SYNTHESIS CHARACTERIZATION AND ANTIMICROBIAL ACTIVITY STUDIES OF SOME TRANSITION METAL COMPLEXES DERIVED FROM 5-CHLORO-3-PHENYL-N^1-((2-THIOXO-1,2-DIHYDROQUINOLINE-3-YL)METHYLENE)-1*H*-INDOLE-2-CARBOHYDRAZIDE

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Abstract
A series of new coordination complexes of Fe(III), Co(II), Ni(II), Zn(II) and Cd(II), with a new Schiff base 5-chloro-3-phenyl-N^1-((2-thioxo-1,2-dihydroquinoline-3-yl) methylene)-1*H*-indole-2-carbohydrazide have been synthesized and characterized by elemental analysis, UV-Visible, IR spectra, ^{1}HNMR spectra, mass spectra, powder X-ray diffraction data, molar conductance, Magnetic susceptibility, ESR and TGA. The new Schiff base has been synthesized by the reaction between 5-chloro-3-phenyl-1*H*-indole-2-carboxyhydrazide and 2-thioxo-1, 2-dihydroquinoline-3-carbaldehyde. The Schiff base behaves as tridentate ONS donor ligand and forms the complexes of the type ML_2 stoichiometry in case of Fe(III), Co(II), Ni(II), Zn(II) and Cd(II). All the complexes have exhibited octahedral geometry. The ligand and its metal complexes have been screened for their antibacterial activity against *Staphylococcus aureus* and *Pseudomonous aeruginosa*, antifungal activity against *Aspergillus niger* and *Aspergillus flavus* by cup plate method respectively and antioxidant activity by free radical scavenging activity using 1, 1- diphenyl-2-picryl hydrazyl (DPPH), with standard drugs vitamin-C and vitamin-E and DNA cleavage activity. Our aim is to investigate active antioxidant compounds from ligand and its complexes and study their antimicrobial and DNA cleavage activities. From this study our newly synthesized ligand and some of its complexes have exhibited good antimicrobial, antioxidant and DNA cleavage activities.
Keywords: Schiff base, Metal complexes, antimicrobial activity, antioxidant activity and DNA cleavage activity.

1. INTRODUCTION

The chemistry of coordination compounds with heterocyclic ligands containing oxygen and nitrogen as donor atoms has attracted the attention of chemists in recent years. It is well known that such ligands coordinate to metal atom in different ways in different media. Transition metal ions are essential in many biological systems in nature.[1] These metal complexes with bidentate and tetra dentate ligands containing both hard and soft donor groups have been used extensively in coordination and organometallic chemistry.[2] The chelating properties of Schiff base display manifold applications in medicine, industry and agriculture.[3]

The indole moiety represents an important structural component associated with a variety of alkaloids[4, 5] and molecules having wide ranging biological activities, such as antiviral,[6-10] antitumor,[11, 12] antimicrobial activities,[13, 14] antituberculosis[15] and antifungal activities.[16] Many quinoline derivatives have been reported to possess antimicrobial[17] and anti-inflammatory[18] activities.

Quinoline derivatives have been reported to possesses anti-inflammatory,[19] antibacterial,[20-22] antifungal,[23, 24] antiallergy,[25] antidepressant,[26] antiasthmatic,[27] antimalarial,[28-30] antiviral,[31, 32] antitumor,[33] neuroleptic,[34] antihypertensive,[35, 36] cytotoxic,[37-39] antihistamine,[40] CVS,[41] antiseptic, analgesic, anthelmintic,[42] hypnotic and sedative[43] beside wide range of other pharmacological activities.

In view of these findings, we have synthesized some metal complexes with a new Schiff base 5-chloro-3-phenyl-N^1-((2-thioxo-1,2-dihydroquinoline-3-yl)methylene)-1*H*-indole-2-carbohydrazide which was prepared by the reaction between 5-chloro-3-phenyl-1*H*-indole-2-carboxyhydrazide and 2-thioxo-1,2-dihydroquinoline-3-carbaldehyde.

2. EXPERIMENTAL

2.1. General Remarks

All solvents and reagents were used as obtained from commercial sources with further purification according to standard procedures.[44] Melting points were determined in open glass capillary tubes. Purity of the compounds was checked on TLC. The IR spectra of compounds were recorded on Perkin-Elmer spectrum one spectrophotometer using KBr disc technique. ^{1}H NMR spectra were recorded on Bruker Avance 400 MHz instrument in d_6-DMSO using TMS as an internal standard and mass spectra on a JEOL GC mate and Agilent 6330 ion trap mass spectrophotometer. Satisfactory C, H, N analysis was recorded for all the compounds.

Compounds 2-chloro-3-formylquinolines, 2-thioxo-1, 2-dihydroquinoline-3-carbaldehyde and 5-chloro-3-phenyl-1*H*-indole-2-carboxyhydrazides[45-47] were prepared according to reported methods.

2.2. Synthesis of ligand HL

An equimolar mixture of 5-chloro-3-phenyl-1*H*-indole-2-carboxyhydrazide (0.285 g, 0.001 mol) and 2-thioxo-1, 2-dihydroquinoline-3-carbaldehyde (0.189 g, 0.001mol) in ethanol (30 mL) were refluxed in presence of catalytic amount of glacial acetic acid (1-2 drops) for about 6-7 h on water bath. The reaction mixture was cooled to room temperature; the separated Schiff base was collected by filtration, washed with ethanol, dried and recrystallized from 1, 4-dioxane Scheme 1.

2.3. Preparation of complexes of Fe(III), Co(II), Ni(II), Zn(II) and Cd(II) with Schiff base 5-chloro-3-phenyl-N^1-((2-thioxo-1,2-dihydroquinoline-3-yl)methylene)-1*H*-indole-2-carbohydrazide

To a hot solution of 5-chloro-3-phenyl-N^1-((2-thioxo-1,2-dihydroquinoline-3-yl) methylene)-1*H*-indole-2-carbohydrazide (0.912 g, 0.002 mol) in ethanol (30 mL) was added a hot ethanolic solution (10 mL) of respective metal chlorides (0.002 mol). The reaction mixture was refluxed on a steam bath for 4 h, and then sodium acetate (0.5 g) was added to it and refluxed for 2 h. It was then poured in to distilled water. The resulting solid complexes were collected by filtration, washed with sufficient quantity of distilled water, then with hot ethanol to apparent dryness and dried in a vacuum over anhydrous calcium chloride in a desiccator (yield 61-80%) table 1.

