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Synthesis, Characterization and Biological Screening of Alkylene Dithiophosphate Derivatives of Macrocyclic Complexes of Pb (II)

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Alkylenedithiophosphate derivatives of macrocyclic complexes of Pb (II), having N_4S_4 potential donors, of the general formula, $[Pb(L)\{S_2P(OR)_2\}_2]$ where L=macrocyclic ligands L^1, L^2, L^3, L^4 and L^5; R= C_2H_5-, $C_3H_7^n$ or $C_3H_7^i$ have been synthesized from the reaction of $[Pb(L)X_2$(where X= Cl^-, NO_3^- or CH_3COO^-) with sodium Alkylenedithiophosphate in 1:2 molar ratios in THF. These complexes have been characterized by elemental analysis, molar conductance, molecular weight determinations, IR, ^1H, ^{13}C NMR. Macrocyclic Complexes of Pb (II) derivatives have been studied by screening them Aspergillus flavus, Fusarium oxysporum, Alternaria alternata and bacteria like Salmonella typhi and Bacillus subtili. Alkylenedithiophosphate derivatives were found to be more fungitoxic and antibacterial than their corresponding macrocyclic complexes.

Keywords: Macrocyclic complexes; alkylenedithiophosphate; microwave.

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1. INTRODUCTION

The chemistry of macrocyclic ligands is a fascinating area of intense study for inorganic chemists. The possibility to tailor make different types of macrocycles for specific use has promoted much of this interest. Among others, these include for biological systems, therapeutic reagents for the treatment of metal intoxication, synthetic ionophores and the selective extraction of heavy and precious metals [1-4]. In spite of vast innovation in macrocyclic chemistry and tremendous interest in mixed ligand complexes, no mixed ligand macrocyclic complex was reported till our publications. Alkylene dithiophosphate has been the area of our thrust since last 3 decades [5-14]. Considering the importance of mixed ligand macrocyclic complexes, we reported synthesis, characterization, antimicrobial of Cr(III), Mn(II), Fe(III), Co(III), Ni(II), Cu(II), Cd(II), Sn(II) and Pb(II) with dialkylene and alkylene dithiophosphate having N_2S_2 potential donors in 14 to 20 membered rings[15-32]. We have also reported the macrocyclic complexes of Ni(II), Sn(II), Sr(II) and Ba(II) with dialkylene and alkylene dithiophosphate having N_4S_4 potential donors in 22-28 membered rings [17,24, 29-32]. In our earlier publication we have reported the synthesis, characterization and biological screening of alkylene dithiophosphate derivatives macrocyclic coplexes of Pb(II) in 14-20 member rings [18]. In continuation to the above work we hereby report the synthesis, characterization and antimicrobial screening of alkylene dithiophosphate derivatives of macrocyclic complexes of Pb(II) having N_4S_4 potential donors in 22 to 28 membered rings.

2. EXPERIMENTAL SECTION

All the lead salts and dicarboxylic acids of A.R. grade were obtained from S. D. fine chemicals, Mumbai and were used without further purification. o-Amino thiophenol was used as obtained from Merck. Solvents were purified and dried by standard methods. The chelating ligand bis-(2-aminophenyl) disulphide was synthesized by the dimerization of the o-amino thiophenol by H_2O_2 as reported in the literature [33]. Alkylene dithiophosphoric acids were prepared by the reactions of various alcohols like ethanol, normal propanol and iso propanol with phosphorus Penta sulphide. Phosphorus Penta sulphide was added slowly in about 2 hours to the anhydrous alcohol heated on a water bath. After complete addition of phosphorus Penta sulphide, reactants were warmed till the evolution of hydrogen sulphide gas ceased. Solvents were removed under reduced pressure and the Alkylenedithiophosphoric acid thus obtained was purified by distillation under reduced pressure.

