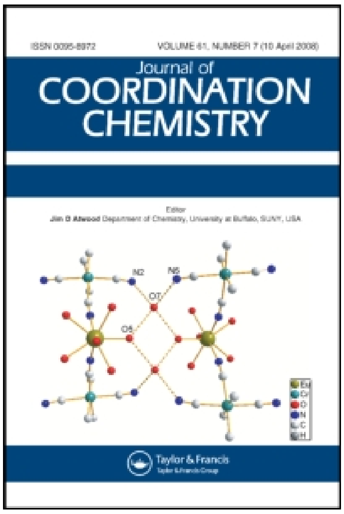

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Synthesis, spectral characterization, <i>in vitro</i> biological and DNA cleavage studies of Co(II), Ni(II), Cu(II), and Zn(II) complexes with 1,2,4-triazole Schiff bases

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Synthesis, spectral characterization, *in vitro* biological and DNA cleavage studies of Co(II), Ni(II), Cu(II), and Zn(II) complexes with 1,2,4-triazole Schiff bases

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A series of metal complexes of cobalt(II), nickel(II), copper(II), and zinc(II) have been synthesized with newly-derived biologically active ligands. These ligands were synthesized by condensation of 3-substituted-4-amino-5-hydrazino-1,2,4-triazole and orthophthalaldehyde. The probable structure of the complexes has been proposed on the basis of elemental analyses and spectral (IR, ^{1}H-NMR, UV-vis, magnetic, ESR, FAB-mass and thermal studies) data. Electrochemical study of the complexes is also made. All complexes are nonelectrolytes in *N,N*-dimethyl formamide and DMSO. The Schiff bases and their Co(II), Ni(II), Cu(II), and Zn(II) complexes have been screened for antibacterial (*Escherichia coli*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and *Pseudomonas aeruginosa*) and antifungal (*Aspergillus niger*, *Aspergillus flavus*, and *cladosporium*) activities by minimum inhibitory concentration method. DNA cleavage is also carried out.

Keywords: Synthesis; Biological activity; Electrochemical; 1,2,4-Triazole; Orthophthalaldehyde; DNA; Complexes

1. Introduction

Coordination chemistry of macrocyclic complexes has undergone spectacular growth due to the synthesis of a large number and variety of synthetic macrocycles [1, 2]. Template reactions have been widely used as synthetic routes for macrocyclic complexes [3–5]. Nitrogen-containing macrocycles have a strong tendency to form stable complexes with transition metals [6]. Some macrocyclic complexes exhibit antibacterial and antifungal activities [7, 8]. Macrocyclic nickel complexes are used in DNA recognition and oxidation [9], whereas macrocyclic copper complexes in DNA binding and cleavage [10]. Triazole derivatives [11–14] possess antibacterial, fungicidal, hypotensive, and hypothermic activities. Metal complexes of 1,2,4-triazole derivatives have been extensively investigated and reported from our laboratory [15–19].

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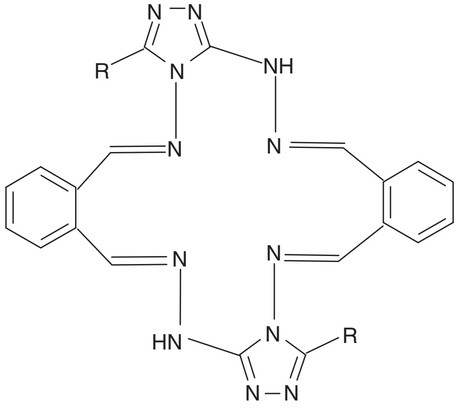
In continuation of our efforts on 1,2,4-triazole derivatives, we have synthesized a new series of macrocyclic Schiff bases are derived from 3-substituted-4-amino-5-hydrazino-1,2,4-triazole and orthophthalaldehyde. Survey of the literature reveals that no work has been carried out on the synthesis of metal complexes with these Schiff bases. We report the Co(II), Ni(II), Cu(II), and Zn(II) complexes with these bioactive Schiff bases (figure 1), which are characterized by spectral, thermal, and molar conductance studies and are evaluated for their antibacterial and antifungal properties against various pathogenic bacterial strains using the minimum inhibitory concentration (MIC) method.