2.4. Antimicrobial activity

Antimicrobial activity was carried out by the cup-plate method[48]. The *in vitro* antibacterial screening of ligand and its complexes was undertaken against *S. aureus* and *P. aeruginosa* by cup-plate method using nutrient agar media. In brief, molten agar nutrient kept at 45 $^{\circ}$C was poured into petri dishes and allowed to solidify. Then wells of 4 mm diameter were punched carefully using a sterile cork borer and were filled with test solution 25 µL (1000 ppm, 500 ppm, 250 ppm, 125 ppm). The plates were incubated for 24 h at 37 $^{\circ}$C. The diameter of the zone of inhibition for all the test compounds was measured and the results were compared

with the standard drug Gentamycin of the same concentration of the test compound under identical conditions.

The antifungal activity of the test compounds were evaluated against *A. niger* and *A. flavus* by the cup plate method cultured on seberose dextrose agar (SDA) medium adapting the procedures as described above. The plates were incubated at 37 °C for 48 h. The diameter of the zone of inhibition for all the test compounds were compared with standard drug Fluconazole of the same concentration as that of the test compounds under identical conditions.

Since all the test compounds and standard drugs were prepared in freshly distilled DMF, its zone of inhibition was found to be negligible and taken as 0 mm.

2.5. Antioxidant activity by DPPH radical scavenging activity

1, 1- Diphenyl-2-picryl hydrazyl (DPPH) radical scavenging activity was measured by spectrophotometric method at 517nm.[49-52] to a methanolic solution of DPPH (0.1 mmol) and standard compounds Vit-C and Vit-E were added separately in different concentrations and an equal amount of methanol (0.05 mL) was added control. After 30 min, absorbance was measured. The percentage of free radical scavenging was calculated by comparing the control and test samples with the fallowing equation.

$$Percentage\,of\ scavenging\ activity = \frac{A_o - A_e}{A_o} \times 100$$

Where A_o corresponds to the absorbance of DPPH without sample and A_e corresponds to the absorbance of sample with complex or ligand. A_o is the absorbance sample containing only DPPH (blank).

2.6. DNA cleavage activity

The degree to which the ligand and its complexes could function as DNA cleavage agents was examined using calf-thymus DNA (Cat. No.105850) as a target, the efficiency of cleavage of these molecules was studied by using agarose gel electrophoresis. Nutrient broth media was used (Peptone 10, NaCl 10 and yeast extract 5 g L^{-1}). The electrophoresis of the samples was done according to the fallowing procedure.[53]

Briefly, 250 mg of agarose was dissolved in 25 ml of tris- acetate-EDTA (TAE) buffer (4.84 g Tris base, pH 8.0, 0.5 M EDTA L^{-1}) by boiling, when the gel attains ~55^0C, it was poured into the gel cassette fitted with comb. The gel was allowed to solidify and then carefully the comb was removed. The gel placed in the electrophoresis chamber flooded with TAE buffer. Load 20 µl of DNA sample (mixed with bromophenol blue dye @ 1:1 ratio) was loaded carefully into the wells, along with standard DNA marker with the constant 50 V of electricity for 45 min. Later gel was removed carefully and stained with ethidium bromide (ETBR) solution (10 µg mL^{-1}) for 10-15 min and the bands were observed under UV gel documentation system.

3. RESULT AND DISCUSSION

3.1. IR, ^{1}H NMR and mass spectra of ligand HL

The IR spectrum of ligand showed peaks at 1655 cm^{-1} due to carbonyl group and the three absorption band at 3288, 3129 and 3088 cm^{-1} due to indole NH, amide NH and NH of

quinolone moiety respectively. The bands observed at 1579 and 1181 cm^{-1} are due to azomethine C=N group and C=S function at 2-position of quinoline moiety respectively.

The ^{1}HNMR spectra of the Schiff base HL in d_6-DMSO at room temperature, showed signal at δ13.9 (s, 1H, quinoline NH), δ12.1 (s, 1H, CONH), δ11.8 (s, 1H,indole NH), δ8.94 (s,1H, N=CH) and δ7.30-8.47 (m,13H,ArH) due to protons of quinolone NH, amide NH, azomethine NH and aromatic protons respectively.

The mass spectrum of ligand HL showed the molecular ion peak at M+1=457, 459 (12%, 3%). This on loss of hydrogen radical gave a fragment ion recorded at m/z 456, 458 (12%, 4%). This on loss of C_9H_5NS species gave a fragment ion recorded at m/z 297, 299 (5%, 2%) which on simultaneous expulsion of methyl, chloride, hydrogen, hydrogen and hydrogen radicals gave a fragment ion recorded at m/z 244 (100%) which is also a base peak. This on simultaneous loss of nitrogen and carbon monoxide molecules gave another fragment ion record at m/z 188 (30%). The molecular ion in another route has also lost $C_{10}H_8N_2S$ species and gave a fragment ion recorded at m/z 268, 270 (33%, 12%) The fragmentation pattern is depicted in Scheme 3. The above fragmentation pattern is in consistency with its structure. The IR, ^{1}HNMR and mass spectral data of ligand HL confirms its formation by the reaction between 5-substituted-3-phenyl-1*H*-indole-2-carboxyhydrazide and 2-thioxo-1,2-dihydroquinoline-3-carbaldehyde.

3.2. IR and mass data of the complexes of ligand HL

The IR spectrum of ligand was compared with those of the complexes of metal ions Fe(III), Co(II), Ni(II), Zn(II) and Cd(II) in order to study the binding mode of Schiff base to the metal ions. In the IR spectra of ligand the band appeared at 1655 cm^{-1} assigned to carbonyl group (C=O) has disappeared in its complexes the above metal ions suggesting the involvement of carbonyl oxygen atom in coordination by the deprotonation after its enolization. All the complexes except Zn(II) complex of the ligand under the present study displayed broad bands in the region 3432-3418 cm^{-1} indicating the presence of lattice or coordinated water molecules. The complexes showed an intense peak in the region 3300-3225 cm^{-1} indicating the non-involvement of indole NH in complexation. The peak observed at 3088 cm^{-1} due to quinolone NH has appeared at about the same region 3100-3053 cm^{-1} in all the complexes indicates the non-involvement of quinolone NH in complexation. The disappearance of a band at 3129 cm^{-1} due to amide NH and appearance of a new band in the region 1603-1590 cm^{-1} due to -C=N-N=C- function in all the complexes further confirms the enolization of O=CNH attached to 2-position of indole during complexation and its deprotonation during complexation. Absorption band at 1579 cm^{-1} in case of ligand due to C=N of azomethine function has been found to be shifted towards the lower frequency side 1555-1536 cm^{-1} in all the complexes indicates the metal ions have coordinated to azomethine nitrogen. A band at 1181 cm^{-1} due to C=S in case of ligand has shifted to lower frequency 1179-1157 cm^{-1} in case of all the complexes indicating the coordination of metal ions with sulfur of C=S function. The IR regions are purely tentative because of various skeletal vibrations associated with metal and ligand vibrations. The bands of weak intensity in the region 563-532 cm^{-1} in case of all the complexes of the ligand HL are assigned M-O vibrations and the bands in the region 469-429 cm^{-1} to M-N vibrations Table 2.