Sodium salts of Alkylenedithiophosphoric acids were prepared by the reaction of Alkylenedithiophosphoric acids with corresponding sodium alkoxide in equimolar ratio. To the sodium alkoxide (prepared by dissolution of sodium metal in excess of parent alcohol) was added drop by drop, the benzene solution of Alkylenedithiophosphoric acid in 1:1 molar ratio. The reaction was exothermic; however, for the sake of completion of reaction the contents were warmed for about 1 hour. Solvent was removed under reduced pressure and the white solid thus obtained was washed with benzene and finally dried under reduced pressure, yielding a white crystalline solid.

2.1 Physical Measurements

Microanalyses for carbon, hydrogen, nitrogen and sulphur were determined from SICART, Vallabh Vidyanagar. Lead and phosphorus were estimated by standard method [34]. The molecular weights were determined by Rast Camphor method. Infrared data were recorded on a Perkin-Elmer FT-IR spectrophotometer as KBr pellets. ^{1}H and ^{13}C NMR spectra were recorded on a Jeol 270 MHz spectrometer using DMSO-d_6 as a solvent and TMS as an internal standard.

2.1.1 Synthesis of Precursor Macrocyclic Complexes Tetra benzo [2,3,9,10,13,14,20,21] [4,8,15,19] Tetraaza [1,11,12,22] Tetra thiacyclodiicosane [5,7,16,18] Tetraone}

Solution of lead chloride (1.621 g, 5 mmol) in methanol was reacted with bis-(2-aminophenyl) disulphide (2.566g, 10 mmol) dissolved in methanol. This was followed by the addition of a methanolic solution of malonic acid (1.075 g, 10 mmol). Reaction mixture was refluxed for 6 hours. The light yellow precipitate obtained was filtered, washed with methanol and dried under vacuum. (Found: C, 39.82; H, 2.65; N, 6.19; Cl, 7.74; S, 14.15; Pb, 22.90%). Calcd. For $C_{30}H_{24}N_4S_4O_4Cl_2Pb$ (fw): C, 39.56; H, 2.64; N, 6.15; Cl, 7.80; S, 14.07; Pb, 22.75%) m.p.241^{o}C; yield 3.47g(74%).

2.1.2 Synthesis of Alkylenedithiophosphate Derivative of Macrocyclic Complexes

Macrocyclic complex mentioned above (1.201 g, 1.3 mmol) was dissolved in THF and was reacted with methanolic solution of ammonium butylene dithiophosphate (0.532g, 2.6 mmol) in 1:2 molar ratio. Reaction mixture was refluxed for 6-8 hours at 200°C. On cooling the yellow crystals of dithiophosphate derivative were separated out, which were filtered through G-3 filtering funnel. This crude product was washed several times with methanol, by vigorous shaking in filtration funnel, to remove the ammonium chloride formed during the reaction. Product was dried under vacuum and was crystallized with THF / C_2H_5OH mixture.

Reaction of lead salts with bis-(2-aminophenyl) disulphide and various dicarboxylic acids in 1:2:2 molar ratio in methanol takes place in the following manner:

The above macrocyclic complexes of Pb(II) in THF react with a methanolic solution of sodium Alkylene dithiophosphate in 1:2 molar ratios to afford the Alkylenedithiophosphate derivatives of the macrocyclic Pb(II) complexes in the following manner:

$$[Pb(L^{1-5})X_2]+2NaS_2P(OR)_2 \quad Pb(L^{1-5})\{S_2P(OR)_2\}_2] + 2NaX$$

R= C_2H_5-, $C_3H_7^n$ or $C_3H_7^i$
L = Macrocyclic ligands L_1, L_2, L_3, L_4 and L_5
L_1= Macrocyclic ligand derived from bis-(2-amino phenyl) disulphide and malonic acid (n=1), 22- membered ring;oi {Tetra benzo[2,3,9,10,13,14,20,21][4,8,15,19]tetraaza [1, 11, 12, 22] tetra thiacyclodiicosane [5, 7, 16,18] tetraone}.

