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## Synthesis, molecular modeling and biological activity of zinc(II) salts with 1,4-bis(benzimidazol-2-yl)benzene

G.Krishnamurthy

Department of Chemistry, Sahyadri Science College (Autonomous), Kuvempu University,  
Shimoga - 577 203, Karnataka, (INDIA)

E-mail : gkmnaik\_sahyadri@yahoo.co.in

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### ABSTRACT

Zinc(II) halides and perchlorate react with 1,4-bis(benzimidazol-2-yl)benzene (L) in 1:2 molar ratio in n-butanol/2-methoxy ethanol at refluxing temperature to produce white/pale yellow colored complexes of the formulae  $[ZnCl_2L]H_2O$ ,  $[ZnBr_2L]3H_2O$  and  $[Zn(OCIO_3)_2L]HOCH_2CH_2CH_2CH_3$ . Zinc(II) iodide also reacts with L in 1:2 molar ratio in n-butanol ( $HOCH_2CH_2CH_2CH_3$ ) to yield white colored complex of the formula  $[ZnL_2(OCH_2CH_2CH_2CH_3)_2]$ . The complexes were characterized by elemental analysis, molar conductance measurements, thermal analysis, IR,  $^1H$  NMR,  $^{13}C$  NMR spectral studies. The complexes showed significant anthelmintic activity. The minimum energy configuration has been obtained for the zinc complexes using molecular modeling pro plus; a tool developed by ChemSW, Inc, U.S.A.

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### KEYWORDS

Zinc complexes;  
Benzimidazole;  
IR and NMR spectra.

### INTRODUCTION

Transition metal complexes containing bis-benzimidazolyl derivatives have been extensively studied. Several of these benzimidazolyl derivatives could modify their reactivity towards metal ions<sup>[1]</sup>. Further, several of their metal complexes may serve as the potential models to mimic the active centers of metalloenzymes<sup>[2]</sup>. Zinc(II) is responsible for the function of carboxy peptidase A and carbonic anhydrase in biological systems<sup>[3]</sup>. Thus, the coordination behaviour of benzimidazoles with zinc is of considerable importance. We report here the synthesis and characterization of zinc(II) complexes containing 1,4-bis(benzimidazol-2-yl)benzene (Figure L-1).

### EXPERIMENTAL

#### Reagents

The hydrated zinc(II) chloride was used as supplied (BDH). Terephthalic acid and o-phenylenediamine were Merck Chemicals. The solvents used were Merck chemicals and they were purified according to literature methods<sup>[4]</sup>. Hydrated zinc(II) bromide, zinc(II) iodide and zinc(II) perchlorate salts were obtained by dissolving the metal, followed by filtration and careful evaporation nearly to dryness under reduced pressure.

#### Drugs

Albendazole (BANDY, Mankind Pharma Ltd., New Delhi), Piperazine citrate, Tween 80.

## Measurements

C, H and N analyses were carried out on a Carlo Erba microanalyser. IR spectra (in nujol) were recorded on a Nicolet 4000D spectrophotometer. Molar conductivity measurements were made with  $10^{-3}$  M solution in dimethylformamide (DMF) using a digital (SYSTRONICS) conductivity meter-304 with a conventional dip type conductivity cell with a cell constant  $1.00\text{cm}^{-1}$ . NMR spectra were recorded (in DMSO- $d_6$ ) were recorded on a Bruker WH-270 or AMX-400 MHz spectrometer using TMS as the internal standard. The thermograms were recorded on a Shimadzu Thermal Analyzer DT30 at a heating rate of  $5^\circ\text{C}$  / minute.

## Preparation of the ligand

The ligand was prepared according to the literature method<sup>[5]</sup>. Terephthalic acid (10 m mole) was stirred with o-phenylenediamine (20 m mole) in syrupy phosphoric acid (20ml) at  $240^\circ\text{C}$  for four hours. The colored melt was poured into cold water (500ml) with stirring to obtain a blue-green colored precipitate. The precipitate was neutralized with 10% aqueous sodium carbonate solution. The resulting solid was recovered by filtration and recrystallized from ethanol to obtain a pale pink compound. (Yield 50%).

## Preparation of the complexes

### $[\text{ZnCl}_2\text{L}]n\text{H}_2\text{O}$ (X = Cl, n = 1; X = Br, n = 3)

To a solution of zinc(II) halide (1 m mol) dissolved in 2-methoxyethanol/ n-butanol (10ml), the ligand (2 m mol) in the same solvent (15ml) was added and the mixture was refluxed for 6-8 hours, followed by concentration of the mixture under reduced pressure, during which time a buff/white/pale-yellow solid separated.