In mass spectrum of Fe(III) complex, molecular ion has not been observed because HCl and H_2O species have expelled out from the molecular ion before it has reached the detector. Therefore the mass spectrum of Fe(III) complex of ligand HL displayed a peak at m/z 964.8, 966.8 (100%, 33%) which is also a base peak. This on further loss of $C_{14}H_9NCl$ radical, CO,

N_2, $C_{10}H_6NS$ radical, C_6H_4N radical, H_2S and H species simultaneously gave fragment ion peak at m/z 385.85, 387.85 (2%, 0.7%). This fragmentation pattern is in agreement with the structure of complex and is depicted in Scheme 4.

3.3. Electronic spectra of Fe(III), Co(II) and Ni(II) complexes of the ligand HL

Electronic spectral data of the Fe(III), Co(II) and Ni(II) complexes of the ligand HL are given in Table-3. Electronic spectral studies of all these complexes were carried out in DMF at 10^{-3} M concentration.

3.3.1. Fe(III) complex

The ground state of high spin octahedral coordinated Fe(III) complex is $^6A_{1g}$. The four lowest energy bands are due to the transition $^6A_{1g}$ to $^4T_{1g}$, $^4T_{2g}$, ^4E_g and $^4A_{1g}$ excited states. For high spin Fe(III) complex three spin allowed transitions are reported[53] around 16800 cm^{-1} (ν_1), 20500 cm^{-1} (ν_2) and 25900 cm^{-1} (ν_3) which are characteristics of octahedral geometry. The iron complex of the ligand HL in the present study displayed three bands at 17090 cm^{-1}, 19685 cm^{-1} and 26635 cm^{-1} which corresponds to ν_1, ν_2 and ν_3 transition respectively. These observed values for Fe(III) complex in its visible spectrum are in agreement with the literature value[54] and there by proved octahedral geometry for the Fe(III) complex of the ligand HL.

3.3.2. Co(II) complex

Cobalt(II) is d^7 ion exists both in octahedral and tetrahedral geometry. In octahedral Co(II) complexes three spin allowed transitions are expected corresponding to the transitions $^4T_{1g}(F) \rightarrow {}^4T_{2g}(F)$ (ν_1) (~ 8000 cm^{-1}), $^4T_{1g}(F) \rightarrow {}^4A_{2g}(F)$ (ν_2) (~ 16000 cm^{-1}) and $^4T_{1g}(F) \rightarrow {}^4T_{2g}(P)$ (ν_3) (~ 20000 cm^{-1}).

Chandra *et al*[55] have reported three bands corresponding to ν_1, ν_2 and ν_3 transition around 9000 cm^{-1}, 14500 cm^{-1}, 20620 cm^{-1} respectively for octahedral Co(II) complex. The Co(II) complex of the ligand HL under present study has showed three bands at 10772 cm^{-1}, 15961 cm^{-1} and 19511 cm^{-1} due to $^4T_{1g}(F) \rightarrow {}^4T_{2g}(F)$ (ν_1), $^4T_{1g}(F) \rightarrow {}^4A_{2g}(F)$ (ν_2) and $^4T_{1g}(F) \rightarrow {}^4T_{2g}(P)$ (ν_3) transition respectively. These transitions suggests octahedral geometry for Co(II) complex. The broad nature of ν_1 band which may be best assigned to the envelope of the transitions from $^4E_g(^4T_{2g})$ to the component $^4B_{2g}$ and ^4E_g of $^4T_{2g}$ which is the characteristic of tetragonally distorted octahedral geometry.[56]

3.3.3. Ni(II) complex

The ground state of Ni(II) in octahedral co-ordination is $^3A_{2g}(t_{2g}{}^6eg^2)$ Ni(II) complexes shows three transitions in an octahedral field viz., $^3A_{2g}(F) \rightarrow {}^3T_{2g}(F)$ (ν_1) (7000-13000 cm^{-1}), $^3A_{2g}(F) \rightarrow {}^3T_{1g}(F)$ (ν_2) (11000- 20000 cm^{-1}) and $^3A_{2g}(F) \rightarrow {}^3T_{1g}(P)$ (ν_3) (20000-27000 cm^{-1}).

Figgis[57] has reported bands in the region ~ 10000, ~12000 and ~ 25000 cm^{-1} due to $^3A_{2g}(F) \rightarrow {}^3T_{1g}(P)$ (ν_3), $^3A_{2g}(F) \rightarrow {}^3T_{1g}(F)$ (ν_2) and $^3A_{2g}(F) \rightarrow {}^3T_{2g}(F)$ (ν_1) for the transitions mentioned above which are characteristics of octahedral geometry.[58]

The electronic spectrum of Ni(II) complex of the ligand HL under present investigation exhibited three bands in the region 10590 cm^{-1}, 16598 cm^{-1} and 25617 cm^{-1} which are assigned to $^3A_{2g}(F) \rightarrow {}^3T_{2g}(F)$ (ν_1), $^3A_{2g}(F) \rightarrow {}^3T_{1g}(F)$ (ν_2) and $^3A_{2g}(F) \rightarrow {}^3T_{1g}(P)$ (ν_3) transition. All these observations favor the octahedral geometry for Ni(II) complex of the present study.

3.4. Magnetic susceptibility data

Magnetic susceptibility measurements of the complexes were performed at room temperature. The magnetic moment values for the various Co(II) complexes is in the range 4.70-5.20 BM is for octahedral complexes. In the present investigation the observed magnetic moment value for Co(II) complex is 5.01 BM indicates octahedral geometry for the Co(II) complex.[54] For Ni(II) complex the observed magnetic moment value is 2.90 BM which is well within the expected range for Ni(II) complex with octahedral stereochemistry 2.83-4.00 BM.[55] For Fe(III) complexes the observed magnetic moment value is 5.91 BM respectively which corresponds to high spin octahedral complex[59] Table 1.

3.5. Molar Conductance

The molar conductance of the complexes was measured in DMF at 10^{-3}M concentration. Measured conductance values of these complexes are too low to account for their electrolytic behavior except Fe(III) complex which is week electrolyte Table 1.

3.6. Powder X-ray diffraction (XRD) studies

The compounds were soluble in polar organic solvents (DMSO & DMF). We did not obtain crystal suitable for single crystal studies. In order to test the degree of crystallinity of the synthesized complexes, we obtained the powder XRD pattern of Fe(III) and Ni(II) complexes.

