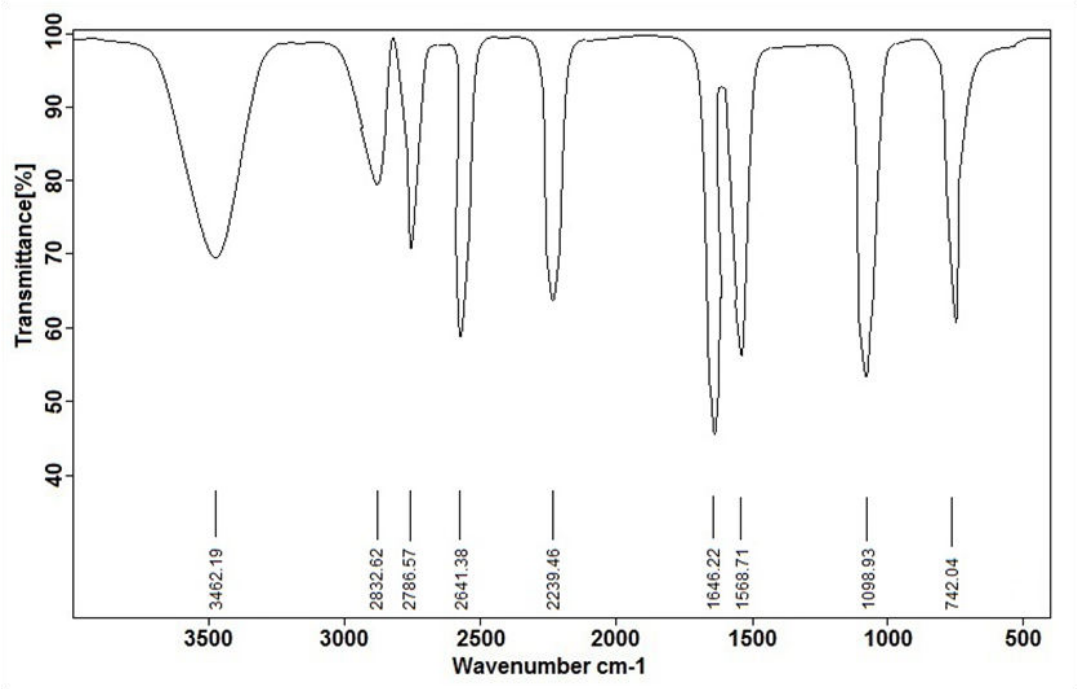


Figure 1. FT-IR spectra of Schiff base mixed ligand L.

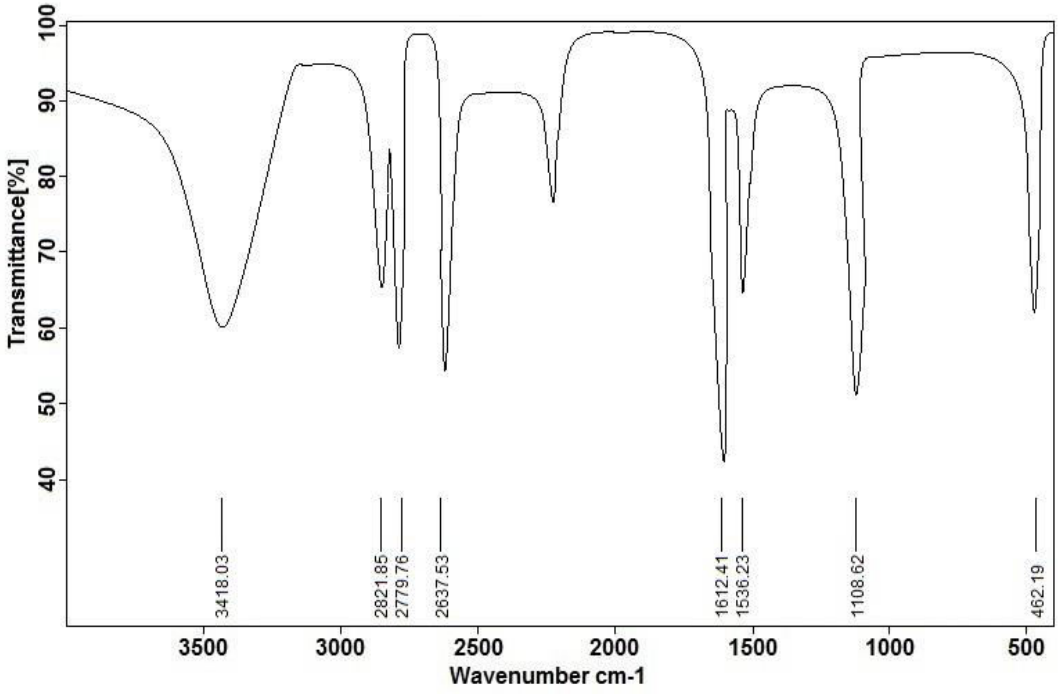


Figure 2. FT-IR spectra of Schiff base Cu (II) complex.

3.1.2 ESI-MS spectrum.

The molecular ion peak [M+] at m/z = 507 confirms the molecular weight of the macrocyclic Schiff base ligand $C_{29}H_{25}N_5O_4$. The peaks at m/z = 475, 447, 415, 367, 330, 305, 255, 215, 161 and 122 corresponds to the various fragments $C_{29}H_{25}N_5O_2$, $C_{27}H_{21}N_5O_2$, $C_{27}H_{21}N_5$, $C_{23}H_{21}N_5$, $C_{21}H_{22}N_4$, $C_{18}H_{19}N_5$, $C_{14}H_{17}N_5$, $C_{11}H_{13}N_5$ and $C_7H_7N_5$ respectively as shown in Figure 3. This confirms the molecular structure of the ligand L. The molecular ion peak [M+] at m/z = 571 confirms the molecular weight of the macrocyclic Schiff base Cu(II) complex $C_{29}H_{25}CuN5O_4$. The peaks at m/z = 511, 479, 428, 354, 302, 274, 224 and 185 corresponds to the various fragments respectively as shown in Figure 4 and Figure S3and S4. The molecular ion peak [M+] at m/z = 566 and 572 confirms the molecular weight of the macrocyclic Schiff base Ni(II) and Zn(II) complex $C_{29}H_{25}N_5O_4M$ [M = Zn and Ni]. The type of fragmentation observed in Zn(II) and Ni(II) complex was similar with that of the Cu(II) complex.