2. Experimental

2.1. *Physical measurements*

IR spectra of the ligands and their Co(II), Ni(II), Cu(II), and Zn(II) complexes were recorded on a HITACHI-270 IR spectrophotometer in the 4000–250 $\mathrm{cm^{-1}}$ region in KBr disks. Electronic spectra of the complexes were recorded in *N,N*-dimethyl formamide (DMF) on a VARIAN CARY 50-BIO UV-spectrophotometer from 200 to 1100 nm. The ^{1}H NMR spectra of ligands and complexes were recorded in DMSO-d_6 on a BRUKER 300 MHz spectrometer at room temperature using TMS as an internal reference. Thermogravimetric (TG) analyses, measured from room temperature to 1000°C at a heating rate of 10°C $\mathrm{min^{-1}}$, were obtained by using a Perkin–Elmer Diamond TG/derivative thermogravimetric (DTG) instrument. Elemental analyses for C, H, and N were done using Elementer Vario EL III Carlo Erba 1108 instrument. The fluorescence study was carried out on a HITACHI F-7000. FAB-mass spectra were recorded on a JEOL SX 102/DA-6000 mass spectrometer/data system using argon/xenon (6 kV, 10 mA) as the FAB gas. The accelerating voltage was 10 kV and the spectra were recorded at room temperature. *m*-Nitrobenzyl alcohol was used as the matrix. The mass spectrometer was operated in the positive ion mode.



R=H, CH_3, C_2H_5, and C_3H_7

Figure 1. Synthesized Schiff bases.

Molar conductivity measurements were recorded on an ELICO-CM-82 T conductivity bridge with a cell having cell constant 0.51. Electrochemical studies were carried out using a CHI1110A-Electrochemical analyzer; magnetic moment measurements were carried out by Faraday balance.

2.2. *Synthesis*

All chemicals for preparing triazoles and their Schiff bases were of reagent grade. The 3-substituted-4-amino-5-mercapto-1,2,4-triazoles were prepared as reported [20, 21]. Orthophthalaldehyde was obtained from Aldrich Chemical Company.

2.3. *Synthesis of 3-substituted-4-amino-5-hydrazino-1,2,4-triazole*

A mixture of 3-substituted-4-amino-5-mercapto-1,2,4-triazole and $N_2H_4 \cdot H_2O$ in 1 : 1 molar proportions in EtOH was boiled under reflux for 4–5 h on a water bath. The reaction mixture was cooled at room temperature and within an hour, the compound separated from the clear solution. It was filtered, washed, and recrystallized from EtOH.

2.4. *Synthesis of Schiff bases [I–IV]*

A 25 mL aqueous methanolic solution (0.067 g, 0.02 mol) of orthophthalaldehyde was added to the 25 mL aqueous methanolic solution (0.065 g, 0.02 mol) of 3-substituted-4-amino-5-hydrazino-1,2,4-triazole dropwise with constant stirring. Then, an equimolar amount of sodium acetate (0.068 g, 0.02 mol) was added and the whole mixture stirred for 20 min on a hot plate stirrer. The pale yellow product was filtered, washed with cold methanol and ether, recrystallized from ethanol and vacuum dried. The purity of the product was monitored by TLC using chloroform : ethanol (7 : 3).

2.5. *Synthesis of Co(II), Ni(II), Cu(II), and Zn(II) complexes [1–16]*

To 25 mL of methanol solution of metal(II) salts, 25 mL of Schiff-base solution was added in a 100 mL round bottom flask. The reaction mixture was stirred magnetically. The resulting solution was concentrated under reduced pressure and a few mL of diethylether was added to initiate crystallization. The resulting precipitate was separated by suction filtration, washed with diethylether, vacuum dried to get a crystalline compound and recrystallized using dichloromethane and diethylether.

2.6. *Analyses*

The metal contents were estimated gravimetrically by the standard method [22]. Carbon, hydrogen, and nitrogen were estimated by C, H, and N analyzer. The presence of chloride is estimated using the standard method [23]. The results of elemental analyses and molar conductance values are listed in table 1.

