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Synthesis, Characterization, Electrochemical Investigation and Antibacterial Activity of Chiral 1, 1' - (4, 6 - Dihydroxy - 1, 3 -Phenylene) Bisethanone based 18 --Membered Tetraaza Macrocyclic Co (II), Ni (II), Cu (II) and Zn (II) Metal Complexes

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Abstract: A template synthesis of 18- membered tetraaza macrocyclic ligand and this ligand were complexes with cobalt, nickel, copper and zinc metals chlorides. These synthesized complexes were characterized by various physicochemical methods such as elemental analysis, molar conductance, magnetic properties, spectral studies of infra-red, electronic spectrum, the redox properties of metal complexes were determined in the cyclic voltammetry and antibacterial activity. This research studies were clarifies the structure of the metal complexes were using with spectral data and molar conductance of the complexes. The structure of Co (II), Ni (II) and Cu (II) transition metal complexes was octahedral and Zn (II) complex was tetrahedral. These transition metal complexes were coordinated through the four nitrogen atoms of the ligand. The molar conductance studies revealed that Co (II), Ni (II) and Cu (II) complexes were non-ionic (1:0) and Zn (II) complex was ionic (1:2) in nature. The in-vitro antibacterial activity method exhibited all the complexes were good antibacterial activities.

Keywords: 18-Membered tetraaza macrocyclic complexes; magnetic properties; electrochemical investigation; electronic spectra; antibacterial activity.

I. INTRODUCTION

The previous few decades' the design and synthesis of macrocyclic complexes of transition metals was elaborate in the consideration of both inorganic and bioinorganic chemistry. The synthesis of macrocyclic complexes of transition metals has been a fascinating area of research work has done and last few decades to growing at a very fast due to the outstanding their resemblance with naturally occurring macrocyclic transition metal complexes in the field of analytical, industrial, and medical applications [1–2]. The template synthesis of tetraaza macrocyclic complexes was mimic the properties of naturally occurring difficult protein derivatives and provides the primary structural components to the coordination site thereby determining the electronic properties of the metal ion were present in the active site macrocyclic complexes [3]. This metal complex was controlled in the electronic properties of the metal ions like heme in haemoglobin, myoglobin and chlorophyll naturally occurring proteins involved redox processes [4]. This type of naturally occurring tetraza macrocyclic complexes are main core of biological mimics like haemoglobin, myoglobin and chlorophyll [5-6]. These tetraaza macrocyclic complexes were playing important role of electron transport, dioxygen transport and catalytic activity of many metalloenzyme reactions [7-8]. The tetraaza macrocyclic complexes are very stable due to best fitting cavity size of the macrocyclic ligands through the coordinated with the transition metal ions [8-10]. The macrocyclic complexes are also important due to its using in preparing the dyes and pigments, trapping of toxic metal ions in the varies chemical industries, sequestration of toxic metals and NMR shift reagents [11-12]. The emergence of the new materials allows the use for organic light emitting devices (OLEDs). A number of emission materials and electron transport materials have been reported such as metal-chelates complexes having tetraaza macrocyclic ligands. In generally heterocyclic tetraaza macrocyclic complexes was containing the electron deficient nitrogen atoms it has suitable for this electron transfer properties [13-16]. Now a day's lot of research effort has done by these transition metals with macrocyclic ligand in studying as antifungal, antibacterial, anticancer and antiviral activities [17-18]. In this view of the above applications we present the paper and report the synthesis, spectral characterization and antibacterial activity of Co (II), Ni (II), Cu (II) and Zn (II) transition metal ions has complexes with 18-membered tetraza macrocyclic ligand (L) has been carried out.

II. EXPERIMENTAL TECHNIQUES

A. Materials and Methods

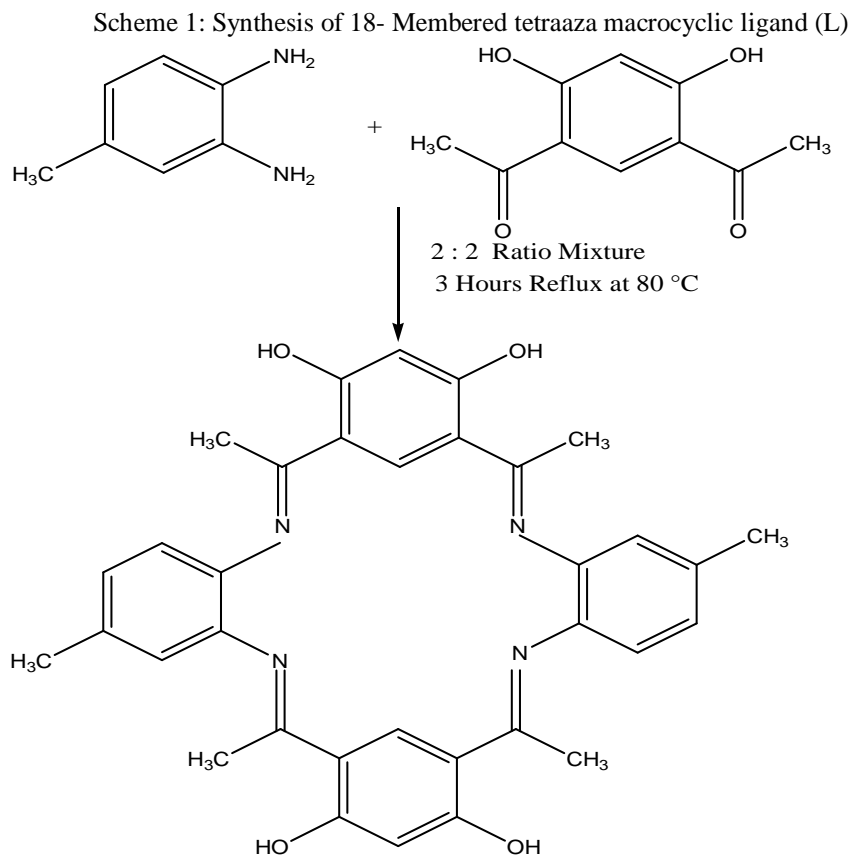
All the chemicals and reagents were used to annular grade and purchased from Sigma–Aldrich and transition metal chlorides were purchased from E. Merck. The elemental analysis (C, H, N and O) were analyzed by a Carlo-Erba 1106 elemental analyzer. Molar conductance was measured by the ELICO (CM82T) conductivity bridge. Magnetic susceptibility was measured at room temperature by a Gouy balance using $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ as a calibrant. ^1H and ^{13}C NMR spectra were recorded by a Hitachi FT-NMR, model R-600 spectrometer using CDCl_3 as solvent. The Chemical movements were given in parts per million with respect to tetramethylsilane. IR spectra (KBr) were recorded on FT-IR range BX-II spectrophotometer. The electronic spectra were recorded in DMF on Shimadzu UV smaller than normal 1240 spectrophotometer. The electrochemical analyses were conveyed by utilizing Auto Lab instrument (Metrohm 663 VA Stand) in dimethylformamide (DMF) covering TEAP as supporting electrolyte by utilizing cyclic voltammetry strategies. This framework contain three anode framework comprises of Pt plate terminal (2mm distance across) as a working cathode, Ag/AgCl (3M KCl) reference anode and Pt wire cathode as helper terminal. Pre-treatment of terminals was done before each cyclic voltammetry experiments.