TABLE 1 : Physical properties and analytical data of the complexes

Complex	Color	M.P ( $^\circ\text{C}$ )	$\Delta_M^a \Omega^{-1}$ $\text{cm}^2\text{mol}^{-1}$	Found(calcd.%)		
				C	H	N
$[\text{ZnCl}_2\text{L}]\text{H}_2\text{O}$	buff	>260	17	51.3 (51.7)	3.6 (3.5)	11.6 (12.1)
$[\text{ZnBr}_2\text{L}]\text{3H}_2\text{O}$	pale-yellow	>260	25	41.3 (42.0)	3.8 (3.2)	8.6 (9.0)
$[\text{ZnL}_2(\text{OCH}_2\text{CH}_2\text{CH}_2\text{CH}_3)_2]$	white	>260	16	68.8 (69.3)	4.7 (5.6)	14.0 (13.5)
$[\text{Zn}(\text{OCIO}_3)_2\text{L}]\text{HOCH}_2\text{CH}_2\text{CH}_2\text{CH}_3$	yellow	>250	13	43.9 (44.4)	4.2 (3.7)	9.5 (8.6)

<sup>a</sup> Molar conductance of  $\sim 10^{-3}$  M solutions around  $25^\circ\text{C}$  in DMF.

This was filtered, washed with petroleum benzene  $40-60^\circ\text{C}$  (20ml) and dried in a vacuum (yield 65%).

### $[\text{ZnL}(\text{OCH}_2\text{CH}_2\text{CH}_2\text{CH}_3)_2]$

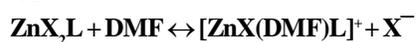
To a solution of hydrated zinc(II) iodide (1 m mol) in n-butanol (20ml), then the ligand (2 m mol) in n-butanol (20ml) was added. The resulting mixture was refluxed for 4-6 hours. The solution was concentrated under reduced pressure when a white colored solid separated. The solid was filtered, washed with petroleum benzene  $40-60^\circ\text{C}$  (20ml) and dried in a vacuum (yield 60-70%).

### $[\text{Zn}(\text{OCIO}_3)_2\text{L}]\text{HOCH}_2\text{CH}_2\text{CH}_2\text{CH}_3$

Zinc(II) perchlorate (1 m mol) was dissolved in n-butanol (5ml), then the ligand (2 m mol) in n-butanol (10ml) was added. The resulting mixture was refluxed for 6 hours when a pale yellow solid separated. This solid was filtered, washed with petroleum benzene  $40-60^\circ\text{C}$  (20ml) and dried in a vacuum (yield 60%).

## RESULTS AND DISCUSSION

The physical properties and analytical data of the complexes are listed in TABLE 1. The complexes are insoluble in common organic solvents but are soluble in DMF and DMSO and show low conductivity in DMF. This may arise from the replacement of the halide by DMF in solution and the existence an equilibrium of the type below<sup>[6]</sup>.



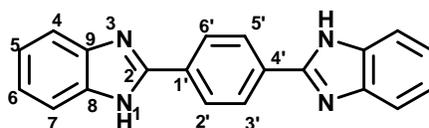
The IR spectra (in nujol mull) of the complexes are similar to the spectra of the uncoordinated N-heterocycle expect for minor shifts in the positions of some of the bands and some changes in their intensities due to coordination. The complexes displayed  $\nu\text{N-H}$  band in the range  $3150-3180\text{cm}^{-1}$  and this increased by  $10-30\text{cm}^{-1}$  as compared to that of the uncoordinated ligand. The  $\nu\text{C}=\text{N}$  and  $\nu\text{C}=\text{C}$  vibrations are very close to each other and occur around  $1616\text{cm}^{-1}$  as weak bands in the spectra of uncoordinated heterocycle and have been observed to shift by about  $10-15\text{cm}^{-1}$  on complexation. The shift in the position of  $\nu\text{C}=\text{N}$  and  $\nu\text{C}=\text{C}$  are suggestive of coordination of the N-heterocycle via the tertiary nitrogen of the imidazole moiety<sup>[7]</sup>. The  $\nu\text{O-H}$  of water of hydration<sup>[8]</sup> is observed around  $3400\text{cm}^{-1}$ . A