Table 1. Elemental analysis of Co(II), Ni(II), Cu(II), and Zn(II) complexes and their magnetic and molar conductance data.

Compound No.	Empirical formula	M%		C%		H%		N%		Molar conductance $\Omega^{-1}\,cm^{-2}\,mol^{-1}$	Magnetic Moments (μ_{eff} BM)
		Obsd	Calcd	Obsd	Calcd	Obsd	Calcd	Obsd	Calcd		
I	$C_{20}H_{16}N_{12}$	–	–	56.57	56.60	3.71	3.77	39.57	39.62	–	–
II	$C_{22}H_{20}N_{12}$	–	–	58.38	58.41	4.38	4.42	37.12	37.17	–	–
III	$C_{24}H_{24}N_{12}$	–	–	59.93	60.00	4.96	5.00	34.92	35.00	–	–
IV	$C_{26}H_{28}N_{12}$	–	–	61.37	61.42	5.48	5.51	33.01	33.07	–	–
1	$[Co(C_{20}H_{16}N_{12})Cl_2]$	10.61	10.66	43.38	43.41	2.82	2.89	30.32	30.38	28	4.62
2	$[Co(C_{22}H_{20}N_{12})Cl_2]$	10.09	10.14	45.40	45.44	3.40	3.44	28.87	28.91	21	4.90
3	$[Co(C_{24}H_{24}N_{12})Cl_2]$	09.61	9.67	47.26	47.29	3.90	3.94	27.51	27.58	20	4.52
4	$[Co(C_{26}H_{28}N_{12})Cl_2]$	09.21	9.25	48.94	48.98	4.34	4.39	26.32	26.37	23	4.63
5	$[Ni(C_{20}H_{16}N_{12})Cl_2]$	10.57	10.62	43.39	43.42	2.84	2.89	30.33	30.39	19	3.35
6	$[Ni(C_{22}H_{20}N_{12})Cl_2]$	10.07	10.11	45.41	45.46	3.41	3.44	28.89	28.93	24	3.22
7	$[Ni(C_{24}H_{24}N_{12})Cl_2]$	09.59	9.64	47.28	47.31	3.90	3.94	27.55	27.60	17	3.15
8	$[Ni(C_{26}H_{28}N_{12})Cl_2]$	09.19	9.22	48.95	49.00	4.33	4.39	26.34	26.38	23	3.23
9	$[Cu(C_{20}H_{16}N_{12})Cl_2]$	11.32	11.39	43.00	43.04	2.81	2.86	30.09	30.13	28	1.68
10	$[Cu(C_{22}H_{20}N_{12})Cl_2]$	10.81	10.85	45.02	45.08	3.38	3.42	28.63	28.69	27	1.80
11	$[Cu(C_{24}H_{24}N_{12})Cl_2]$	10.30	10.35	46.91	46.94	3.86	3.91	27.34	27.38	21	1.72
12	$[Cu(C_{26}H_{28}N_{12})Cl_2]$	09.86	9.90	48.60	48.63	4.31	4.36	26.15	26.18	23	1.65
13	$[Zn(C_{20}H_{16}N_{12})Cl_2]$	11.62	11.68	42.85	42.90	2.81	2.86	30.00	30.03	27	–
14	$[Zn(C_{22}H_{20}N_{12})Cl_2]$	11.09	11.13	44.90	44.94	3.38	3.40	28.55	28.60	21	–
15	$[Zn(C_{24}H_{24}N_{12})Cl_2]$	10.58	10.62	46.73	46.79	3.83	3.89	27.26	27.29	26	–
16	$[Zn(C_{26}H_{28}N_{12})Cl_2]$	10.12	10.16	48.44	48.49	4.31	4.35	26.08	26.11	20	–

2.7. *Preparation of culture media for DNA study*

Nutrient broth (peptone, $10\,\mathrm{g\,L^{-1}}$; Yeast extract, $5\,\mathrm{g\,L^{-1}}$; NaCl, $10\,\mathrm{g\,L^{-1}}$) is used for culturing of *Escherichia coli* and potato dextrose broth (potato, $250\,\mathrm{g\,L^{-1}}$; dextrose, $20\,\mathrm{g\,L^{-1}}$) is used for culture of *Aspergillus niger*. The 50 mL media is prepared and autoclaved for 15 min at 121°C under 15 lb pressure. The autoclaved media are inoculated with the seed culture and *E. coli* is incubated for 24 h and *A. niger* for 48 h at 37°C.