B. Synthesis of the Macrocyclic Ligand (L)

An ethanolic solution of 4, 6- diacetyl resorcinol 0.194 g (1 mM) and 4- Methyl - O- Phenylenediamine 0.122 g (1 mM) dissolved separately in absolute ethanol with molar ratio (1:1) were mixed together and the solution mixture was left under reflux for three hours. The reaction mixture was collected and then cooled to form yellow colour precipitate, which was separated out by filtration and washed several times with a small amount of absolute ethanol. The ligand was recrystallized, in absolute ethanol. The crystals were dried over anhydrous calcium chloride under vacuum and by end of the process 62% of the yield was collected (Scheme 1).

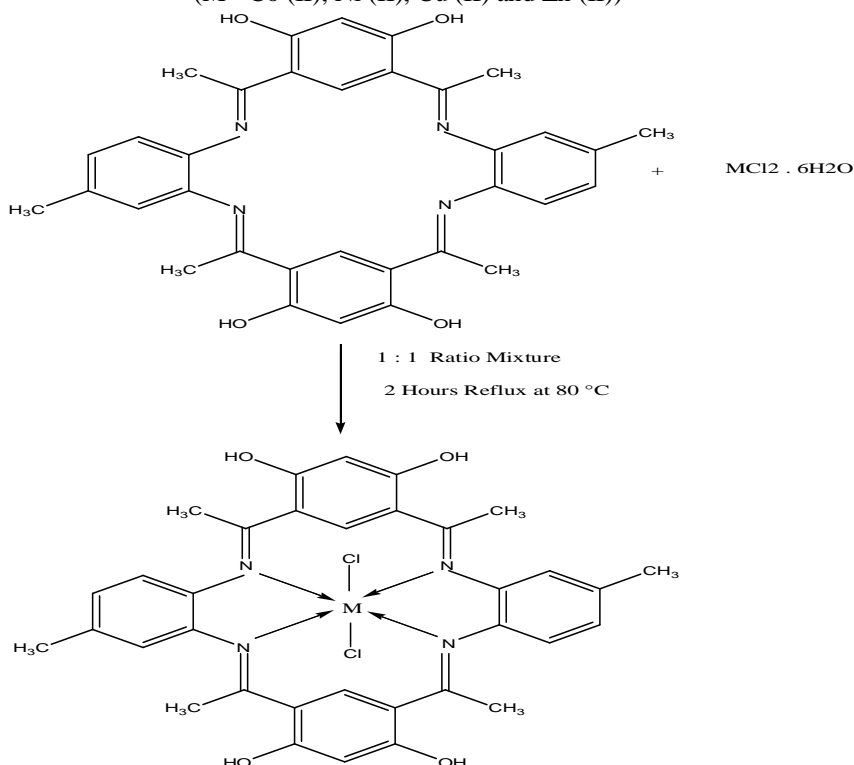
C. Synthesis of the Macrocyclic Complexes

A transition metal chlorides (1 mM) were dissolved in absolute ethanol was added in to the (1mM) dissolved ethanolic solution of macrocyclic ligand (L). The resulting mixture was refluxed for 3hrs and then cooled to form the colored precipitate which is separated by filtration. This filtrate is washed repeatedly with absolute ethanol and then dried over anhydrous calcium chloride in vacuum (Scheme 2).



Scheme 2: Synthesis of Macrocyclic Complexes

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



D. Antibacterial Activity Studies

The antibacterial activity of macrocyclic ligand (L) and macrocyclic complexes were tested against three types of different bacterial species, Escherchia coli (E.Coli), Streptococcus pneumoniae (S.pneumonia) and Pseudomonas. The procedure as followed from reported literatures [19-20].

III. RESULTS AND DISCUSSION

The macrocyclic ligand (L) and macrocyclic complexes were soluble in ethanol, methanol, acetone, acetonitrile, DMSO and DMF. The transition metals and macrocyclic ligand (L); stoichiometry ratio were (1:1), which is confirmed from the analytical reports.