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TABLE 2 : <sup>1</sup>H-NMR spectral data of the ligand and complexes in DMSO-d<sub>6</sub> (δ in p.p.m)

Compound <sup>a</sup>	H-2'	H-3'	H-5'	H-6'	H-4	H-5	H-6	H-7	N-H
L	8.35s	8.35s	8.35s	8.35s	7.41d	7.23m	7.25m	7.69d	13.05s
[ZnCl <sub>2</sub> L]H <sub>2</sub> O	8.35s	8.35s	8.35s	8.35s	7.72d	7.24t	7.24t	7.56d	13.04s
[ZnBr <sub>2</sub> L].3H <sub>2</sub> O	8.43s	8.43s	8.43s	8.43	7.76q	7.41m	7.41m	7.75q	13.05
[ZnL <sub>2</sub> (OCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> ) <sub>2</sub> ]	8.34s	8.34	8.34	8.34	7.68d	7.55t	7.23t	7.23t	13.03s
[Zn(OCIO <sub>3</sub> ) <sub>2</sub> L]HOCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>	8.36s	8.36s	8.36s	8.36	7.65b	7.25s	7.25	7.65b	13.04
	(0.01)	(0.01)	(0.01)	(0.01)	(0.24)	(0.02)	-	(-0.04)	(-0.01)

<sup>a</sup>spectra recorded in dmsO-d<sub>6</sub>, s = singlet, d = doublet, t = triplet, m = multiplet, b = broad. \*values in the parentheses are coordination induced shifts (c.i.s) = δ<sub>complex</sub> - δ<sub>ligand</sub>.

TABLE 3 : <sup>13</sup>C-NMR spectral data of the ligand and complexes (in δ p.p.m)

Compound <sup>a</sup>	C-1'	C-2'	C-6'	C-2	C-4	C-5	C-6	C-7	C-8	C-9
L	131.19s	126.92s	126.92s	143.94s	111.41s	121.85s	119.01s	122.81s	143.94s	131.19s
[ZnL <sub>2</sub> (OCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> ) <sub>2</sub> ]	131.17s	126.91s	126.91s	143.92s	111.41s	121.85s	119.00s	122.81s	150.61s	132.17s
	(-0.02)	(-0.01)	(-0.01)	(-0.02)	(0.00)	-	(-0.01)	(0.0)	(6.67)	(0.98)
[Zn(OCIO <sub>3</sub> ) <sub>2</sub> L]HO(CH <sub>2</sub> ) <sub>3</sub> CH <sub>3</sub> ]	130.25s	127.16s	127.16s	137.50s	114.91s	123.01s	114.91	123.01	149.87s	130.25s
	(-0.84)	(0.24)	(0.24)	(-6.44)	(3.50)	(1.16)	(-4.10)	(0.20)	(5.93)	(-0.94)

<sup>a</sup> spectra recorded in dmsO-d<sub>6</sub>, s = singlet, d = doublet, t = triplet, m = multiplet, b = broad. \* values in the parentheses are coordination induced shifts (c.i.s) = δ<sub>complex</sub> - δ<sub>ligand</sub>.

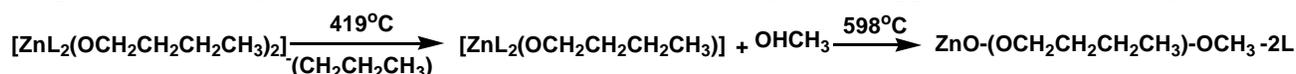
strong band at 1574cm<sup>-1</sup> and a band around 1550cm<sup>-1</sup> are assigned to N-H in-plane bending vibrations of ligand and the complexes respectively. The νC-N and δN-H vibrations are probably very close to one another and occur at 1320cm<sup>-1</sup>. The band due to p-disubstituted benzene ring vibrations occur around 1300, 1250 and 760cm<sup>-1</sup>. The bands due to benzimidazole ring vibrations are located around 1280, 1010 and 960cm<sup>-1</sup>. The assignments are tentative and are based on the literature reports on related compounds<sup>[7]</sup>. However, in the infrared spectrum of [Zn(OCIO<sub>3</sub>)<sub>2</sub>L]HO(CH<sub>2</sub>)<sub>3</sub>CH<sub>3</sub> complex, in addition to the ligand bands, the peaks around 1100 and 622cm<sup>-1</sup> of ν<sub>3</sub> and ν<sub>4</sub> of perchlorate are also observed. The former band is split indicating the presence of perchlorates are coordinated to the metal ion<sup>[9]</sup>.