The powder XRD pattern of Fe(III) and Ni(II) complexes have provided the supplementary material. The Fe(III) complex showed six reflections in the range 20-80° (2θ), arising the diffractions of X-ray by the planes of complex. All important peaks have been indexed and observed values of inter–planar distance (d) have been compared with the calculated ones. The unit cell calculations were performed for cubic system and the (h^2+ k^2+ l^2) values were determined. The absence of forbidden numbers (7, 15 and 23) indicates that the Fe(III) complex has cubic symmetry. The experimental values are in good agreements with (h^2+ k^2+ l^2) values of primitive type cubic cell with lattice parameter equal to $a = b = c = 3.66$ Å Table 4**.**

Similar calculations were performed for Ni(II) complex and this complex showed fifteen reflection in the range 0-80° (2θ). The experimental values are in good agreements with (h^2+ k^2+ l^2) values of primitive type cubic cell with lattice parameter equal to $a = b = c = 8.37$ Å Table- 5.

3.7. ESR spectrum of Fe(III) complex of ligand HL

In order to obtain more information about the environment of Fe(III) complex, the X- band ESR spectrum of the polycrystalline Fe(III) complex has been recorded at room temperature. The value of $g_\perp$ and $g_\parallel$ were calculated and presented in the Table 3. The observed ESR spectrum is characteristics of octahedral geometry with rhombic distortion. The 'g' value averaged to overall distortion where calculated using the relation

$$g_{av}^{\ 2} = \frac{1}{3}(2g_{\square}^{\ 2} + g_{\perp}^{\ 2})$$

G = ($g_\parallel$ - 2) / ($g_\perp$ - 2) which measures the exchange interaction between iron centers in a polycrystalline solid has been calculated. According to Hathaway[60], if the G value is greater than 4, the exchange interaction is negligible, while a value of G less than 4 indicates a

considerable exchange interaction between metal ions in the solid complex. In the present case, G =10 indicates that there is no exchange interaction in the Fe(III) complex of ligand HL.

The observed $g_{\parallel}$ =2.250, $g_{\perp}$= 2.025 values of the Fe(III) complex under the present study followed the same trend $g_{\parallel} > g_{\perp} > g_e$ which suggest the presence of unpaired electron in d_{x2-y2} orbital giving octahedral geometry.[61] The observed G=10 for the complexes under present study evidenced the monomeric nature of the complexes.[62] This fact is further supported by the absence of a band corresponding to ΔMs = ±2 transition in the observed ESR spectrum which is characteristic of monomeric complex.

$$G = \frac{(G \parallel -2)}{(G \perp -2)} = 10$$

3.8. Thermal studies

From TG curve, information related to the thermal stabilities, composition of the initial sample, intermediate compounds that formed and the final residue could be obtained.

The TGA study on $Fe[C_{50}H_{32}N_8O_2S_2Cl_2]Cl.H_2O$ and $Ni[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$ were carried out in the temperature range 35 °C to 1000 °C.

3.8.1. Fe(III) complex

The decomposition studies of the Fe(III) complex, $Fe[C_{50}H_{32}N_8O_2S_2Cl_2]Cl.H_2O$ has been carried out. In the thermogram of the $Fe[C_{50}H_{32}N_8O_2S_2Cl_2]Cl.H_2O$, the first stage of the decomposition represents the weight loss of HCl at 290 °C, with weight loss of 3.8%. The theoretical weight loss for this decomposition was 3.5% agreeing with observed value of 3.8%. The complex underwent further degradation and gave break at 360 °C with a weight loss of 12.85%, which corresponds to the decomposition of the H_2O and C_7H_5N species. This practical weight loss 12.85% is in accordance with theoretical weight loss of 12.31%. The third stage of decomposition occurs at 505 °C, with weight loss of 52.65% which corresponds to the decomposition of $C_{26}H_{15}N_4SCl$ species. This practical weight loss 52.65% is in accordance with theoretical weight loss of 52.21%. Thereafter compound showed a gradual decomposition up to 1000 °C and onwards. The weight of residue corresponds to iron oxide. The thermal decomposition of $Fe[C_{50}H_{32}N_8O_2S_2Cl_2]Cl.H_2O$ with probable assignments are given in Table 6.

3.8.2 Ni(II) complex

The decomposition studies of the Ni(II) complex $Ni[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$ has been carried out. In the thermogram of the $Ni[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$, the loss of H_2O observed at 285 °C, with weight loss of 2%. This practical weight loss of 2% is in accordance with the theoretical weight loss 1.82%. The resultant intermediate complex underwent further degradation and gave another break at 385 °C with a weight loss of 4.88%, which corresponds to the elimination of the SO species from the above intermediate complexes. This theoretical weight loss corresponds to 4.88% is agreeing well with the observed value 4.95%. The third stage of decomposition occurs at 740 °C, with weight loss of 66.06% which corresponds to the decomposition of $C_{35}H_{23}N_6OSCl$ species. This practical weight loss 66.06% is in accordance with theoretical weight loss of 66.25%. Thereafter compound showed a gradual decomposition up to 1000 °C and onwards. The weight of residue corresponds to nickel

oxide. The thermal decomposition of $Ni[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$ with probable assignments is given in Table 7.

3.9. Antimicrobial activity

The antibacterial activity of all the newly synthesized ligand and its complexes were studied in a series of dilutions (1000 ppm, 500 ppm, 250 ppm and 125 ppm) by cup-plate method using nutrient agar media. Ligand **HL** and **Ni(II)** complex exhibited high activity towards *S. aureus in vitro* tests at minimum inhibitory concentration of 125 ppm. **Fe(III)**, **Co(II)** and **Cd(II)** complexes were active at minimum inhibitory concentrations of 250 ppm against *S. aureus* when compared with standard drug Gentamycin at 125 ppm. **Fe(III)** and **Cd(II)** complexes exhibited high activity towards *P. aeruginosa in vitro* tests at minimum inhibitory concentration of 125 ppm. Ligand **HL**, **Ni(II)** and **Zn(II)** complexes were active at minimum inhibitory concentrations of 250 ppm against *P. aeruginosa*. **Co(II)** showed less activity at 500 ppm when compared with standard drug Gentamycin 125 ppm Table 8.

The antifungal activity of the ligand HL and its complexes was studied in a series of dilutions (1000 ppm, 500 ppm, 250 ppm and 125 ppm) by seberose dextrose agar (SDA) method. **Ni(II)** complex exhibited high activity towards *A. niger in vitro* tests at minimum inhibitory concentration of 125 ppm. **Co(II)**, **Zn(II)** and **Cd(II)** complexes were moderately active at minimum inhibitory concentrations of 250 ppm against *A. niger*. Ligand **HL** and **Fe(III)** complex showed less activity at 500 ppm when compared with standard drug Fluconazole at 125 ppm.