A. Characterization of Macrocyclic Ligand (L)

The macrocyclic ligand (L) was synthesis and characterized on the basis of elemental analysis, Infra-red spectra, ^1H NMR and ^{13}C NMR spectrum. The elemental analysis to provide the percentage of (C,H,N and O) were analysis and verified the macrocyclic ligand was formed, IR spectra the carbonyl stretching frequency is very much decrease from 1680 cm^{-1} to 1605 cm^{-1} it is indicate the carbonyl group (C=O) is converted to imine group (C=N). So that ligand (L) was conformed to formed, it has cyclic in nature and it was verified by some chemical test. ^1H NMR spectrum of the macrocyclic ligand (L) does not give any signal corresponding to primary amine protons. The multiplet in the range of $8.2\text{ }\delta\text{ ppm}$ it indicate that aromatic imine group, it has propose that the imine (C=N) groups is formed. A multiplet in the $\delta\text{ ppm}$ range is $2.20\text{--}2.64$ $\delta\text{ ppm}$ region, may be recognized to methylene protons was attached with phenyl ring, another multiplet in the region of $7.28\text{--}7.52\text{ }\delta\text{ ppm}$, can be assigned to the aromatic ring protons. ^{13}C NMR spectrum range is $100\text{--}150\text{ }\delta\text{ ppm}$ aromatic double bond carbon atoms and the range $170\text{ }\delta\text{ ppm}$ has aromatic imine carbon atoms. So these all above data's has to provide the essential conformation of the formation of cyclic 18- membered tetraza macrocyclic ligand (L).

B. Characterization of the Macrocyclic Complexes

The macrocyclic ligand (L) was reacted with Co (II), Ni (II), Cu (II) and Zn (II) metal chlorides to contribute the cyclic 18-membered tetraza macrocyclic complexes were designed. These macrocyclic complexes were characterized on the basis an elemental analysis, molar conductance measurements, magnetic moments values, electronic spectrum, infrared spectrum, cyclic voltammetry and antibacterial activity.

- 1) *Elemental Analysis*: The elemental analysis of the macrocyclic complexes were shown in Table and Fig. 1. These indicated that the observed percentage of ligand (C, H, N and O) with metal ions, these metal ions and ligand has present in decent arrangement with the values were in found and calculated the molecular weight were practically identical. So this indicated that was assuming the 1:1 molar ratio of metals and ligand to form the macrocyclic complexes.
- 2) *Molar Conductance Measurements*: The molar conductance data of the macrocyclic complexes were shown in Table 2. All the transition metal macrocyclic complexes were dissolved in 10^{-3} M concentration of DMF solution, the molar conductivities of the macrocyclic complexes was taken and measured at room temperature. The molar conductivity values of Co (II), Ni (II) and Cu (II) complexes were non- electric in nature and the value were experimentally observed in 23.24, 25.65 and 21.36. The conductance values of Zn (II) complexes was electric in nature and the value was experimentally observed in 96.86 [21].
- 3) *Magnetic Moments*: The magnetic moments value of the macrocyclic complexes were shown in Table 2. The experimentally observed magnetic moment value of Co(II) complex in present investigation were found to be 4.63 B.M. at room temperature indicating the presence of three unpaired electrons in the Co(II) complex. These magnetic moment values of Co (II) complex were slightly higher than that of expected from the original spin only value of 3.87 B.M. due to contribution of spin orbit coupling. So that Co (II) complex was paramagnetic in nature and high spin octahedral in geometry. The experimentally observed magnetic moment value of Ni (II) complex the value was 3.26 B.M. at room temperature it suggesting that the presence of two unpaired electrons in Ni (II) complex. These magnetic moment values of Ni (II) complexes were slightly higher than that of expected from the original spin only value of 2.83 B.M. due to involvement of spin orbit coupling. The Ni (II) complex was paramagnetic in nature and high spin octahedral in geometry. The experimentally observed magnetic moment value of Cu (II) complex the value was 1.84 B.M. at room temperature implying the existence of one unpaired electrons in Cu (II) complex. The Cu (II) complex was paramagnetic in nature and high spin octahedral in geometry. The experimentally observed magnetic moment value of Zn (II) complex has 0 B.M. at room temperature indicating no unpaired electrons in Zn (II) complex. The Zn (II) complex was diamagnetic in nature and high spin tetrahedral in geometry [22].
- 4) *Electronic Spectrum*: The electronic spectral data of the complexes were shown in Table 3 and Fig. 5. In this Co (II) complex revealed that the three electronic transitions were observed in the range of ${}^4T_{1g} \rightarrow {}^4T_{2g}$ (F) (10173 cm^{-1}), ${}^4T_{1g} \rightarrow {}^4A_{2g}$ (F) (18687 cm^{-1}) and Charge transfer (25580 cm^{-1}). So geometry of Co (II) complex was high spin octahedral in geometry. The Ni (II) complex has exhibited the three electronic transitions are observed in the range ${}^3A_{2g} \rightarrow {}^3T_{2g}$ (F) (9360 cm^{-1}), ${}^3A_{2g} \rightarrow {}^3T_{1g}$ (F) (14826 cm^{-1}) and Charge transfer (25890 cm^{-1}). So geometry of Ni (II) complex was high spin octahedral in geometry. The Cu (II) complex has exhibited the three electronic transitions are observed in the range of ${}^2B_{1g} \rightarrow {}^2A_{1g}$ (13497 cm^{-1}), ${}^2B_{1g} \rightarrow {}^2E_g$ (16320 cm^{-1}) and Charge transfer (27340 cm^{-1}). So geometry of Cu (II) complex was high spin octahedral in geometry. The Zn (II) complex revealed that the three electronic transitions are observed in the range of ${}^1A_{1g} \rightarrow {}^1A_{2g}$ ($18,240\text{ cm}^{-1}$), ${}^1A_{1g} \rightarrow {}^1B_{1g}$ ($20,950\text{ cm}^{-1}$) and Charge transfer (24570 cm^{-1}). So geometry of Zn (II) complex was high spin tetrahedral in geometry [23].
- 5) *Infrared Spectrum*: The IR spectra of the macrocyclic complexes were deliberate by conflicting with the IR spectra of macrocyclic ligand (L) to discover the essential approach of transition metals and macrocyclic ligand (L) were shown in Table and Fig. - 4. In IR spectra of the free ligand L, a sharp intensity $\nu(\text{C}=\text{N})$ band appeared at 1605 cm^{-1} [21]. In these macrocyclic complexes were imine nitrogen peaks appeared at 1596 and 1588 cm^{-1} which showing that the imine nitrogen atom takes part in the complexation. On these complexes formation the stretching frequency value of $\nu(\text{C}=\text{N})$ band is shifted by 8 to 15 cm^{-1} towards to decrease the stretching frequency. This suggests that the coordination through the nitrogen atoms of the imine groups with transition metal ions to form the metal complexes; it was indicating that imine groups have complexes with transition metal ions. These coordination behaviors of this ligand with transition metal complexes were also proved by the appearance of IR stretching frequency range were 426 to 408 cm^{-1} $\nu(\text{M}-\text{N})$ [22] due to coordination of nitrogen with metal complexes. The IR stretching frequency range is 298 to 321 cm^{-1} $\nu(\text{M}-\text{Cl})$ due to coordination of through the chlorides with transition metal complexes [24].
- 6) *ESR spectrum of Cu (II) complex*: The ESR spectrum of Copper complex was shown in Table 3 and Fig. 5. The ESR spectrum of Copper (II) complex was recorded in DMSO at 300 and 77 K . The g -tensor value of the complex was $g_{\parallel} > g_{\perp} > g_{\perp}$ (2.0023) specify that the unpaired electron present in the macrocyclic complexes. The ESR spectrum of Cu (II) complex provides the useful information about metal ion environment in the Copper (II) complex. The ESR spectrum of Copper (II) complex showed that $g_{\parallel} = 2.21$ and $g_{\perp} = 2.11$ suggest that the high spin octahedral environment. The bonding between the ligand and the Copper (II) metal complex were possesses a more covalent character than the ionic character. In the complexes