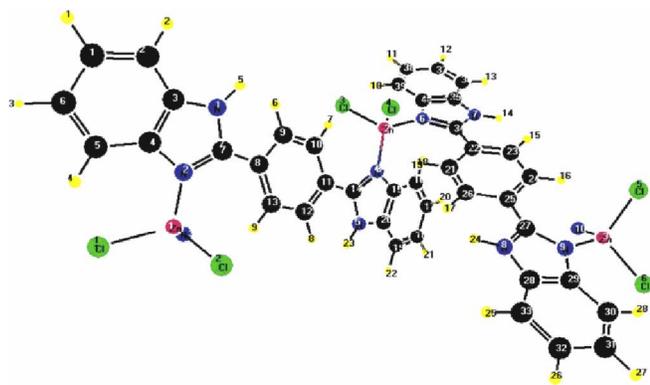
The <sup>1</sup>H NMR spectra of the complexes recorded in DMSO-d<sub>6</sub> exhibit resonances due to coordinated N-heterocycle. The spectral data of the complexes along with the assignments are compiled in TABLE 2. The spectra of the complexes are almost similar to those

TABLE 4 : Molecular modeling data

Compound	Total energy (kJ/mol)	Strain on metal (%)	Molar volume (cm <sup>3</sup> /mol)
Ligand (A)	334.824	-	264.9268
[ZnCl <sub>2</sub> L]H <sub>2</sub> O (B)	969.36	1Zn 33.46 2Zn 41.38 3Zn 34.88	709.4789
[ZnBr <sub>2</sub> L].3H <sub>2</sub> O (C)	946.26	1Zn 32.19 2Zn 39.25 3Zn 39.25	695.4039
[ZnL <sub>2</sub> (OCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> ) <sub>2</sub> ] (D)	836.262	Zn 61.49	716.2531
[Zn(OCIO <sub>3</sub> ) <sub>2</sub> L]HOCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> (E)	975.156	Zn 48.24	458.4137

of the ligand, except for a slight shift in the positions of the signals. The X-ray crystal structure study of the ligand has been established by Fingli Bei and coworkers<sup>[5]</sup>, that it has centro symmetric. The molecule is twisted in such a way that the part of the molecule is in the plane opposite to the plane of the other part. The N-H resonance signal occurs at 13.0ppm. The protons of the benzene ring, which are almost equivalent, give resonance signal at 8.3ppm, the resonance signals due to

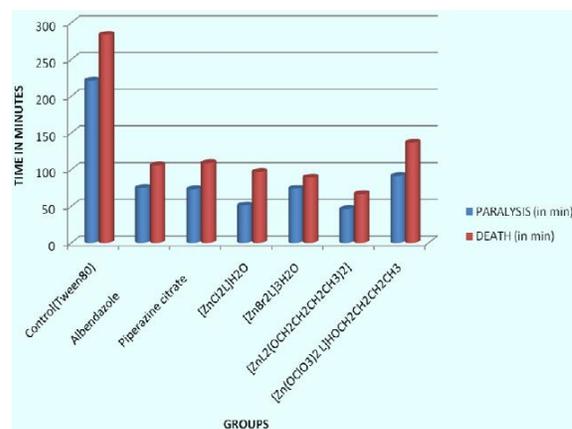




### Molecular model I : Minimum energy configuration of $[ZnCl_2L]H_2O$

protons of benzimidazole ring are found in the range  $\delta$  7.2-7.8ppm<sup>[10]</sup>. In addition to the ligand resonance signals, the resonance signals due to the protons of n-butanol are also observed in  $[ZnL_2(OCH_2CH_2CH_2CH_3)_2]$  and  $[Zn(OCIO_3)_2L]HOCH_2CH_2CH_2CH_3$  complexes. The resonance signal due to  $-CH_3$  protons of n-butanol are located at 0.86ppm and signal due to  $-CH_2-$  protons are observed at 1.3ppm. The protons of  $-OCH_2-$  group show resonance signal at 4.31ppm. The presence of solvent molecules in the complexes was confirmed by comparison with the spectrum of n-butanol in DMSO- $d_6$ . The coordination-induced shifts for various protons of the coordinated ligands are in the range 0.01-0.46ppm.