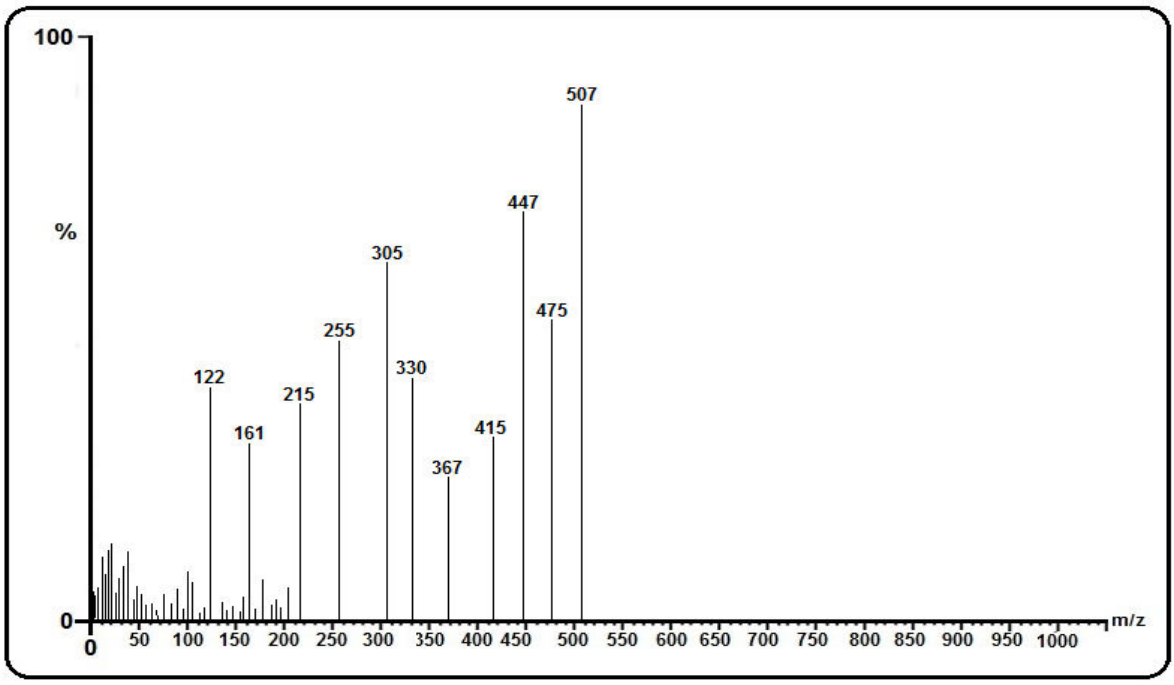


Figure 3. Mass Spectra of Schiff base mixed Ligand L.

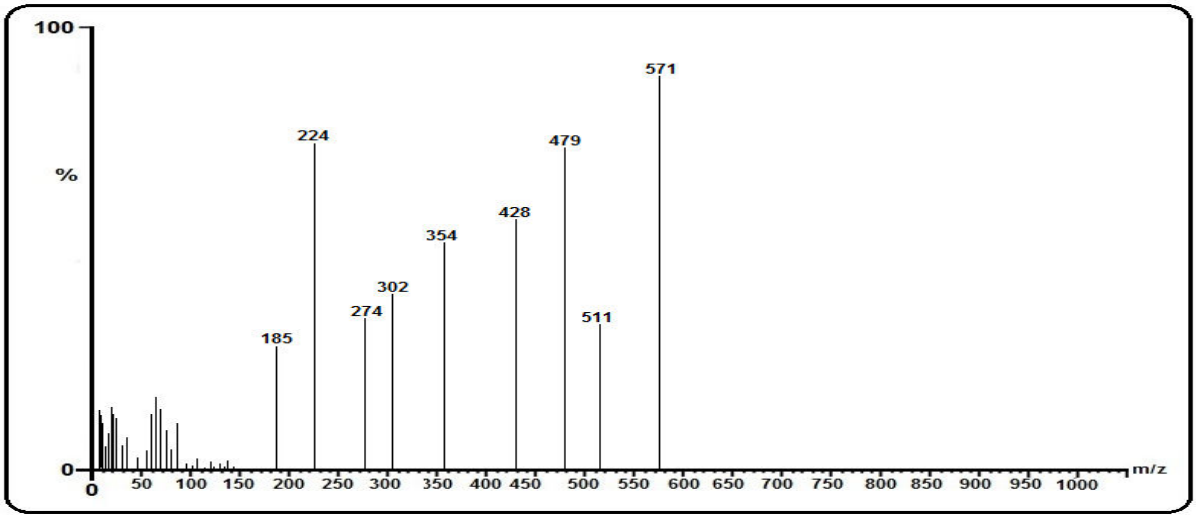


Figure 4. Mass Spectra of Schiff base mixed Ligand Complex Cu(II).

3.2 DNA binding experiments

3.2.1 Absorption spectral studies

Electronic absorption spectroscopy is usually employed to determine the binding of complexes with the DNA helix. A complex binding to DNA through intercalation usually results in hypochromism and bathochromism, due to intercalation involving a strong π-π* stacking interaction between an aromatic chromophore and the base pairs of DNA. The extent of hypochromism in the UV band is consistent with the strength of intercalative interaction [24]. The prominent variations in the spectra also suggest the tight complexation of synthesized molecule with DNA, which resulted in the alteration in the absorption maxima of the DNA as presented in Figure 5 and S5 –S6. These results recommended an intimate association of the compounds with CT-DNA and it is also expected that these compounds bind to the helix *via* intercalation [25]. Later than the compounds intercalate to the base pairs of DNA, the π* orbital of the intercalated compounds could couple with π orbitals of the base pairs, thus diminishing the π → π* transition energies. Therefore, these interactions resulted in the observed hypochromism [26]. The Cu (II) complex showed more hypochromicity than the other complexes, indicating that the binding strength of the copper (II) complexes are much stronger than that of the other synthesized complexes. In order to affirm quantitatively the affinity of the complex bound to DNA, the intrinsic binding constants K_b of the complex with DNA was obtained by monitoring the changes in absorbance at 354 nm for the title complex with increasing concentration of DNA as shown in inset of Figure 5. The Kb values obtained for the synthesized complexes are as follows: for Cu(II) complex 6.03 x 10^4 M^{-1}, for Ni(II) complex 4.87 x 10^4 M^{-1} and for Zn (II) complex is 3.78 x 10^4 M^{-1}.