were both the parallel and the perpendicular g values were can be calculate by the G value of the following equation:

$$G = \frac{g_{\parallel} - 2.002}{g_{\perp} - 2.002}$$

We have using the above the equation, the G value of the Copper (II) complexes was 1.92 which clearly indicates that the Copper (II) metal ions was strongly complex with the macrocyclic ligand [25].

- 7) *Electrochemical Studies*: The cyclic voltammetry of macrocyclic complexes is shown in Table 3 and Fig. 7. The electrochemical studies of the macrocyclic complexes were carried out in the CH₃CN. There was a specific correlation between E_{1/2} and solvent it has depends on the interaction between the solvent and macrocyclic complexes [26]. These all the macrocyclic complexes considering the one electron quasireversible redox process on the of basis solvent effect on peak separation. The cyclic voltammetry behaviour of these macrocyclic complexes were studied in the range was -1.5 V to +1.5 V. The cyclic voltammograms of these macrocyclic complexes show quasireversible wave couples peaks. The E_{1/2} values are also similar with the reported values for the analogous complexes under the identical conditions [28]. The difference between the E_{pc} and E_{pa} peak potential for these macrocyclic complexes were very close to 59 mV which may be assigned to a one electron quasireversible process: Co(II)/Co(III) and Co(III)/Co(II) with similar to all other macrocyclic complexes with respect to the anodic and cathodic peaks respectively. The value of I_{pa} / I_{pc} ratio is also close to unity suggesting that these complexes show quasireversible electrochemical process [26-27].
- 8) *Antibacterial Activity*: The antibacterial activities of the macrocyclic complexes were shown in the Table 5 and Fig. 8. Antibacterial activity of macrocyclic complexes were screened against E. Coli, S. pneumonia and Pseudomonas using Muller-Hindon agar medium by well diffusion method using DMSO as solvent. All the transition metal complexes were very good antibacterial activity compare to the 18- membered tetraza macrocyclic ligand. We were taken in three different bacteria E.coli, Pseudomonas and S.pneumoniae. The MIC (Minimum Inhibitory Concentration) of test compounds against all these bacteria were taken in the sample has 200 µg, 400 µg, 600 µg and 800 µg at which 100% inhibition were experimentally observed. The increased activity of all the transition metal complexes can be explained on the basis of the concentration of the macrocyclic complexes. They results of all the macrocyclic complexes were indicate that concentration plays a major vital role in destroying the all these three bacteria. When the increasing the concentration of transition metal complexes increasing the degree of inhibition; as the concentration increases, the antibacterial activity transition metal complexes also increases. The complexes of transition metals has antibacterial activity against to the E.coli and Pseudomonas bacteria Ni (II) complex was more activity than other transition metal complexes; the S.pneumoniae bacteria Co(II) complex is more activity than other complexes [28].

Table 1: Elemental Analysis

S.No	Name of the Complexes	%M		% C		% H		% N		% O	
		Calc	Foun	Calc	Foun	Calc	Foun	Calc	Foun	Calc	Foun
1	L	-	-	72.85	71.76	05.35	05.12	10.00	09.65	05.35	05.23
2	[Co(II)LCl ₂]	08.41	08.20	59.21	58.98	04.35	04.25	08.12	07.89	09.28	09.20
3	[Ni(II)LCl ₂]	08.38	08.35	59.20	59.10	04.34	04.23	08.10	08.02	09.26	09.22
4	[Cu(II)LCl ₂]	09.00	08.90	58.78	58.65	04.31	04.27	08.06	08.01	09.22	09.16
5	[Zn(II)L]Cl ₂	09.30	09.20	58.62	57.87	04.30	04.10	08.04	08.00	9.19	9.12

Table 2: Molar conductance and Magnetic moments

S. No	Name of the Complexes	Colour	Molar conductance of the complexes (ohm ⁻¹ cm ² mol ⁻¹)		Magnetic Moment Value µ _{eff} (B.M)		Magnetic Properties
			Conductance	Electrolyte	Calc	Expt	
1	L	Yellow	-	-	-	-	-
2	[Co(II)LCl ₂]	Brown	23.24	1:0	4.63	3.87	Paramagnetic
3	[Ni(II)LCl ₂]	Light Brown	25.65	1:0	3.26	2.83	Paramagnetic
4	[Cu(II)LCl ₂]	Dark Brown	21.36	1:0	1.84	1.73	Paramagnetic
5	[Zn(II)L]Cl ₂	Yellow	96.86	1:2	0	0	Diamagnetic