The  $^{13}C$  NMR spectra of the ligand,  $[ZnL_2(OCH_2CH_2CH_2CH_3)_2]$  and  $[Zn(OCIO_3)_2L]HOCH_2CH_2CH_2CH_3$  complexes recorded in DMSO- $d_6$  reveal distinct resonances that are in agreement with the expected carbon environments and the data are collected in TABLE 4. The assignments of the signals are made comparison with the reported values for benzimidazole and substituted benzimidazoles<sup>[11]</sup>. The  $^{13}C$  spectra for the chloro and bromo complexes could not be recorded due to their poor solubility in DMSO- $d_6$ . The resonance signal due to C-1' carbon is observed around 135.0ppm. The resonance signal at 126.0ppm is assigned to C-2' and C-6' carbons. The resonances due to aromatic carbons of benzimidazole ring are found in the range 119.0 to 150.6ppm. The resonance due to C-8 is observed around 150.0ppm and has shown positive coordination induced shift of 6.0ppm. The resonance signal due to C-9 carbon is observed around 130.0ppm. In addition to the ligand resonance signals, the peaks due to n-butanol are also observed. The reso-



nance signal due to  $CH_3$  is found at 13.8ppm. The carbon atom of the two  $-CH_2$  groups n-butanol is resonates at 18.6 and 34.6ppm. The carbon of  $-CH_2-O-$  is observed at 60.3ppm. Both positive and negative coordination induced shifts are observed in the spectra of the complexes due to the ligand to metal  $\sigma$ -donation and metal to ligand  $\pi$ -donation respectively<sup>[12]</sup>.

Thermo gravimetric analysis data of  $[ZnL_2(OCH_2CH_2CH_2CH_3)_2]$  complex has shown that there is a loss of a part of the solvent molecule ( $CH_3CH_2CH_2$ ) (0.58%, theoretical value 4.4% and found 5.1%) around 419°C. The weight loss due to 1.42% solvent molecule ( $OCH_2-$  and  $OCH_2CH_2CH_2CH_3$ ) and two molecules of N-heterocycle takes place in the temperature around 600°C, which corresponds to a theoretical loss of 92.1% (found 93.4%). The final step of the decomposition corresponds to the formation of ZnO<sup>[13]</sup>.

Molecular modeling studies were carried out with an interactive graphics molecular program<sup>[14]</sup>. Energy minimization was repeated several times to obtain the global minimum. The Leonnard - Jones equation was applied on M-N bond to obtain a configuration with minimum repulsion and hence minimum steric strain. A representative example of the structure of the complex  $[ZnCl_2L]H_2O$ , with minimum energy configuration is shown in molecular model I. After global minimum configuration is attained, the total energy of the molecule in kJ/mol, percentage strain on the metal atom and selected bond lengths, bond angles have been computed. The values are given in the TABLE 4 and 5.

Based on above discussion, the chloro, bromo and perchlorate-complexes are proposed to possess tetrahedral geometry. In the case of the perchlorate ion is monodentately coordinated to the metal ion as supported

TABLE 5 : Molecular modeling data

Compound	Bond length (Å)			Bond angle (Degrees)								
A	-	-	-	-	-	-	-	-	-	-		
	1Zn-2N	1.76	2Zn-6N	1.79	2N-1Zn-3N	109.30	2N-1Zn-1Cl	110.94	2N-1Zn-2Cl	109.30	3N-1Zn-2Cl	109.26
	1Zn-1Cl	2.10	2Zn-4Cl	2.10	3N-1Zn-1Cl	108.96	1Cl-1Zn-2Cl	109.07	4N-2Zn-3Cl	108.45	4N-2Zn-4Cl	110.62
	1Zn-3N	1.82	3Zn-9N	1.76	4N-2Zn-6N	110.85	6N-2Zn-3Cl	110.64	6N-2Zn-4Cl	107.28	9N-3Zn-10N	109.90
B	1Zn-2Cl	2.10	3Zn-5Cl	2.10	9N-3Zn-5Cl	109.93	9N-3Zn-6Cl	110.07	10N--3Zn-9N	109.90	10N-3Zn-5Cl	109.92
	2Zn-4N	1.77	3Zn-1O	1.82	5Cl-3Zn-6Cl	109.41						
	2Zn-3Cl	2.10	3Zn-6Cl	2.10								
	1Zn-2N	1.76	2Zn-4N	1.78	2N-1Zn-1Br	110.94	3N-1Zn-2Br	109.30	3Br-2Zn-7N	111.25	9N-3Zn-10N	110.07
	1Zn-1Br	2.28	2Zn-3Br	2.28	2N-1Zn-2Br	109.30	3N-1Zn-2Br	109.24	4Br-2Zn-7N	105.26	10N-3Zn-5Br	110.06
C	1Zn-3N	1.82	2Zn-7N	1.79	2N-1Zn-3N	109.31	4N-2Zn-3Br	107.01	9N-3Zn-5Br	110.04	10N-3Zn-6Br	109.87
	1Zn-2zr	2.27	2Zn-4Br	2.28	3N-1Zn-1Br	108.74	4N-2Zn-4Br	111.23	9N-3Zn-6Br	110.08	5Br-3Zn-6Br	106.67
					4N-2Zn-7N	111.23						
D	Zn-1N	1.75	Zn-2O	1.78	1N-Zn-2N	109.45	1N-Zn-1O	72.53	1O-An-2O	110.83	1O-Zn-2N	110.80
	Zn-1O	1.77	Zn-2N	1.78	1N-Zn-2O	108.16						
E	Zn-N	1.79	Zn-2O	1.75	N-Zn-1O	98.379	N-Zn-3O	141.88	1O-Zn-3O	94.25	1O-Zn-2O	138.06
	Zn-1O	1.76	Zn-3O	1.78	N-Zn-2O	95.25						