L_2= Macrocyclic ligand derived from bis-(2-aminophenyl) disulphide and succinic acid (n=2), 24- membered ring;

{Tetrabenzo[2,3,10,11,14,15,22,23][4,9,16,21]tetraaza[1,12,13,24]tetrathiacyclotetraicosane[5,7,17,20]tetraone}.

L_3=Macrocyclic ligand derived from bis-(2-aminophenyl) disulphide and glutaric acid (n=3), 26- membered ring;

{Tetrabenzo[2,3,11,12,15,16,24,25][4,10,17,23]tetraaza[1,13,14,26]tetrathiacyclohexaicosane[5,9,18,22]tetraone}.

L4=Macrocyclic ligand derived from bis-(2-aminophenyl) disulphide and adipic acid (n=4), 28- membered ring.

{Tetrabenzo[2,3,12,13,16,17,26,27][4,11,18,25]tetraaza[1,14,15,28]tetrathiacyclooctaicosane[5,10,19,24]tetraone}.

L_5=Macrocyclic ligand derived from bis-(2-amino phenyl) disulphide and phthalic acid ($(CH_2)_n$ = *o*-C_6H_4-), 24- membered ring;
{Hexabenzo[2,3,6,7,10,11,14,15,18,19,22,23][4,9,16,21]tetraaza[1,12,13,24]tetrathiacyclotetraicosane[5,8,17,20]tetraone}.

The derivatives of macrocyclic complexes of the following alkylene dithiophosphoric acids have been synthesized. Physical Properties and Analysis of Alkylene dithiophosphoric Acids has been given in Table - 1.

2.1.3 Synthesis of precursor macrocyclic complexes tetra benzo [2,3,9,10,13,14,20,21] [4,8,15,19] tetraaza [1,11,12,22] tetra thiacyclodiicosane [5,7,16,18] tetraone}

Solution of lead chloride (1.621 g, 5 mmol) in methanol was reacted with bis-(2-aminophenyl) disulphide (2.566g, 10 mmol) dissolved in

methanol. This was followed by the addition of a methanolic solution of malonic acid (1.075 g, 10 mmol). Reaction mixture was refluxed for 6 hours. The light yellow precipitate obtained was filtered, washed with methanol and dried under vacuum. (Found: C, 39.82; H, 2.65; N, 6.19; Cl, 7.74; S, 14.15; Pb, 22.90%). Calcd. For C30H24N4S4O4Cl2.Pb (fw): C, 39.56; H, 2.64; N, 6.15; Cl, 7.80; S, 14.07; Pb, 22.75%) m.p.241^oC; yield 3.47g(74%). All derivatives are yellow or Light-Yellow, Yellow in colour, which melts with at high temperature [35-45]. The Physical properties of these derivatives are given in Table-1.

2.1.4 Synthesis of alkylenedithiophosphate derivatives of macrocyclic complexes

Macrocyclic complex Tetrathiacyclodiicosane [5, 7, 16, 18] Tetraone (1.201 g, 1.3 mmol) was dissolved in THF and was reacted with methanolic solution of ammonium butylene dithiophosphate (0.532g, 2.6 mmol) in 1:2 molar

ratio. Reaction mixture was refluxed for 6 hours at reflux temp on cooling the yellow crystals of dithiophosphate derivative were separated out, which were filtered through G-3 filtering funnel. This crude product was washed several times with methanol, by vigorous shaking in filtration funnel, to remove the ammonium chloride formed during the reaction. Product was dried under vacuum and was crystallized with THF / C_2H_5OH mixture.