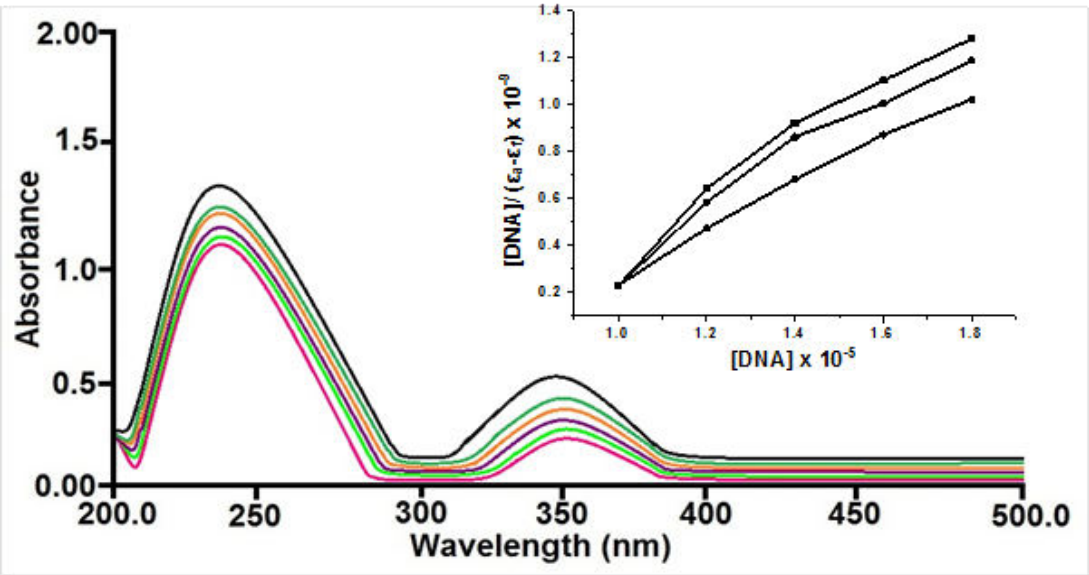


Figure 5 **The absorption spectra of complex Cu(II)(1 x 10^{-5} M) in the absence and presence of increasing amounts of CT-DNA (0-2.5 x 10^{-3} M) at normal temperature in 50 mM Tris-HCl / NaCl buffer (pH = 7.5). The Inset shows the plots of [DNA] / (ϵ_a-ϵ_f) versus [DNA] for the titration of DNA with Cu(II), Ni(II) and Zn(II) complexes.**

3.3.2. Fluorescence emission spectrometry

The competitive binding experiment was carried out to clarify the interaction of the complexes with DNA. The fluorescence intensity of DNA or EB in Tris buffer is very low. However, EB, which is one of the most sensitive fluorescence probes, emits intense fluorescence at about 600 nm in the presence of DNA due to its strong intercalation between the adjacent DNA base pairs[27] and this enhanced fluorescence could be quenched by the addition of another molecule due to the decreasing binding sites of DNA available for EB [28].

The emission spectra of EB bound to DNA in the absence and presence of the complexes are prearranged in Figure 6, Figure S7 and S8. It can be renowned that the fluorescence intensity of the EB-DNA solutions reduces with the addition of the complex visibly. The results suggest that the complex can restore the EB and bind to the DNA molecule. The quenching plots (insets in Figure 6) illustrate that the fluorescence quenching of EB bound to DNA by Cu(II), Ni(II) and Zn(II) complexes in linear agreement with the Stern–Volmer equation, which corroborates that the complexes bound to DNA. The Kapp values for Cu(II), Ni(II) and Zn(II) complexes are found to be 6.63 x 10^5 M^{-1}, 5.12 x 10^5 M^{-1} and 4.53 x 10^5 M^{-1} respectively. The achieved results are in reliable with that of absorption spectroscopic studies. Anyway, it may be concluded that the whole complexes bound to DNA *via* the similar mode and the quenching constants of the synthesized complexes reveals that the interaction of the compound with DNA should be intercalation [29].

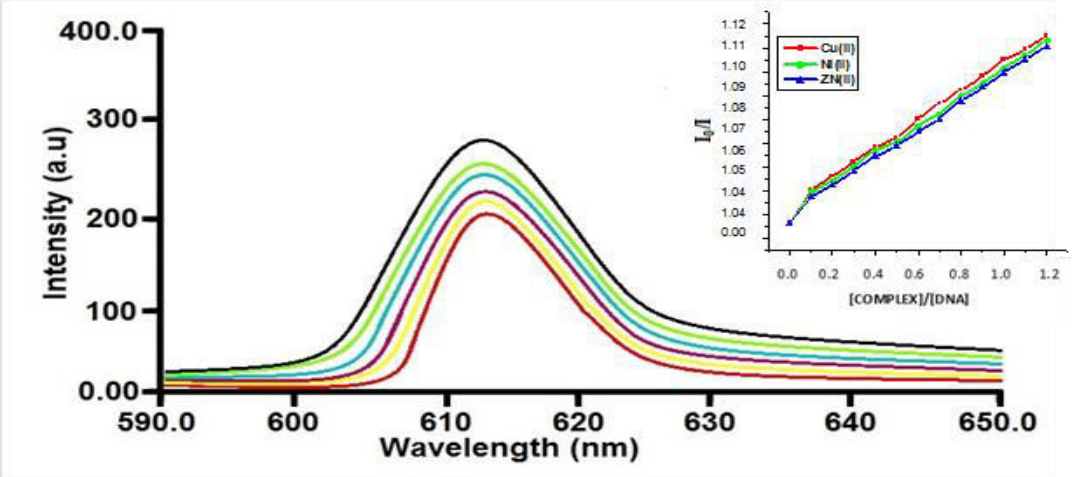


Figure 6. Emission spectrum of EB bound to DNA in the presence of Cu(II):([EB] =3.3 µM,[DNA] = 40 µM, [complex] = 0-25 µM, λex= 440 nm). Inset shows the plots of emission intensity I_o / I *vs* [DNA] / [complex] for the titration of DNA with Cu(II), Ni(II) and Zn(II) complexes.