TABLE 6 : Anthelmintic activity of synthesized complex compounds with standard

Sl. No.	Group	Paralysis (in min)	Death (in min)
1	Control(Tween80)	221.17±0.48	283.67±1.05
2	Albendazole	74.83±0.60	105.67±0.67
3	Piperazine citrate	72.83±0.87	108.83±0.60
4	[ZnCl <sub>2</sub> L]H <sub>2</sub> O	51.00±1.18	96.83±0.48
5	[ZnBr <sub>2</sub> L]3H <sub>2</sub> O	73.67±1.02	89.00±0.58
6	[ZnL <sub>2</sub> (OCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub> ) <sub>2</sub> ]	46.00±0.86	66.50±0.62
7	[Zn(OCIO <sub>3</sub> ) <sub>2</sub> L]HOCH <sub>2</sub> CH <sub>2</sub> CH <sub>2</sub> CH <sub>3</sub>	91.00±0.97	136.50±0.76

by IR spectrum. The molecular modeling suggests that the N-heterocycle bridges between the two zinc metal ions. In the case of [ZnL<sub>2</sub>(OCH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub>)<sub>2</sub>] complex, <sup>1</sup>H and <sup>13</sup>C NMR spectral studies and the thermo gravimetric data show the presence of two solvent molecules i.e., n-butanol, which are coordinated to the metal ion through oxygen atom and the complex is also proposed to have tetrahedral geometry around the metal ion.

#### Anthelmintic activities of synthesized complex

Synthesized complexes were dissolved separately in minimum amount of Tween 20 and then volume was adjusted to 25ml with dextrose solution. All solutions were freshly prepared before starting the experiment. Four groups of six earthworms each were released into 25ml of desired formulation as follows.

1. Group I (Vehicle): 5% Tween 80 in dextrose solution.
2. Group II (Albendazole): 125mg
3. Group III (Piperazine citrate): 125mg
4. Group IV ([ZnCl<sub>2</sub>L]H<sub>2</sub>O): 125mg
5. Group VI ([ZnBr<sub>2</sub>L]3H<sub>2</sub>O): 125mg
6. Group VII ([ZnL<sub>2</sub>(OCH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub>)<sub>2</sub>): 125mg
7. Group VIII ([Zn(OCIO<sub>3</sub>)<sub>2</sub>L]HOCH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub>): 125mg

Observations were made for the time taken to paralysis and death time of individual worm. Paralysis was said to occur when the worm did not revive even in normal saline. Death was considered when worms lost their body colour and even they will not react when they are exposed to steam. This shows the death of the earthworm. The data revealed that synthesized complex compounds showed significant anthelmintic activity at 5mg/ml concentrations. The results are comparable with standard drugs Albendazole and piperazine citrate at same concentration. TABLE 6 reveals that almost all complexes showed a significant activity compared to the standard drug and out of four synthesized complexes, [Zn(OCIO<sub>3</sub>)<sub>2</sub>L]HOCH<sub>2</sub>CH<sub>2</sub>CH<sub>2</sub>CH<sub>3</sub> complex showed the better anthelmintic activity.

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