Ligand **HL, Fe(III), Co(II)** and **Cd(II)** complexes exhibited high activity against *A. flavus* at minimum inhibitory concentration of 125 ppm. **Ni(II)** and **Zn(II)** complexes were moderately active at minimum inhibitory concentrations of 250 ppm against *A. flavus* when compared with standard drug Fluconazole at 125 ppm Table 9.

3.10. Antioxidant activity by DPPH radical scavenging activity

Antiradical activity evaluation for complexes and its ligand was measured in terms of decreases in absorbance at 517 nm of DPPH methanol solution (0.1 mmol) produced by the effect of each complex or ligand as a result of their ability to donate a hydrogen giving place to the reduced form of DPPH radical. The amount of DPPH reduced form was obtained in every established period of time using the fallowing equation.

$$Percentage\,of\ scavenging\ activity = \frac{A_o - A_e}{A_o} \times 100$$

Where A_o corresponds to the absorbance of DPPH without sample and A_e corresponds to the absorbance of sample with complex or ligand. A_o blank sample containing only DPPH; the negative control is methanol and standard Vit-C and Vit-E as a control.

DPPH scavenging activity of ligand and its complexes v/s vitamin-C and vitamin-E as standards were analyzed at 1mg/mL in DMF solution at T = 30 min. This investigation indicates that there is a great possibility of finding potent antioxidants. The ligand HL and its Zn(II) complex have exhibited very good free radical scavenging activity, Fe(III), Co(II), Ni(II) and Cd(II) complexes of ligand HL showed less activity compared with Vit-C and Vit-E. The bar graph representation of percentage of free radical scavenging activities is shown in Fig 1.

3.11. DNA cleavage activity

The calf-thymus (CT) DNA gel electrophoresis experiment was conducted at 37 ^{o}C using our newly synthesized ligand and its complexes, as can be seen from the result at 100 µg mL^{-1} concentration of all the samples showed complete cleavage of DNA as compared to control DNA figure 2.

4. CONCLUSION

A new ligand 5-chloro-3-phenyl-N^{1}-((2-thioxo-1,2-dihydroquinoline-3-yl) methylene)-1*H*-indole-2-carbohydrazide and its complexes have been synthesized and characterized by IR, ^{1}HNMR, mass, TGA, ESR and Powder-XRD data which concludes that all the complexes are of the type ML_2 stoichiometry and these complexes exhibited octahedral geometry. Some of these complexes have exhibited good antimicrobial, antioxidant and DNA cleavage activities.

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Table 1 Physical, Analytical, and Molar conductance data of ligand HL and its complex

Compounds	Molecular formula	Mol. Wt.	M.P ^{0}C (Yield in%)	Elemental analysis (%) Calcd (Found) M	C	H	N	Cl	Molar cond (λ_M) ohm^{-1} cm^2 mol^{-1}	μ eff (B.M)	Colour
*HL	$C_{25}H_{17}N_4OSCl$	456	315-317 (65)	-	65.78 (65.71)	3.72 (3.70)	12.28 (12.22)	7.67 (7.60)	18	-	Yellow
Fe-complex	$Fe[C_{50}H_{32}N_8O_2S_2Cl_2]Cl.H_2O$	1018.85	304-306 (80)	5.48 (5.59)	58.88 (58.92)	3.33 (3.25)	10.99 (10.91)	10.30 (10.21)	53	5.91	Brown
Co-complex	$Co[C_{50}H_{32}N_8O_2S_2Cl_2]2H_2O$	1004.93	330-332 (73)	5.86 (5.78)	59.70 (59.65)	3.58 (3.60)	11.14 (11.08)	6.96 (6.92)	37	5.01	Brown
Ni-complex	$Ni[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$	986.69	>360 (61)	5.94 (5.90)	60.80 (60.68)	3.44 (3.40)	11.35 (11.30)	7.09 (6.99)	10	2.90	Reddish brown
Zn-complex	$Zn[C_{50}H_{32}N_8O_2S_2Cl_2]$	975.39	324-326 (68)	6.70 (6.81)	61.51 (61.45)	3.28 (3.20)	11.48 (11.41)	7.17 (7.10)	14	-	Yellow
Cd-complex	$Cd[C_{50}H_{32}N_8O_2S_2Cl_2]H_2O$	1040.41	332-334 (78)	10.80 (10.65)	57.66 (57.65)	3.26 (3.15)	10.76 (10.80)	6.72 (6.65)	20	-	Light green

*HL - Ligand HL

Table 2 IR Data of ligand HL and its complex

Compounds	ν_{H_2O}	Indole NH	Amide NH	Quinoline NH	$\nu_{C=O}$	$\nu_{C=S}$ Quinoline	$\nu_{C=N}$ Azomethine	>C=N-N=C< azine	ν_{M-O}	ν_{M-N}
HL	-	3288	3129	3088	1655	1181	1579	-	-	-
Fe-complex	3425	3300	-	3053	-	1162	1549	1594	532	436
Co-complex	3418	3225	-	3053	-	1172	1547	1603	563	469
Ni-complex	3432	3247	-	3054	-	1157	1555	1595	542	462
Zn-complex	-	3280	-	3075	-	1179	1536	1600	537	429
Cd-complex	3425	3266	-	3100	-	1169	1537	1590	550	439

Table 3 Electronic and EPR spectral data of complex of the ligand HL

Compounds	Electronic spectral data (in cm^{-1})				E. S. R data			
	ν_1	ν_2	ν_3	ν_4	$g_\perp$	g_{II}	g_{av}	G
Fe-complex	16429	20524	25635	-	2.025	2.250	2.10	10.0
Co-complex	10772	15961	19511	-	-	-	-	-
Ni-complex	10590	16598	25617	-	-	-	-	-

Table 4 Powder X-ray data of Fe(III) complex of ligand HL

Peak	2Θ	Θ	SinΘ	$Sin^2\Theta$	h k l	d		$h^2+k^2+l^2$	a in A^o
						Calc.	Obser.		
1	24.270	12.135	0.2102	0.0441	1 0 0	3.6631	3.6642	1	3.66
2	27.560	13.78	0.2381	0.0566	1 0 0	3.2339	3.2338	1	3.65
3	27.730	13.865	0.2396	0.0574	1 0 0	3.2136	3.2144	1	3.66
4	63.360	31.68	0.5251	0.2757	2 1 1	1.4663	1.44667	6	3.66
5	73.930	36.965	0.6013	0.3615	2 2 0	1.2805	1.2810	8	3.66
6	74.340	37.17	0.6041	0.3649	2 2 0	1.2749	1.2749	8	3.66