3.2.3 Circular dichoric spectral studies

CD spectroscopy is a useful technique to analyze interactions between complex and CT-DNA. It is also useful because CD signals are quite sensitive to the mode of DNA interaction with small molecules [30]. The CD spectrum of CT DNA consists of a positive band at 275 nm that can be due to base stacking and a negative band at 245 nm that can be due to helicity and it is also characteristic of DNA in a right-handed B form [31]. As represented in Figure 7, the CD spectrum of DNA exhibits a positive absorption at 277 nm due to the base stacking and a negative band at 240 nm due to the helicity of B-DNA. In the presence of the complex, both the positive and negative peak intensities of the CD spectra of DNA were increased. The changes in the CD spectra in the presence of the complex show stabilization of the right handed B form of CT-DNA. These observations clearly indicate that the binding mode of the complexes should be intercalative, the stacking of the complex molecules between the DNA base pairs leads to an enhancement in the positive band and the partial unwinding of the helix decreases intensity of the negative band. So, the main interactions of the complexes with DNA can be ascribed to the intercalative mode.

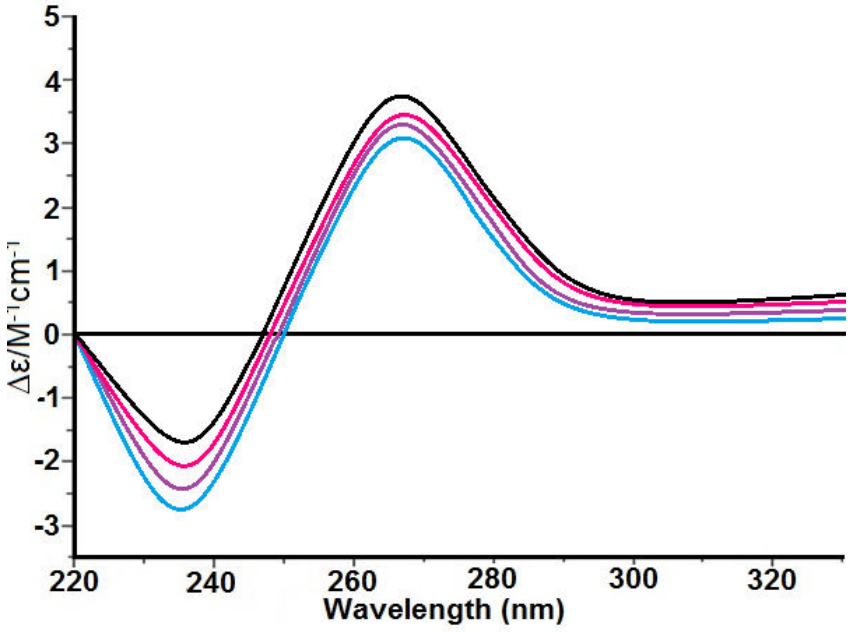


Figure 7CD spectra recorded over the wavelength range 220-320 nm for solutions containing 2:1 ratio of CT-DNA (200 µM) and mononuclear Cu(II), Ni(II) and Zn(II) complexes (100 µM).

3.3.4 Viscosity measurements

In the absence of crystallographic structure data, hydrodynamic methods which are sensitive to DNA length increases are regarded as the least ambiguous and the most critical tests of binding in solution. A classical intercalative mode demands that the DNA helix must lengthen as base pairs are separated to accommodate the binding ligand, leading to the increase of DNA viscosity [32]. The effects of complexes together with the viscosity of rod-like DNA are shown in Figure 8. On increasing the amounts of complexes, the relative viscosity of DNA increases steadily. The experimental results suggest that complexes bind to DNA through a classical intercalation mode. This helps complexes intercalate into the DNA base pairs deeply [33].

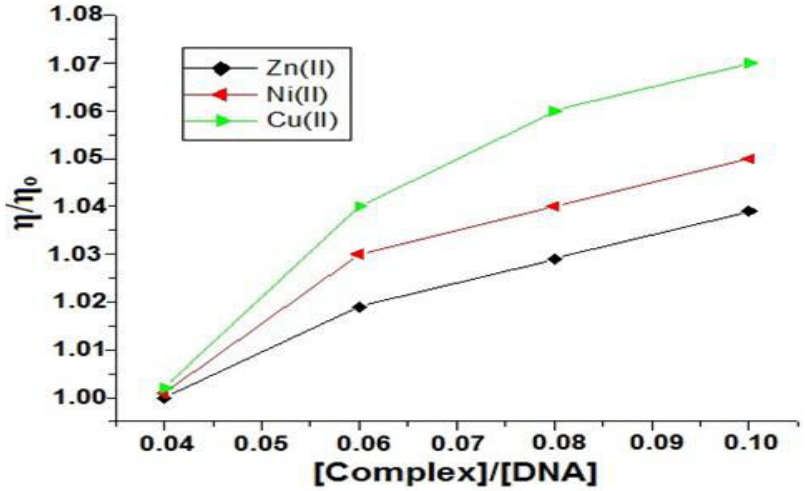


Figure 8 Viscosity measurements of the Cu(II), Ni(II) and Zn(II) complexes.

CONCLUSION:

In conclusion, we have successfully synthesized three new transition metal hydrazone complexes and characterized using various spectroscopic methods like FT-IR, UV-Vis and Mass spectra. Furthermore, we explored the binding interaction of the complexes CT-DNA in physiological buffer using UV–Vis, fluorescence, viscosity and circular dichoric spectral studies. The results suggest that complexes interact with CT-DNA by intercalative modes. Among the investigated complexes, the one containing copper as the central metal ion showed better binding affinity than the other two complexes containing zinc and nickel ions as metal counterparts respectively.