Table 3: Electronic Spectrum

S. No	Name of the Complexes	ESR		Electronic Transitions (cm^{-1})			Stereochemistry
		g_{\parallel}	g_{\perp}				
1	L	-	-	-	-	-	-
2	[Co(II)LCI ₂]	-	-	$^4T_{1g} \rightarrow ^4T_{2g}(F)$ 10173	$^4T_{1g} \rightarrow ^4A_{2g}(F)$ 18687	C.T 25580	Octahedral
3	[Ni(II)LCI ₂]	-	-	$^3A_{2g} \rightarrow ^3T_{2g}(F)$ 9360	$^3A_{2g} \rightarrow ^3T_{1g}(F)$ 14826	C.T 25890	Octahedral
4	[Cu(II)LCI ₂]	2.21	2.11	$^2B_{1g} \rightarrow ^2A_{1g}$ 13497	$^2B_{1g} \rightarrow ^2E_g$ 16320	C.T 27340	Octahedral
5	[Zn(II)L]Cl ₂	-	-	$^1A_{1g} \rightarrow ^1A_{2g}$ 18,240	$^1A_{1g} \rightarrow ^1B_{1g}$ 20,950	C.T 24570	Tetrahedral

Table 4: Infrared Spectra of the complexes

S. No	Name of the Complexes	$\gamma(\text{C}=\text{N}) \text{ cm}^{-1}$	$\gamma(-\text{OH}) \text{ cm}^{-1}$	$\gamma(-\text{CH}_3) \text{ cm}^{-1}$	$\gamma(\text{M}-\text{N}) \text{ cm}^{-1}$
1	L	1605	3520	2986	-
2	[Co(II)LCI ₂]	1596	3545	2989	408
3	[Ni(II)LCI ₂]	1595	3537	2950	414
4	[Cu(II)LCI ₂]	1588	3526	2985	426
5	[Zn(II)L]Cl ₂	1597	3546	2992	423

Table 5: Cyclic Voltammetry

S. No	Name of the Complexes	EP_a (V)	EP_c (V)	$E_{1/2}$ (V)	ΔE	IP_c / IP_a
1	L	-	-	-	-	-
2	[Co(II)LCI ₂]	0.64 1.32	0.93 1.06	0.77 1.18	-0.23 -0.26	1.09
3	[Ni(II)LCI ₂]	1.03 -1.06	0.66 -1.57	0.87 -1.36	0.36 0.27	1.12
4	[Cu(II)LCI ₂]	1.31 -0.62	1.12 -0.54	1.23 0.57	-0.22 -0.13	0.97
5	[Zn(II)L]Cl ₂	1.02 -1.15	0.84 -1.66	0.93 -1.47	0.24 0.46	1.06

Table 5: Antibacterial Activity

S.No	Name of the Complexes	Concentration of extract in μl / zone of inhibition in mm (μg)											
		E.Coli				Pseudomonas				S.Pneumoniae			
		200	400	600	800	200	400	600	800	200	400	600	800
1	L	5.3	6.7	7.5	8.2	5.7	6.4	7.5	8.2	5.4	6.5	7.7	8.9
2	[Co(II)LCI ₂]	9.0	10.1	11.3	12.6	8.5	9.4	10.5	11.8	12.0	13.2	14.1	15.0
3	[Ni(II)LCI ₂]	12	13.3	14	16.0	11	12.1	13.2	14.3	9.2	9.7	10.7	12.3
4	[Cu(II)LCI ₂]	8.0	9.0	9.9	11.4	7.3	7.9	8.5	8.9	7.2	7.5	7.8	8.7
5	[Zn(II)L]Cl ₂	9.0	10.1	11.3	13.2	9.4	11.1	12.2	14.1	8.4	8.9	9.8	11.6