Table 5 Powder X-Ray data of Ni(II) complex of ligand HL

Peak	2Θ	Θ	SinΘ	$Sin^2Θ$	h k l	D		$h^2+ k^2+ l^2$	a in A^o
						Calc.	Obser.		
1	10.560	5.28	0.092	0.0084	1 0 0	8.3695	8.3705	1	8.37
2	11.070	5.535	0.096	0.0092	1 0 0	8.0208	7.9860	1	8.33
3	11.840	5.92	0.103	0.0106	1 0 0	7.4757	7.4683	1	8.38
4	15.510	7.755	0.134	0.0179	1 1 0	5.7462	5.7084	2	8.33
5	15.840	7.92	0.137	0.0187	1 1 0	5.6204	5.5902	2	8.32
6	16.730	8.365	0.145	0.0210	1 1 0	5.3103	5.2948	2	8.37
7	17.150	8.575	0.149	0.0222	1 1 0	5.2380	5.1661	3	8.39
8	20.940	10.47	0.181	0.0327	2 0 0	4.2541	4.2388	4	8.36
9	21.280	10.64	0.184	0.0338	2 0 0	4.1847	4.1719	4	8.36
10	21.620	10.81	0.187	0.0349	2 0 0	4.1776	4.1070	4	8.36
11	22.520	11.26	0.195	0.0380	2 0 0	3.9487	3.9449	4	8.36
12	23.180	11.59	0.200	0.0400	2 1 0	3.8500	3.8340	5	8.38
13	23.860	11.93	0.206	0.0424	2 2 1	3.7378	3.7263	9	11.20
14	24.680	12.34	0.213	0.0453	2 1 0	3.6150	3.6043	5	8.36
15	25.160	12.58	0.217	0.0470	2 1 0	3.5483	3.5366	5	8.36

Table 6 Thermal decomposition of Fe(III) complex of ligand HL

Complex	Stage	Peak temp TG(^{0}C)	Loss of mass (in %)		Probable assignments
			Practical	Theoretical	
Fe-complex					$[C_{50}H_{34}N_8O_3S_2Cl_3]Fe$
					$\downarrow$ -HCl
	I	290	3.8	3.5	
					$[C_{50}H_{33}N_8O_3S_2Cl_2]Fe$
					$\downarrow$ $-H_2O$ $-C_7H_5N$
	II	360	12.85	12.31	
					$[C_{43}H_{26}N_7O_2S_2Cl_2]Fe$
					$\downarrow$ $-C_8H_5N$ -
	III	505	52.65	52.21	$C_{18}H_{10}N_3SCl$
					$[C_{17}H_{11}N_3O_2SCl]Fe$
					$\downarrow$
					Fe_2O_3

Table 7 Thermal decomposition of Ni(II) complex of ligand HL

Complex	Stage	Peak temp TG(^{0}C)	Loss of mass (in %)		Probable assignments
			Practical	Theoretical	
Ni-complex					$[C_{50}H_{34}N_8O_3S_2Cl_2]Ni$
					$\downarrow$ $-H_2O$
	I	285	2	1.82	
					$[C_{50}H_{32}N_8O_2S_2Cl_2]Ni$
	II	385	4.88	4.95	$\downarrow$ -SO
					$[C_{50}H_{32}N_8OSCl_2]Ni$
	III	740	66.06	66.25	$\downarrow$ -
					$C_{35}H_{23}N_6OSCl$
					$[C_{15}H_9N_2Cl]Ni$
					$\downarrow$
					NiO

Table 8 Antibacterial activity of ligand and its complexes

Compound	1000 ppm		500 ppm		250 ppm		125 ppm	
	S. aureus	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*	*S. aureus*	*P. aeruginosa*
HL	8	4	6	3	5	3	4	2
Fe-complex	6	5	5	5	4	3	2	3
Co-complex	3	4	3	4	3	2	2	2
Ni-complex	6	5	4	4	3	3	3	2
Zn-complex	5	5	4	3	3	3	2	2
Cd-complex	5	3	5	3	4	3	2	3
DMF(Control)	-	-	-	-	-	-	-	-
Gentamycin	8	6	8	6	7	5	5	4

Table 9 Antifungal activity of ligand and its complexes

Compound	1000 ppm		500 ppm		250 ppm		125 ppm	
	A. Niger	*A. Flavus*	*A. Niger*	*A. Flavus*	*A. Niger*	*A. Flavus*	*A. Niger*	*A. Flavus*
HL	6	8	6	6	4	6	4	6
Fe-complex	6	8	6	8	4	8	3	6
Co-complex	6	8	6	8	6	8	5	6
Ni-complex	12	6	10	6	8	6	6	4
Zn-complex	6	6	6	6	5	6	4	4
Cd-complex	6	6	5	6	5	6	4	6
DMF(Control)	-	-	-	-	-	-	-	-
Fluconazole	10	10	8	10	8	10	8	8

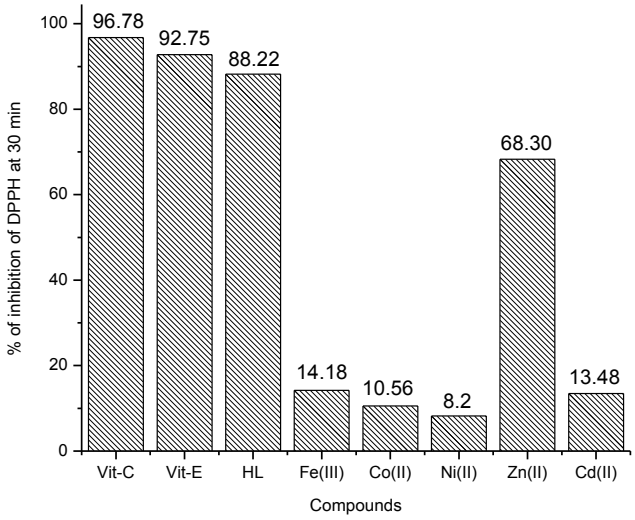


Fig.1 DPPH radical scavenging activity of ligand HL and its complexes.