Abbreviations

DNA -	Deoxyribose nucleic acid
CT -	Calf Thymus DNA
Cu -	Copper
Ni -	Nickel
Zn -	Zinc
Tris-HCl -	Tris(hydroxymethyl)aminomethane
EB -	Ethidium Bromide
CD -	Circular Dichroism

REFERENCES:

1. R.R Gupta, Kumar, V.Gupta, Heterocyclic Chemistry I, (1998).
2. M. Fleck, D. Karmakar, M. Ghosh, A. Ghosh, R. Saha, D. Bandyopadhyay, Polyhedron 34 (2012) 157
3. N. Raman, R. Mahalakshmi, T. Arun, S. Packianathan, R. Rajkumar
4. E.K. Efthimiadou, G. Psomas, Y. Sanakis, N. Katsaros, J. Inorg. Biochem. 101 (2007) 525–535.
5. F. Tisato, C. Marzano, M. Porchia, M. Pellei and C. Santini , Medicinal Research Reviews. Vol. 30, 2010, pp.708- 749.
6. J.F.M. Da Silva, S.J. Garden and A.C. Pinto, J. Braz. Chem. Soc., 67(3), 273(2001).
7. N.R. Penthala, T.R. Yerramreddy, N.R. Madadi, P.A. Crooks, Bioorg. Med. Chem.Lett. 20 (15) (2010) 4468.
8. S.N. Pandeya, S. Smitha, M. Jyoti, S.K. Sridhar, Acta Pharm. 66 (2008) 43.
9. S.A. Imam, R.S. Varma, Experientia 31 (1975) 1287.
10. H. Hatcher, R. Planalp, J. Cho, F.M. Torti, S.V. Torti, Cell. Mol. Life Sci. 65 (2008) 1631.
11. M.I. Khalil, A.M. AL-Zahem, M.M. Qunaibit, Med Chem Res 23 (2014) 1683–1689.
12. Y. Mawani, C. Orvig, J. Inorg. Biochem.132 (2014) 52–58.
13. R.S. Priya, S. Balachandran, J. Daisy, P.V. Mohanan, Univ J Phys Appl. 3 (2015) 6–16
14. Nurchi VM, Crespo-Alonso M, Toso L, Lachowicz JI, Crisponi G (2013) J Inorg Biochem 128: 174-182.
15. Ramadan AAT, Rahman RMA, Seda MH (1992) Asian J. Chem 4: 569-578.
16. Tarafder MTH, Chew K, Crouse KA, Ali AM, Yamin BM, Fun HK (2002) Polyhedron 21(27–28):2683–2690.
17. M. B. Halli, V. B. Patil, R. B. Sumathi, Kinni Mallikarjun, Der Pharma Chemica, 2012, 4(6):2360-2367.
18. A.I. Vogel, "A Text Book of Quantitative Inorganic Analysis", 3rd Ed.; ELBS, Longman Green: London, 1961, 415-456.
19. S. Satyanarayana, J.C. Dabrowiak, J.B. Chaires, Biochemistry, 32 (1993) 2573-2584
20. S. Banerjee, S. Mondal, W. Chakraborty, S. Sen, R. Gachhui, R.J. Butcher, A.M.Z. Slawin, C. Mandal, S. Mitra, Polyhedron, 28 (2009) 2785-2793.
21. A. Wolfe, G.H. Shimer, T. Meehan, Biochemistry, 26 (1987) 6392-6396
22. J.R. Lakowiez, G. Webber, Biochem. 12 (1973) 4161.
23. M Belicchi-Ferrari, F Bisceglie, G Pelosi, P Tarasconi, Polyhedron, 2008, 27, 1361–1367.
24. A.M. Pyle, J.P. Rehmann, R. Meshoyrer, C.V. Kumar, N.J. Turro, J.K. Barton. J. Am. Chem. Soc., 111, 3051 (1989).
25. S.M. Hecht, J. Nat. Prod., 2000, 63,158.
26. M. Eriksson, M. Leijon, C. Hiort, B. Norden, A. Graeslund, Biochem., 1994, 33, 5031.
27. F.J. Meyer-Almes, D. Porschke, Biochemistry 32 (1993) 4246–4253.
28. B.C. Baguley, M.L. Bret, Biochemistry 23 (1984) 937–943.
29. D. S. Raja, N. S. P. Bhuvanesh, K. Natarajan, Inorg. Chim. Acta 385 (2012) 81.
30. E. J. Gao, Y. G. Sun, Q. T. Liu, L. Y. Duan, J. Coord. Chem., 2006, 59, 1295.
31. V.I. Ivanov, L. E. Minchenkova, A. K. Schyolkina, A. I. Poletayev, Biopolymers, 1973, 12, 89–110.
32. S. Satyanarayana, J.C. Dabrowiak, J.B. Chaires, J. Biochem. 31 (1992) 9319–9324.
33. E.C. Long, J.K. Barton, Acc. Chem. Res. 23 (1990) 271–273.

Supplementary Figure captions.
Figure S1. FT-IR spectrum of Ni(II) complex.
Figure S2. FT-IR spectrum of Zn(II) complex.
Figure S3. Mass spectrum of Ni(II) complex.
Figure S4. Mass spectrum of Zn(II) complex.
Figure S5. Absorption spectra of complexes Ni(II), (1 x 10-5 M) in the absence and presence of increasing amounts of CT-DNA (0-2.5 x 10-3 M) at room temperature in 50 mM Tris-HCl / NaCl buffer (pH = 7.5).
Figure S6. Absorption spectra of complexes Zn(II), (1 x 10-5 M) in the absence and presence of increasing amounts of CT-DNA (0-2.5 x 10-3 M) at room temperature in 50 mM Tris-HCl / NaCl buffer (pH = 7.5).
Figure S7. Emission spectrum of EB bound to DNA in the presence of Ni(II); ([EB] = 3.3 µM, [DNA] = 40 µM, [complex] = 0-25 µM, λex= 440 nm).
Figure S8. Emission spectrum of EB bound to DNA in the presence of Zn(II); ([EB] = 3.3 µM, [DNA] = 40 µM, [complex] = 0-25 µM, λex= 440 nm).