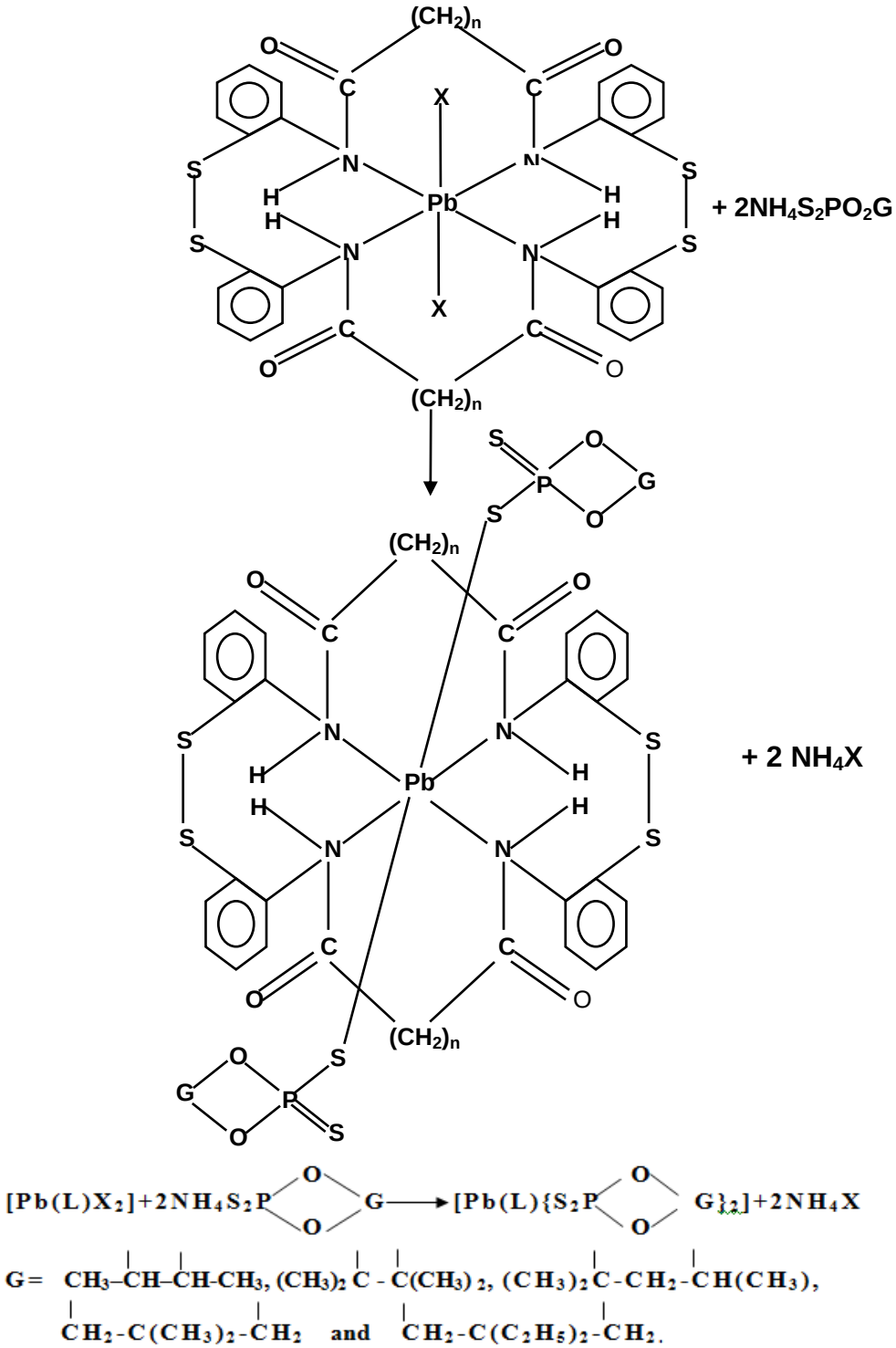

Fig. 1 General structure of macrocyclic complexes of Pb (II)

Where n = 1, 2, 3, 4 or $(CH_2)_n$ = o-C_6H_4- ; Pb (II), X=Cl^-, NO_3^-, CH_3CHOO^-