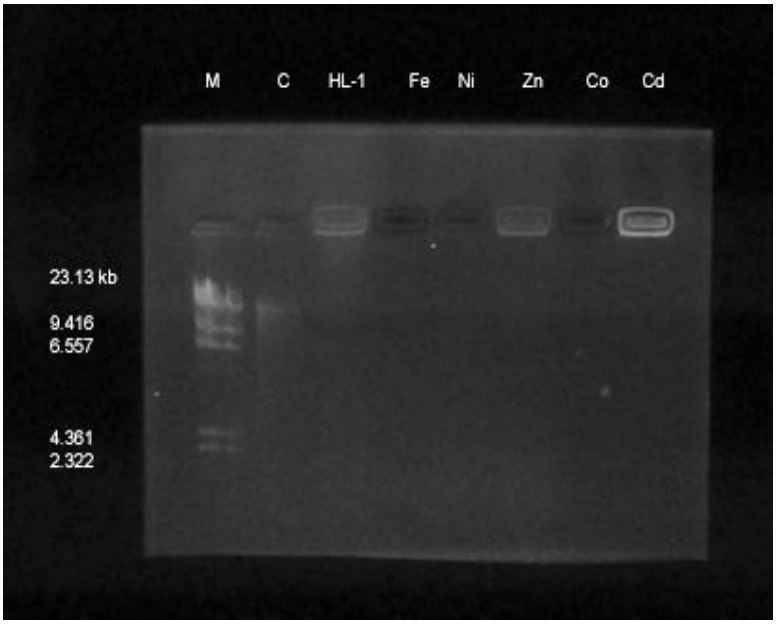
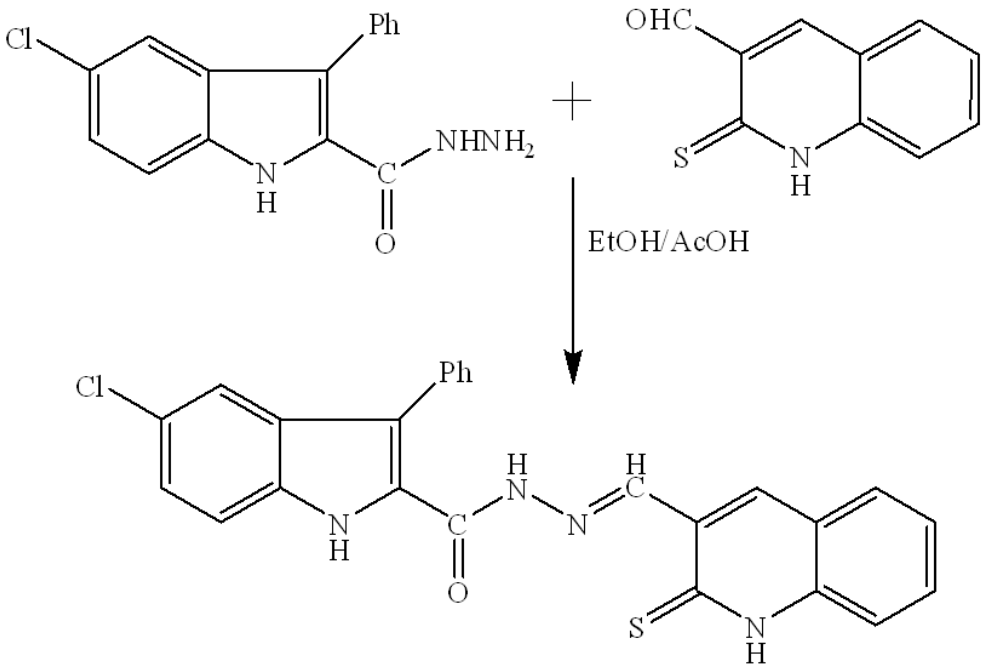
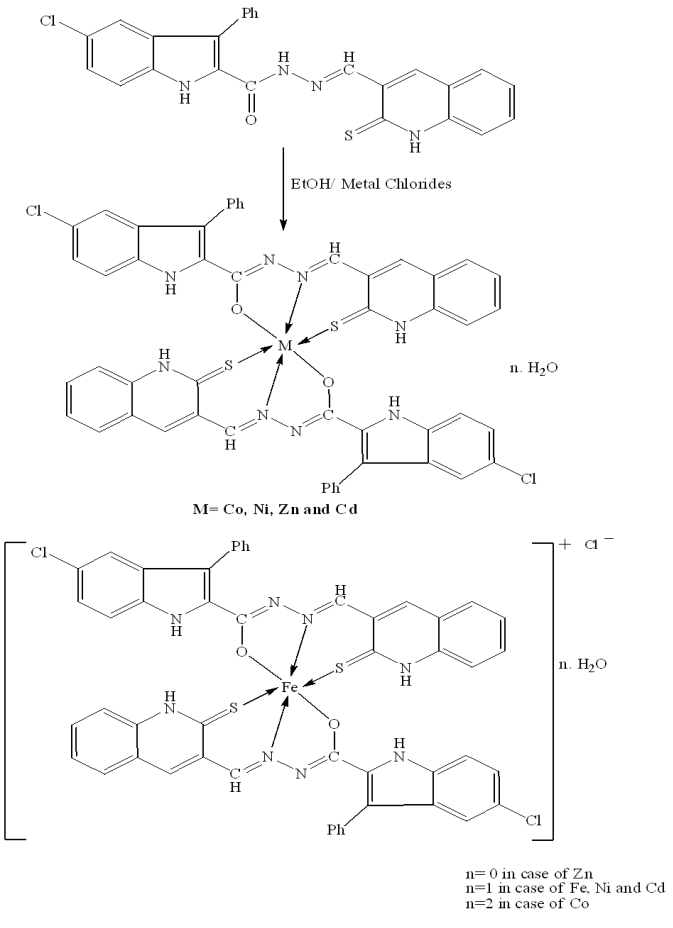