Figure 1: Elemental Analysis

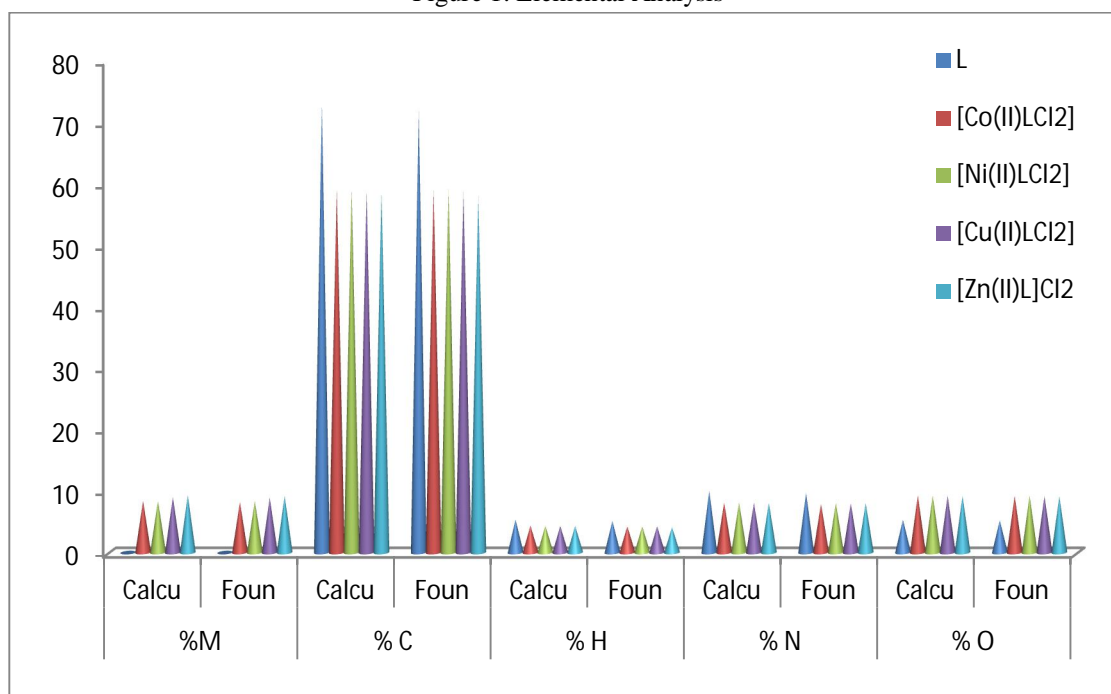


Figure 2: ^1H NMR Spectrum

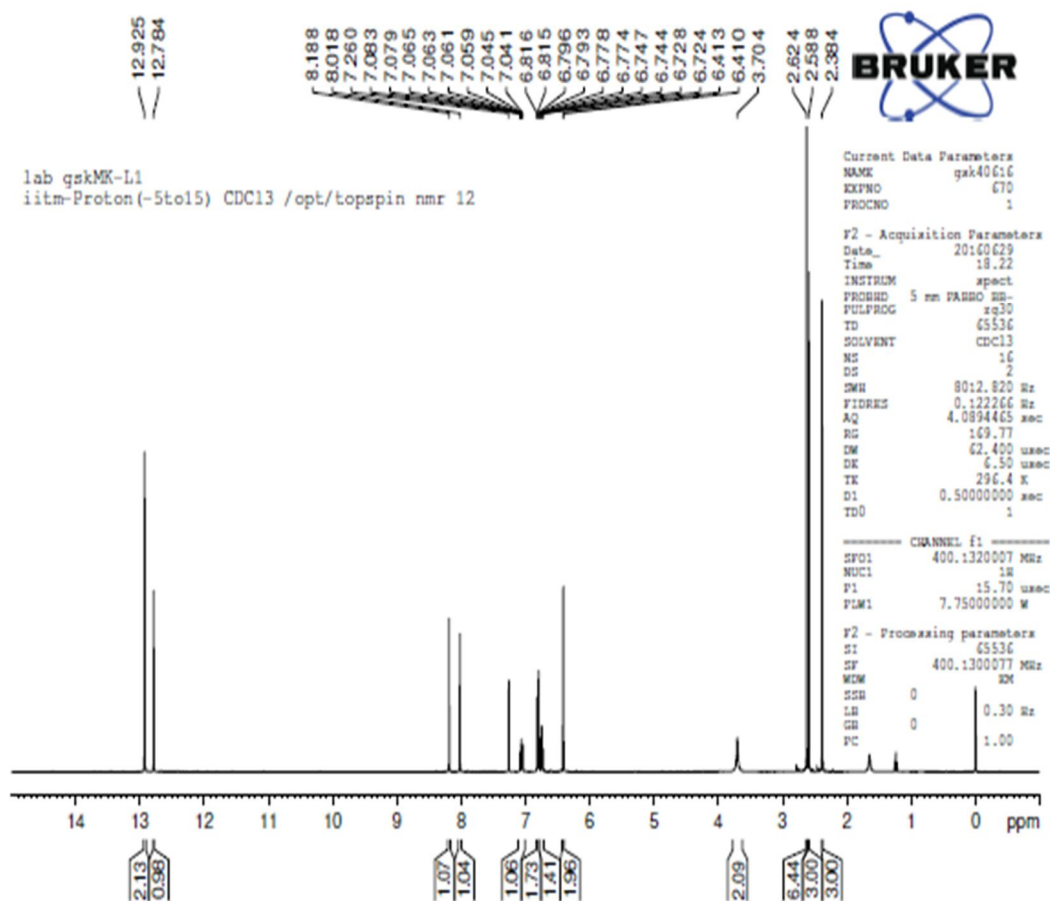


Figure 3: ^{13}C NMR Spectrum

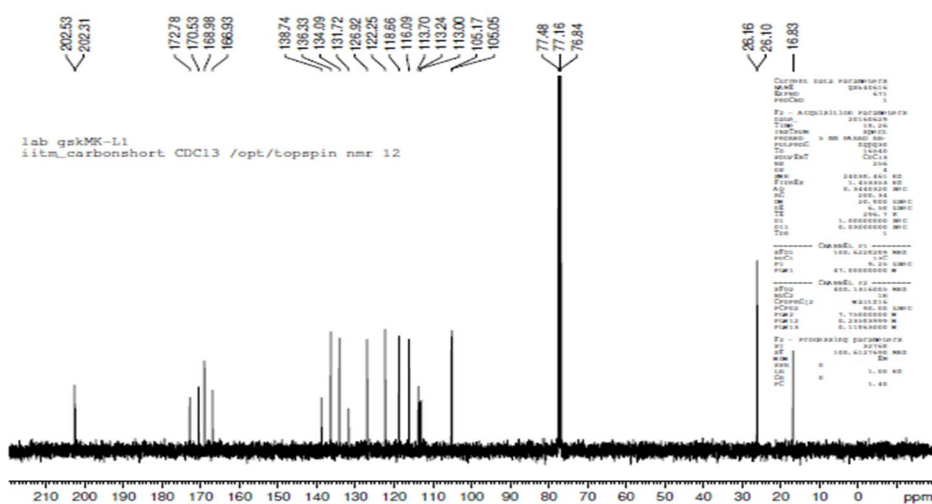


Figure 4: IR Spectrum

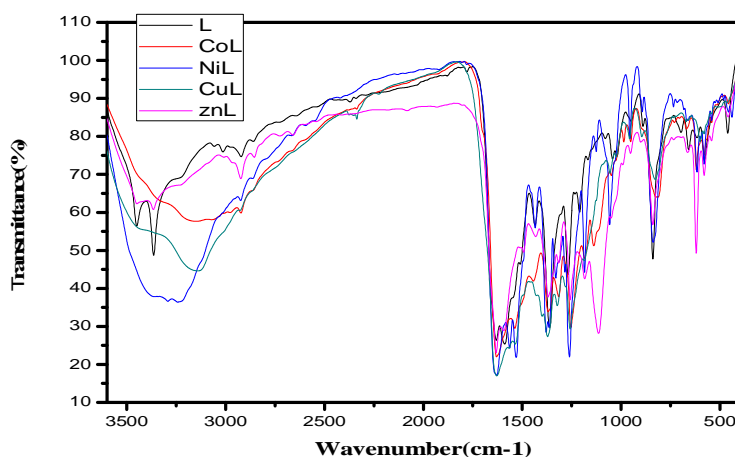


Figure 5: Electronic spectrum

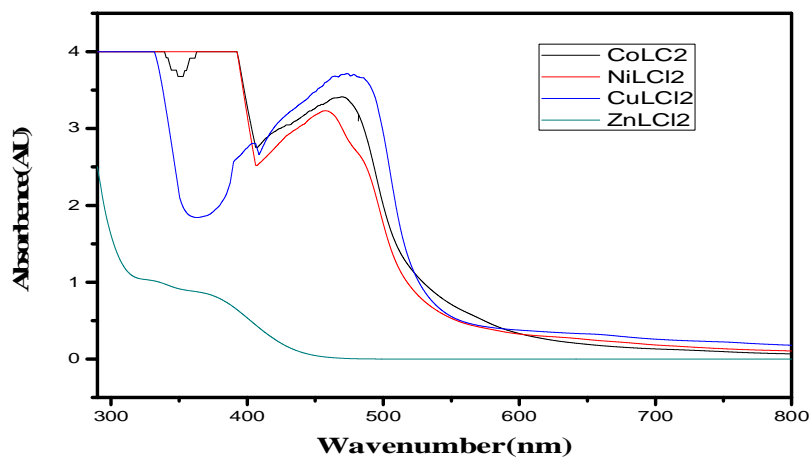


Figure 6: ESR Spectra of the Cu (II) Complexes

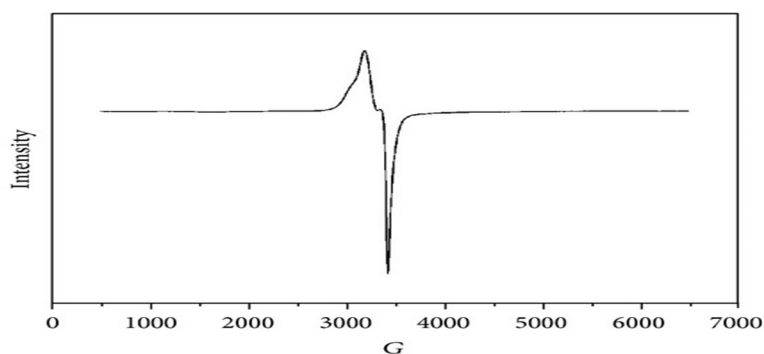


Figure 7: Cyclic Voltammetry

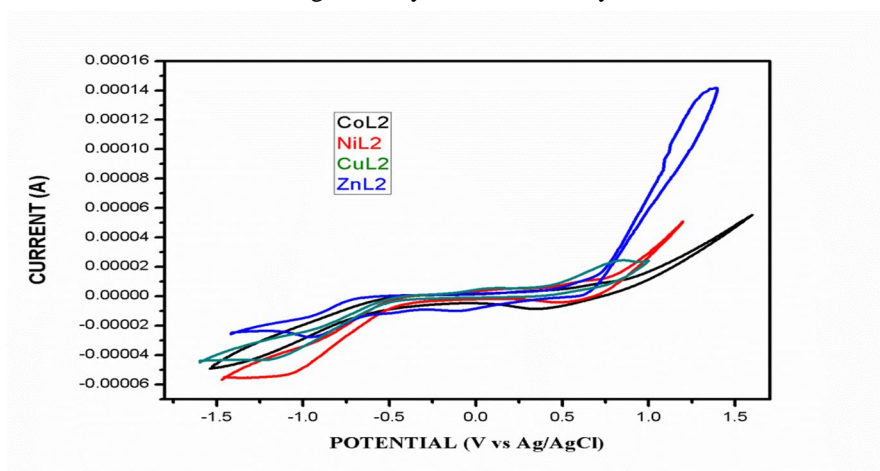