2.8. *Isolation of DNA*

Fresh bacterial culture (1.5 mL) is centrifuged to obtain the pellet, which is then dissolved in 0.5 mL of lysis buffer (100 mM *tris* pH 8.0, 50 mM EDTA, 10% SDS). To this 0.5 mL of saturated phenol is added and incubated at 55°C for 10 min. Then it is centrifuged at 10,000 rpm for 10 min and equal volume of chloroform : isoamyl alcohol (24 : 1) and 1/20-th volume of 3M sodium acetate (pH 4.8) is added to this supernatant and centrifuged at 10,000 rpm for 10 min. To this supernatant three volumes of chilled absolute alcohol is added. The precipitated DNA is separated by centrifugation. The pellet is dried and dissolved in TE buffer (10 mM *tris* pH 8.0, 1 mM EDTA) and stored cold.

3. Results and discussion

All the Co(II), Ni(II), Cu(II), and Zn(II) complexes are stable at room temperature, nonhygroscopic and soluble in DMF and DMSO. The elemental analyses shown in table 1 agree with formation of $\mathrm{ML\cdot Cl_2}$. The molar conductivity measurements at 10^{-3} M in DMF indicate that these complexes are nonelectrolytes.

3.1. *Infrared spectra*

In the IR spectra of the Schiff bases, a medium intensity broad band observed from 3330 to 3358 $\mathrm{cm^{-1}}$ is attributed to the ν(NH). This band was observed in all the complexes in the range 3315–3340 $\mathrm{cm^{-1}}$, confirming that NH is not involved in coordination. A medium to high-intensity band in the range 1625–1600 $\mathrm{cm^{-1}}$ is assigned to ν(C=N) [24] which shifts lower by 20–25 $\mathrm{cm^{-1}}$ to 1602–1580 $\mathrm{cm^{-1}}$ [25]. This supports Schiff-base coordination to metal through the nitrogen of C=N [26]. This is further supported by the appearance of a medium intensity band in the region 525–505 $\mathrm{cm^{-1}}$ assigned to ν(M–N) [27]. A band in the range of 320–305 $\mathrm{cm^{-1}}$ in the far IR spectra of all the complexes indicates the presence of two *trans* chlorides [28, 29]. Various absorption bands in the region 1400–1575 $\mathrm{cm^{-1}}$ are assigned to ν(C=C) aromatic stretching vibrations [30, 31]; bands in the region 740–785 $\mathrm{cm^{-1}}$ are assigned to ν(C–H) out of plane bending of aromatic rings [32, 33].

ν(M–Cl) vibrations are sensitive to metal oxidation state and coordination number [34, 35]. These bands usually appear as medium to weak intensity bands from 400–300 $\mathrm{cm^{-1}}$; weak intensity bands at 320–305 $\mathrm{cm^{-1}}$ are assigned to ν(M–Cl).

3.2. *NMR spectra*

The ^{1}H NMR spectra of the Schiff bases have signals at 13.90 (s, 1H, NH) [36], 8.68 and 7.1–7.8 (m, 4H, Ar-H) ppm due to –NH, –CH=N [37], and aromatic protons, respectively.

The Zn(II) complex has the azomethine protons at 9.02 ppm inferring coordination through the azomethine nitrogen [30].

3.3. *FAB mass spectral studies*

The FAB mass spectrum of Schiff base (II) (Supplementary Material online) shows a molecular ion peak at *m*/*z* 452 which is equivalent to its molecular weight. The fragments in the spectrum lead to the formation of $[C_{22}H_{20}N_{12}]^+$.