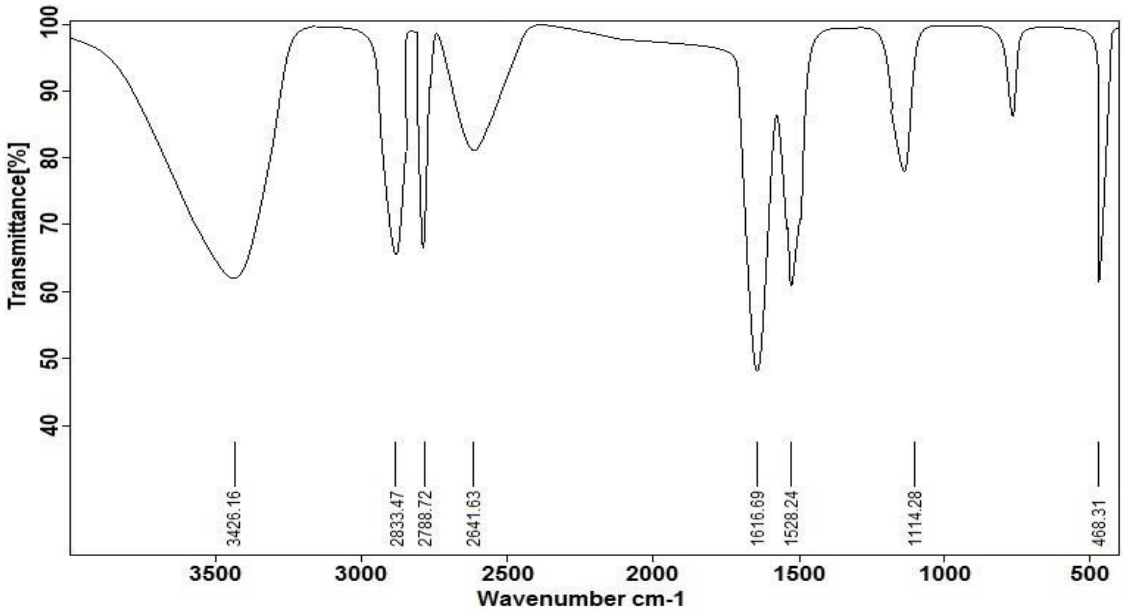


Figure S1: FT-IR spectrum of Ni(II) complex.

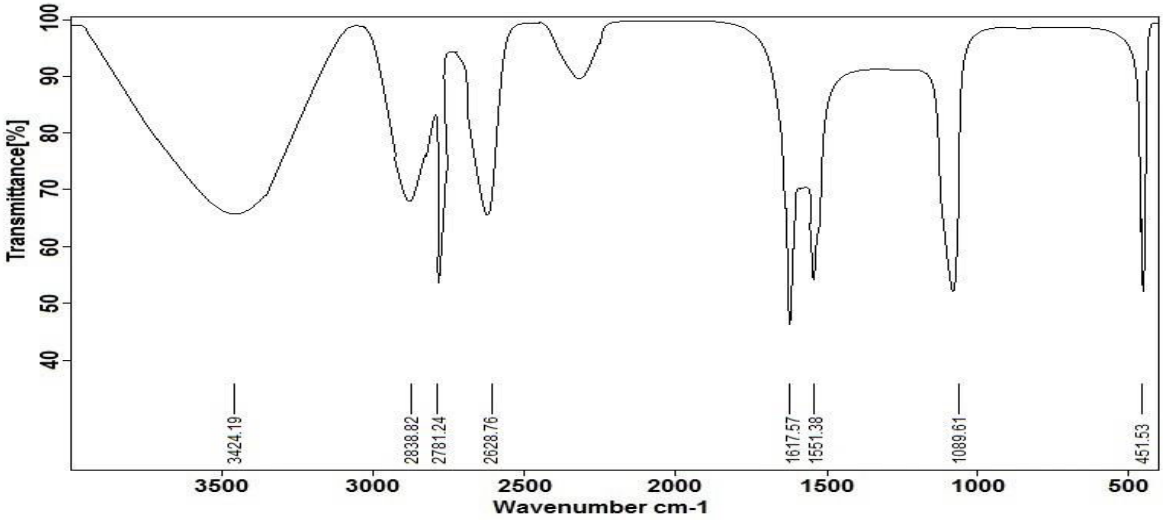


Figure S2: FT-IR spectrum of Zn(II) complex.

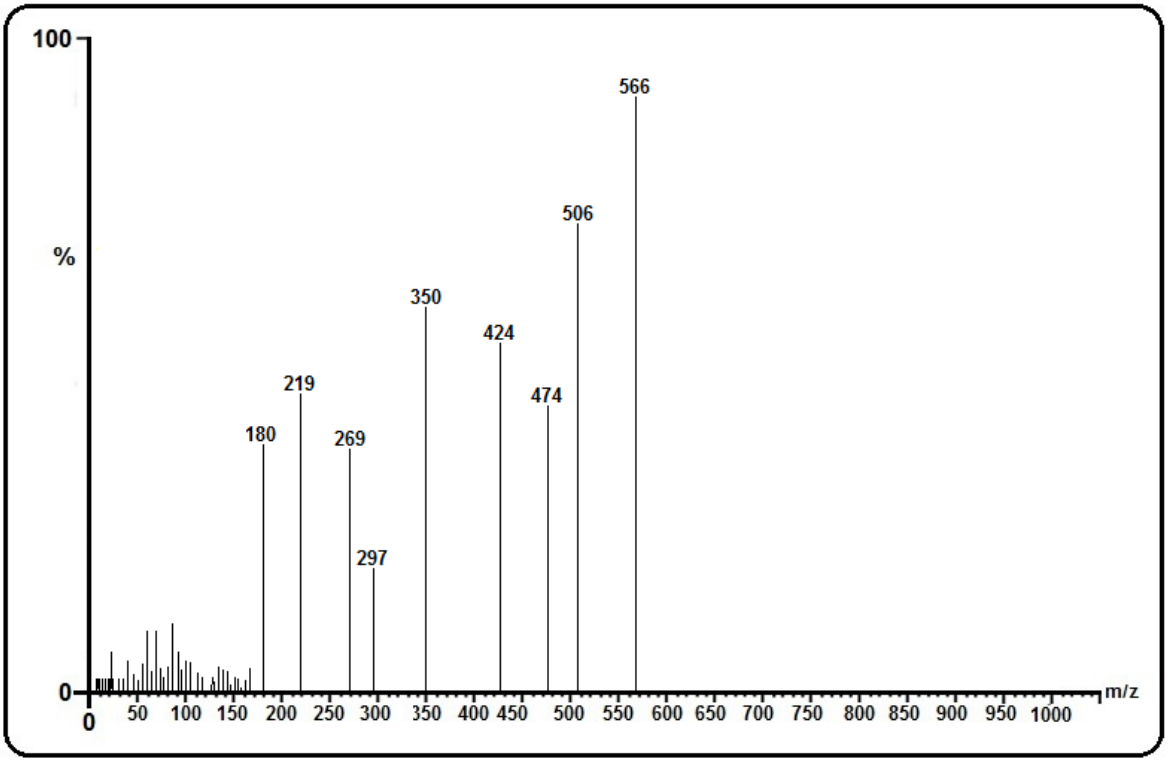


Figure S3: Mass spectrum of Ni(II) complex.