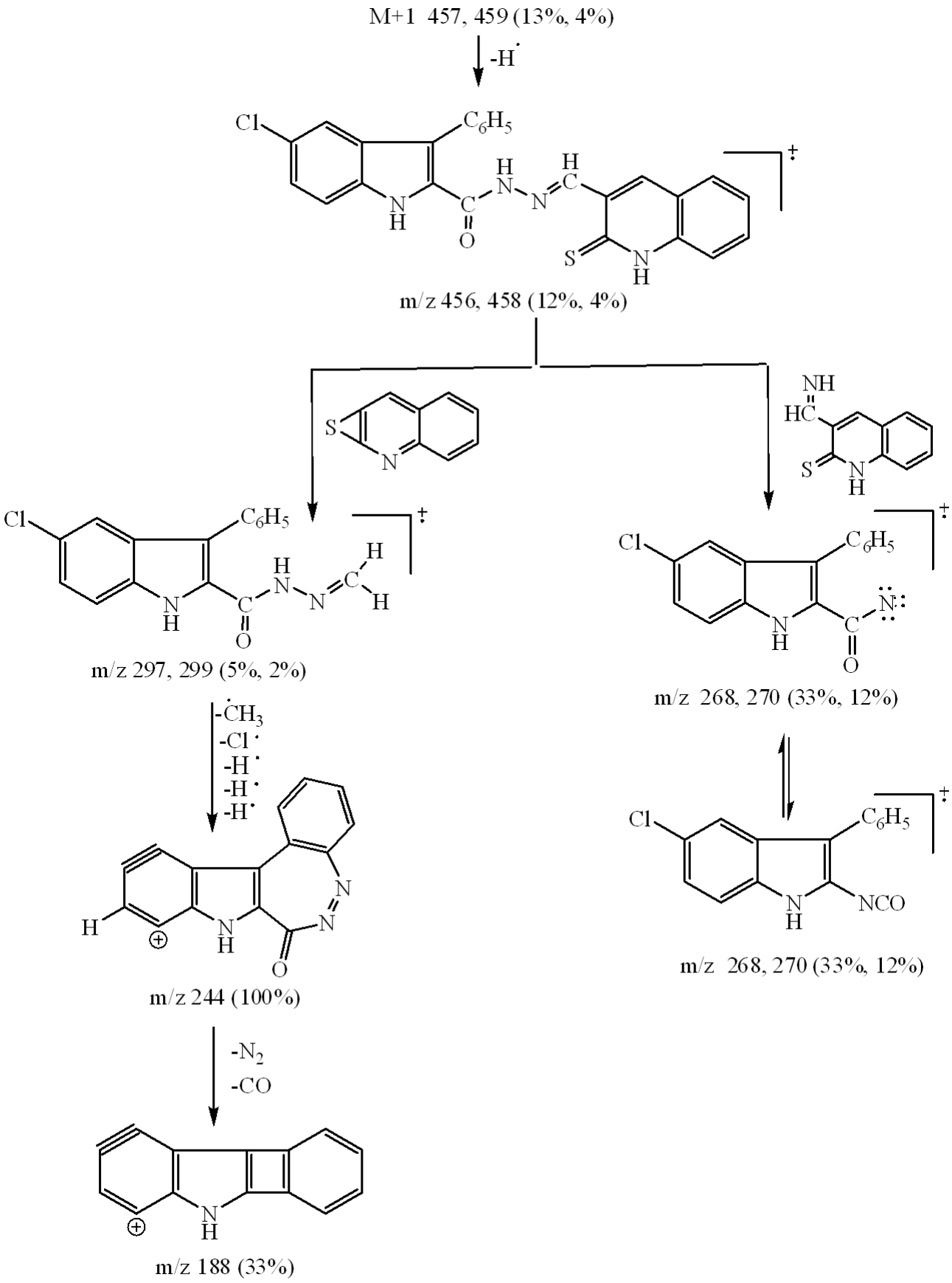
Fig. 2 DNA cleavage activity of ligand HL and its complexes.



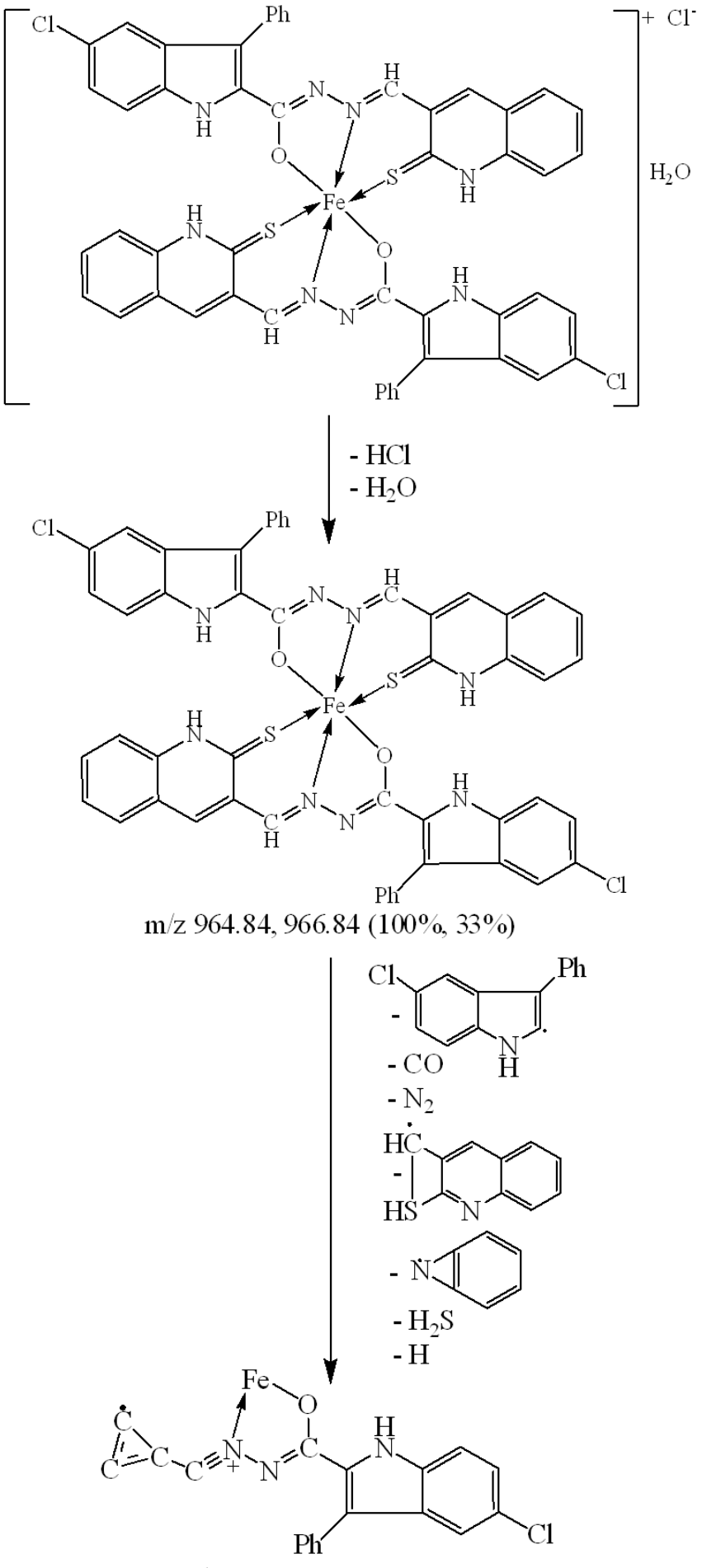
Scheme 1 synthesis of ligand



Scheme 2 Synthesis of metal complexes



Scheme 3 Mass fragmentation pattern of ligand



Scheme 4 Mass fragmentation of Fe(III) complex of ligand HL