The FAB mass spectrum of the Ni(II) complex (**6**) have a molecular ion peak M^+ at *m*/*z* 580 (Supplemental Material). The molecular ion peak fragmentation with the loss of two chlorides gave a peak at *m*/*z* 510 due to $[Ni(C_{22}H_{20}N_{12})]^+$; further, loss of $C_3H_4N_6$ gave a fragment ion at *m*/*z* 386.

3.4. *Electronic spectra*

Cobalt complexes: The electronic spectra of cobalt(II) complexes exhibit absorptions at 8070–9060(ν_1), 12,490–15,680(ν_2), and 18,780–20,290 cm^{-1}(ν_3). The spectra resemble those for complexes reported to be octahedral [38]. The spectra show a band in the visible region, split due to the presence of low symmetry. Assuming the effective symmetry to be D_{4h}, the various bands can be assigned to: $^4T_{1g}$ (F) → $^4T_{2g}$(F) (ν_1), $^4T_{1g}$ (F) → $^4A_{2g}$(F) (ν_2) and $^4T_{1g}$ (F) → $^4T_{1g}$ (P) (ν_3), respectively. The assignment of the first spin-allowed band seems plausible since the first band appears at approximately half the energy of the visible band [39].

Nickel complexes: Electronic spectra of Ni(II) complexes exhibit a band with a shoulder on the low-energy side. The other two bands generally observed in the region at 16,680–17,060 cm^{-1} (ν_2) and 27,760–28,220 cm^{-1} (ν_3) are assigned to $^3A_{2g} \rightarrow {}^3T_{1g}$(F) ($\nu_2$) and $^3A_{2g} \rightarrow {}^3T_{1g}$(P) ($\nu_3$), respectively. The first two bands result from splitting of ν_1 and are in the range ca 9730–10,500 and 11,810–12,340 cm^{-1}, assigned to $^3B_{1g} \rightarrow {}^3E_g$ and $^3B_{1g} \rightarrow {}^3B_{2g}$, assuming the effective symmetry to be D_{4h} (component of $^3T_{2g}$ in O_h symmetry) [40]. The intense higher energy band at ca. 34,520 cm^{-1} may be due to a π–π^* transition of the (C=N) group. The spectra are consistent with distorted octahedral complexes.

Copper complexes. Electronic spectra of copper(II) complexes exhibit bands in the region *ca* 17,840–19,660 cm^{-1} with a shoulder on the low-energy side at *ca* 14,630–16,070 cm^{-1}, showing that these complexes are distorted octahedra [38, 39]. Assuming tetragonal distortion in the molecule, the d-orbital energy level sequence for these complexes may be: x^2–$y^2 > z^2 > xy > xz > yz$ and the shoulder can be assigned to: $z^2 \rightarrow x^2 - y^2$ ($^2B_{1g} \rightarrow {}^2B_{2g}$). The broad band contains both the $xy \rightarrow x^2 - y^2$ ($^2B_{1g} \rightarrow {}^2E_g$) and xz, $yz \rightarrow x^2 - y^2$ ($^2B_{1g} \rightarrow {}^2A_{2g}$) transitions [40]. The band separation in the spectra is of the order of 2500 cm^{-1}, consistent with the proposed geometry [40].

3.5. *Magnetic studies*

The magnetic moments obtained at room temperature are listed in table 1. The magnetic measurements for Co(II) and Ni(II) complexes are 4.80–4.96 B.M. and 2.8–3.5 B.M., respectively [41], consistent with their octahedral environment. The Cu(II) complexes show magnetic moments of 1.75–1.87 B.M., slightly higher than the spin-only value 1.73 B.M. expected for one unpaired electron [42].

3.6. *ESR spectra of (10)*

The ESR spectrum of (**10**) shows $g_{\|}$ and $g_{\perp}$ values of 2.0948 and 2.02698, respectively. The trend $g_{\|} > g_{\perp} > g_e$ observed for the present complex indicates that the unpaired electron is localized in the $d_{x^2-y^2}$ orbital of Cu(II). The axial symmetry parameter G observed for this complex (3.3537) is <4 indicating exchange interaction in the solid complex, consistent with distorted octahedral configuration [43].