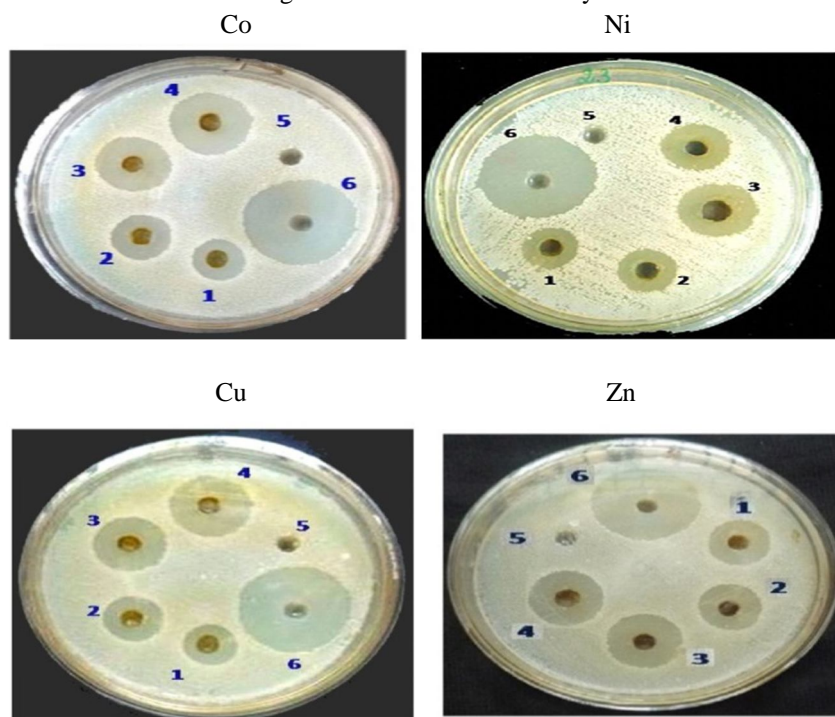
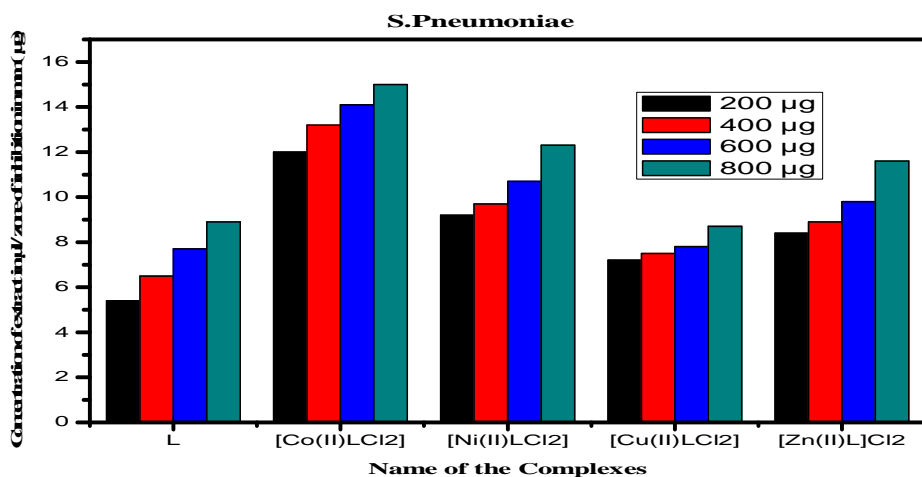
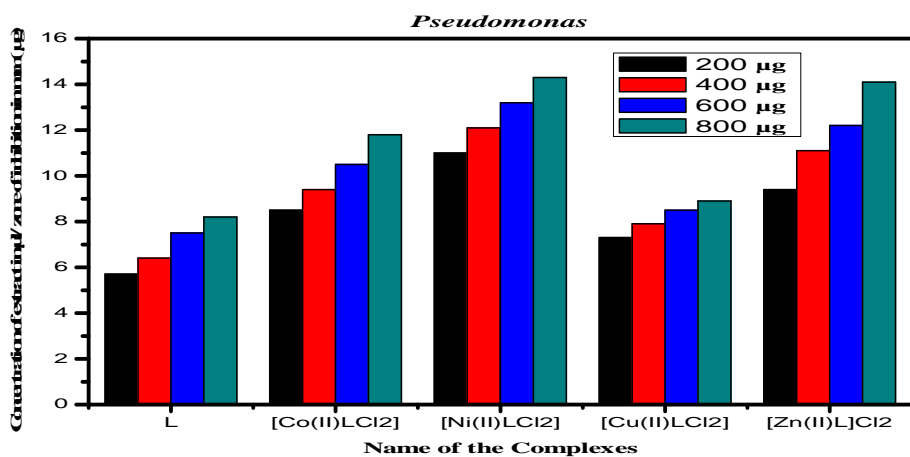
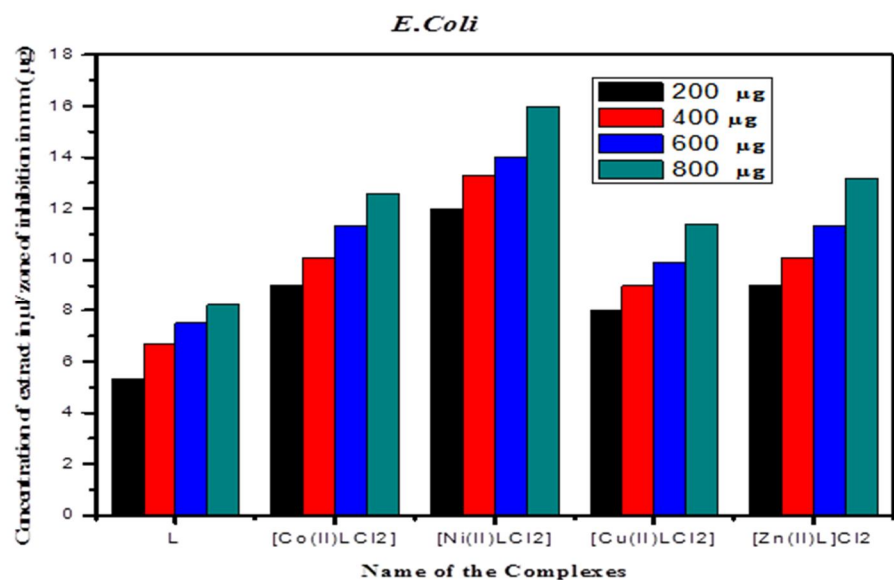


Figure 8: Antibacterial Activity





IV. CONCLUSION

The results of complexes Co (II), Ni (II) and Cu (II) are non-ionic in nature, high spin octahedral and paramagnetic. The complex of Zn (II) is ionic in nature, high spin tetrahedral and diamagnetic. All the metal complexes are very good antibacterial activity when increase the concentration of complexes. The cyclic voltammograms of these all the macrocyclic complexes were show in quasireversible wave couples peaks. The complexes of transition metal antibacterial activity against to the E.coli and Pseudomonas bacteria Ni (II) complex is more activity than other complexes similar the S.pneumoniae bacteria Co (II) complex is more activity than other complexes due to the stability and activity of the complexes.

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