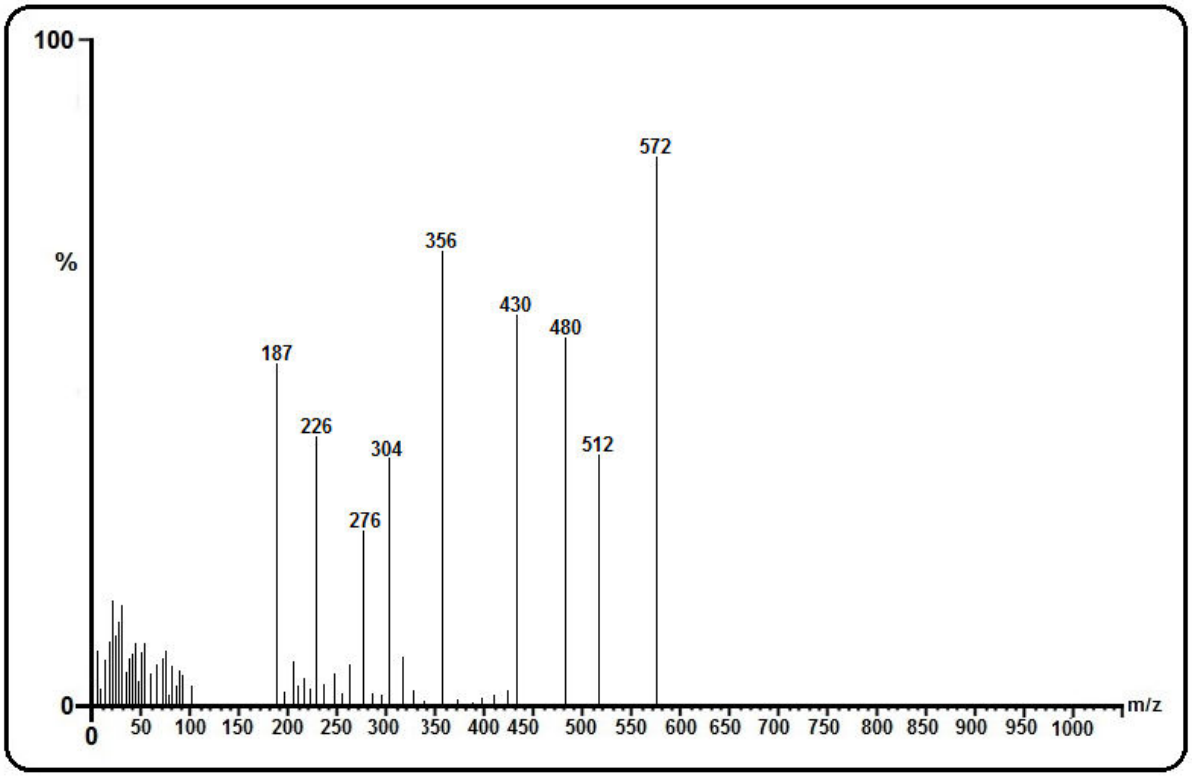


Figure S4: Mass spectrum of Zn(II) complex.

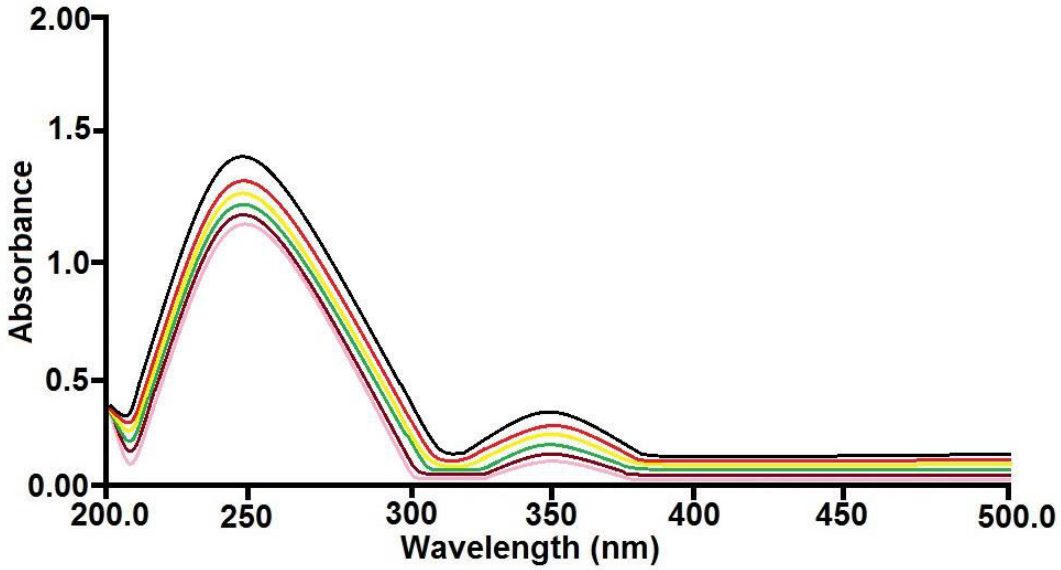


Figure S5. **The absorption spectra of complex Ni(II)(1 x 10^{-5} M) in the absence and presence of increasing amounts of CT-DNA (0-2.5 x 10^{-3} M) at normal temperature in 50 mM Tris-HCl / NaCl buffer (pH = 7.5).**