3.7. *TG analyses*

TG and DTG showed only a single decomposition curve around 290–305°C corresponding to loss of organic moiety (Supplemental Material). Above 500°C, metal(II) compounds decomposed to their respective metal oxides.

3.8. *Electrochemical studies*

Electrochemical properties of the complexes were studied on a CHI1110A-Electrochemical analyzer in DMF containing 0.05 M *n*-Bu_4NClO_4 as the supporting electrolyte. A cyclic voltammogram (Supplemental Material) of (**10**) displays a reduction peak at $E_{pc} = 0.1073$ V with a corresponding oxidation peak at $E_{pa} = 0.6327$ V. The peak separation (ΔE_p) is 0.5254 V at a scan rate 0.1 V and increases with scan rate [44, 45]. The difference between forward and back peak potentials provides a rough evaluation of the degree of reversibility of one electron transfer. Cyclovoltammetric responses with the scan rate varying from 50 to 250 mV s^{-1} gives evidence for quasi-reversible one electron oxidation.

The ratio of cathodic to anodic peak height was less than one. However, the peak current increases with increase of the square root of the scan rates, establishing the electrode process as diffusion controlled [46]. Separation in peak potential increases at higher scan rates consistent with quasi-reversibility of the Cu(II)/Cu(I) couple.

4. Biological activities

4.1. In vitro *antibacterial and antifungal assay*

All the synthesized Schiff bases (I–IV) and their corresponding Co(II), Ni(II), Cu(II), and Zn(II) complexes were screened *in vitro* for their biological activity by using bacteria, *E. coli*, *Staphylococcus aureus*, *Streptococcus pyogenes*, and

Pseudomonas aeruginosa and fungi, *A. niger*, *Aspergillus flavus* and *cladosporium* by the reported method [47, 48]. Stock solution (1 mg mL^{-1}) was prepared by dissolving 10 mg of the test compound in 10 mL of DMF. Control for each dilution was prepared by diluting 10 mL of solvent instead of stock solution with sterilized distilled water.

The bacteria were subcultured in agar medium. The petridishes were incubated for 24 h at 37°C. Standard antibacterial drug (Gentamycine) was also screened under similar conditions for comparison. The fungi were subcultured in potato dextrose agar medium. Standard antifungal drug (Fluconazole) was used for comparison. The petridishes were incubated for 48 h at 37°C. Wells were dug in the agar media using a sterile metallic borer. Activity was determined by measuring the diameter of the zone showing complete inhibition (mm). Growth inhibition was compared with the standard drugs. In order to clarify any effect of DMF on the biological screening, separate studies were carried out with solutions of DMF and they showed no activity against any microbial strains.

4.2. *Antimicrobial results*

Microbial results are systematized in tables 2 and 3. The antibacterial and antifungal studies suggest that all Schiff bases were biologically active and their metal(II) complexes showed significantly enhanced antibacterial and antifungal activity [49, 50].

In the case of bacteriological studies, the results were compared with the standard drug (Gentamycine). Some Schiff bases were active against all bacterial strains. Compound (IV) shows high activity against all bacterial strains, especially with *P. aeruginosa* and *E. coli*. Potency was enhanced on coordination with metal ions with Ni(II) and Cu(II), exhibiting high activity.

Antifungal activities were compared with the standard drug (Flucanazole). All Schiff bases show high activity against fungal species with (I) and (III) showing very high activity, and (III) showing higher activity than Flucanazole against *A. niger*. Metal(II) complexes showed much enhanced activity, especially with *Cladosporium*.

5. DNA cleavage experiment

5.1. *Agarose gel electrophoresis*

Cleavage products were analyzed by agarose gel electrophoresis. Test samples (1 mg mL^{-1}) were prepared in DMF and (25 µg) added to the isolated DNA of *E. coli* and *A. niger*. The samples were incubated for 2 h at 37°C and then 20 µL of DNA sample (mixed with bromophenol blue dye at 1 : 1 ratio) added carefully into the electrophoresis chamber wells along with standard DNA marker containing TAE buffer (4.84 g *Tris* base, pH 8.0, 0.5 M EDTA/1 L) and finally loaded on agarose gel and treated with 50 V of electricity for around 30 min. Removing the gel and staining with 10.0 µg mL^{-1} ethidium bromide for 10–15 min, bands were observed under UV transilluminator and photographed to determine the extent of DNA cleavage; the results are compared with standard DNA marker.

Table 2. Antibacterial and antifungal results of Schiff bases (I–IV).

		Antibacterial activity (Zone of inhibition in %)				Antifungal activity (Zone of inhibition in %)		
Compound	Concentraction (μg mL^{-1})	*E. coli*	*S. aureus*	*S. pyogenes*	*P. aeruginosa*	*A. flavus*	*Cladosporium*	*A. niger*
I	100	–	62	50	60	50	54	–
II	100	64	–	–	59	–	–	57
III	100	60	58	49	62	53	48	42
IV	100	59	61	–	65	61	60	55
Standards	100	100	100	100	100	100	100	100

Table 3. MIC (μg mL^{-1}) results for metal(II) complexes.

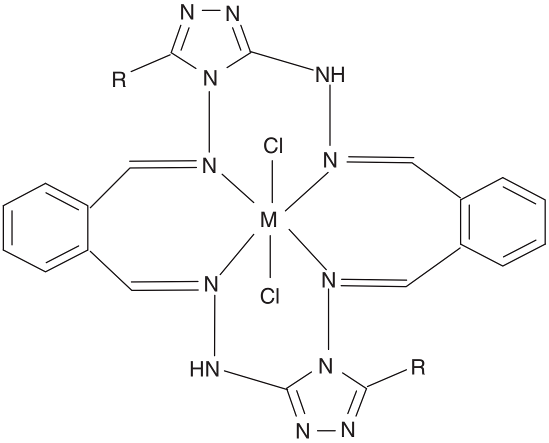
	Antibacterial activity (Zone of inhibition in %)				Antifungal activity (Zone of inhibition in %)		
Compound	*E. coli*	*S. aureus*	*S. pyogenes*	*P. aeruginosa*	*A. flavus*	*Cladosporium*	*A. niger*
1	15	15	–	10	20	25	25
2	15	–	25	25	20	25	25
3	–	25	25	15	25	20	–
4	10	–	25	–	20	–	20
5	10	20	25	20	–	15	20
6	10	20	–	15	–	20	–
7	10	–	20	15	15	–	25
8	15	20	–	20	15	–	–
9	–	25	20	10	20	10	20
10	10	–	20	15	15	15	25
11	15	10	15	–	25	10	20
12	10	15	15	10	20	10	20
13	–	15	–	20	15	–	20
14	10	–	15	15	15	20	10
15	–	20	10	10	–	20	–
16	15	10	15	10	25	20	15

5.2. *Electrophoretic analysis*

The Schiff bases and their Co(II), Ni(II), and Cu(II) complexes were studied for DNA cleavage activity by agarose gel electrophoresis method revealing that only the Cu(II) complex acted on DNA. The difference was observed in the bands (Lanes 2 and 6 of Supplemental Material) compared to the control DNA of *E. coli* and *A. niger*. The control DNA alone does not show cleavage whereas Cu(II) complexes do. The nature of reactive intermediates in DNA cleavage by the complexes is not clear. The results indicated the role of metal in DNA cleavage reactions.

6. Conclusion

The synthesized macrocyclic Schiff bases (I–IV) are tetradentate through azomethine nitrogens, confirmed by analytical, IR, ^{1}H NMR, electronic, magnetic, ESR, FAB



R=H, CH_3, C_2H_5, and C_3H_7

M = Co(II), Ni(II), Cu(II), and Zn(II)

Figure 2. Octahedral structure of metal(II) complexes.

mass, and thermal studies. Electrochemical study of Cu(II) complex has quasi-reversible character.

Biological results confirm that all the Schiff bases are biologically active and their metal(II) complexes have more activity.

We propose the structures shown in figure 2 for $MLCl_2$ [M = Co(II), Ni(II), Cu(II), and Zn(II)].

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