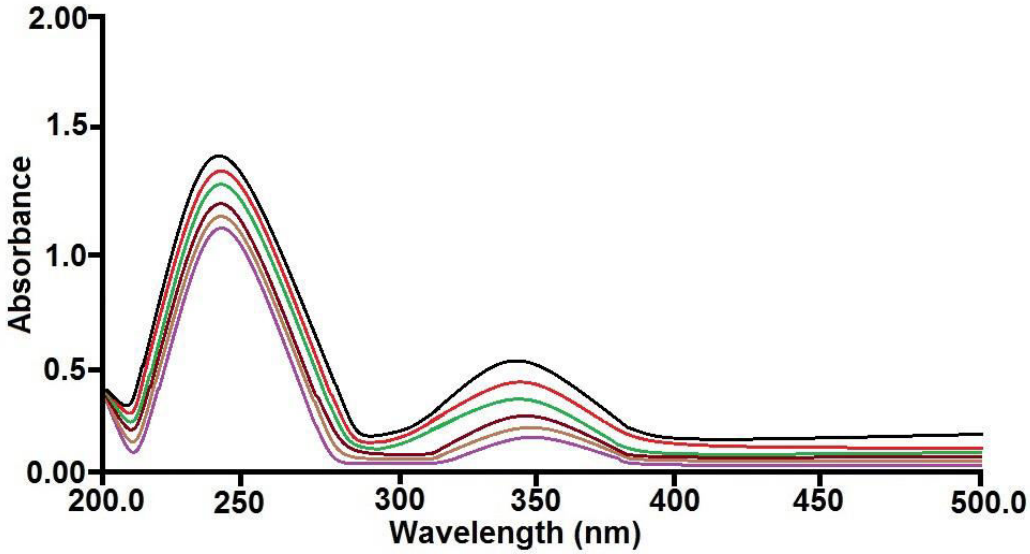


Figure S6 **The absorption spectra of complex Zn(II)(1 x 10^{-5} M) in the absence and presence of increasing amounts of CT-DNA (0-2.5 x 10^{3} M) at normal temperature in 50 mM Tris-HCl / NaCl buffer (pH = 7.5).**

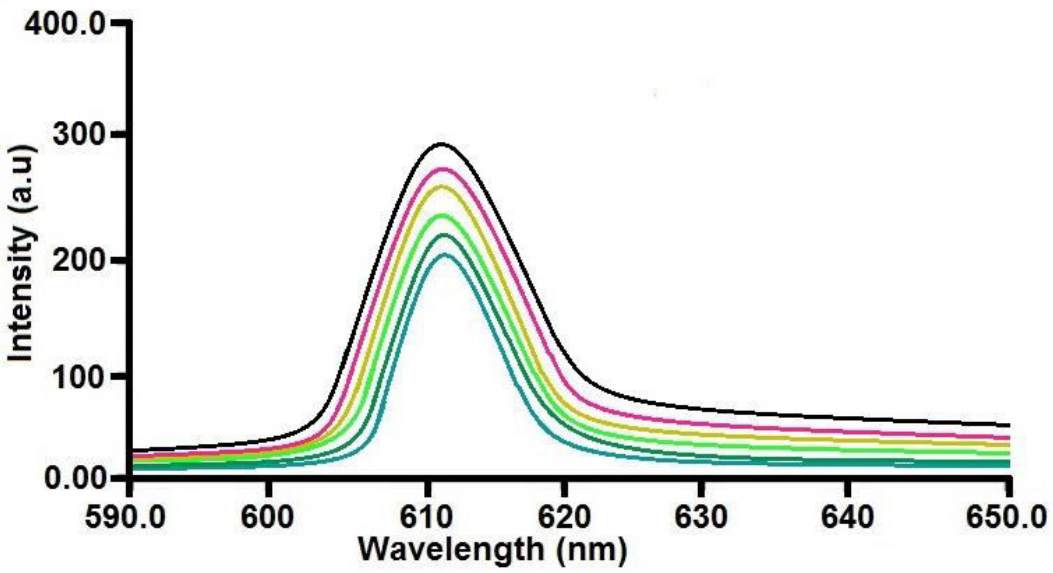


Figure S7 **Emission spectrum of EB bound to DNA in the presence of Ni(II):([EB] =3.3 µM,[DNA] = 40 µM, [complex] = 0-25 µM, λex= 440 nm).**

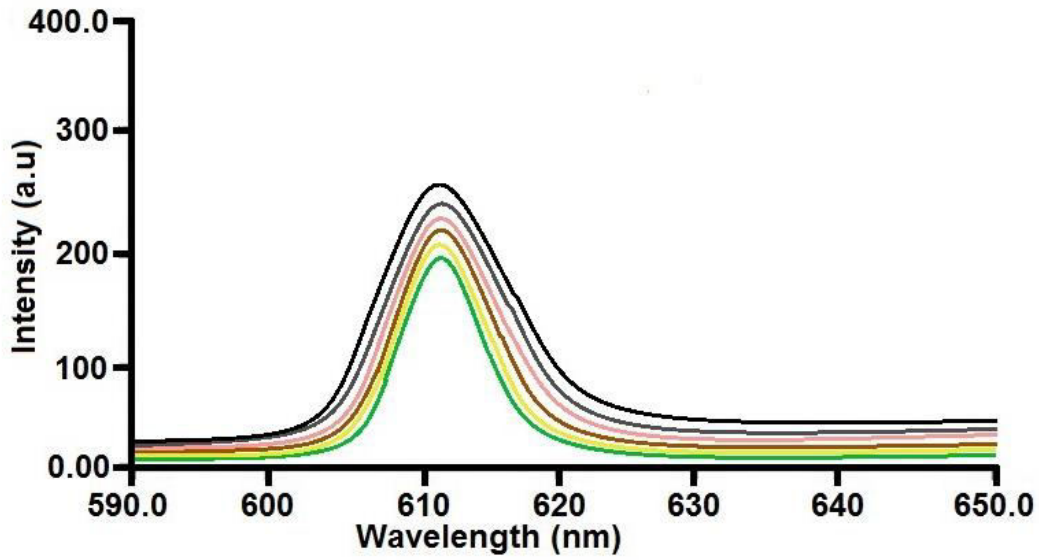


Figure S8 **Emission spectrum of EB bound to DNA in the presence of Zn(II):([EB] =3.3 µM,[DNA] = 40 µM, [complex] = 0-25 µM, λex= 440 nm).**