2.1.5 Synthesis of Alkylenedithiophosphate Derivatives of Macrocyclic Complexes

To an aqueous solution (10 cm^3) of Macrocyclic complex Tetrathiacyclodiicosane [5,7,16,18] Tetraone (1.201 g,1.3 mmol) (0.237 g, 1 mmol was dissolved in THF and was reacted with methanolic solution of ammonium butylene dithiophosphate (0.532g, 2.6 mmol) in 1:2 molar ratio. To this hot solution, an aqueous solution (10 cm^3) of an amino acid (HL) was (1 mmol) with constant stirring. The reaction mixture was kept in the microwave for about 10-13 min. The complexes were obtained by raising the pH of the reaction mixture by adding a diluted $NaHCO_3$ solution.The solid complex obtained was filtered and washed with Water-ethanol. A comparison of these two methods is given in Table 2.

3. RESULTS AND DISCUSSION

The reaction mixture was refluxed for 3 h. on cooling the crystals of the dithiophosphate derivatives separated out. Except THF and DMSO, these derivatives are insoluble in almost all organic solvents. The molar conductance of 10-3 M solution in DMSO lie in the range 03-06 ohm-1cm2mol-1, showing that these complexes are non-electrolyte. The molecular weight determinations indicate their monomeric nature (Table-3).

Table 1. Comparison Colour, molecular weight, decomposition temperature and pH of the lead complexes

Sr. No	Empirical formula of Complexes	Molecular Weight	Colour	Melting Point ^0C	pH
1	$C_{38}H_{40}N_4O_8P_2S_8Pb$	1222 (1205)	Yellow	224	5.2
2	$C_{42}H_{48}N_4O_8P_2S_8Pb$	1251 (1261)	Light Yellow	218	6.6
3	$C_{46}H_{56}N_4O_8P_2S_8Pb$	1302 (1317)	Light Yellow	228	6.4
4	$C_{44}H_{52}N_4O_8P_2S_8Pb$	1299 (1289)	Yellow	236	5.8
5	$C_{52}H_{52}N_4O_8P_2S_8Pb$	1371 (1385)	Yellow	224	5.7

Table 2. Comparison between microwave and thermal method

Sr. No	Empirical formula of Complexes	Thermal Method		Microwave Method	
		% Yield	Time in Hrs	% Yield	Time in minutes
1	$_{38}H_{40}N_4O_8P_2S_8Pb$	65	7	97	13
2	$_{42}H_{48}N_4O_8P_2S_8Pb$	60	6	94	12
3	$_{46}H_{56}N_4O_8P_2S_8Pb$	56	7	91	16
4	$_{44}H_{52}N_4O_8P_2S_8Pb$	67	6	98	14
5	$_{52}H_{52}N_4O_8P_2S_8Pb$	62	7	93	12

Table 3. Elemental analysis data, molar conductance and magnetic moment of lead complexes

Sr. No.	Emperical Formula	Elemental analysis found (calculated)				Molar conductance (Mhos. cm^2. mol^{-1})	μeff (B.M.)
		M %	C %	H %	N %		
1	$C_{38}H_{40}N_4O_8P_2S_8Pb$	16.93 (17.18)	37.31 (37.84)	3.27 (3.32)	4.58 (4.65)	04	2.68
2	$C_{42}H_{48}N_4O_8P_2S_8Pb$	16.55 (16.41)	40.29 (39.96)	3.84 (3.80)	4.48 (4.44)	04	2.73
3	$C_{46}H_{56}N_4O_8P_2S_8Pb$	15.90 (15.72)	42.40 (41.91)	4.30 (4.25)	4.30 (4.25)	03	2.77
4	$C_{44}H_{52}N_4O_8P_2S_8Pb$	15.93 (16.06)	40.65 (40.96)	4.00 (4.03)	4.31 (4.34)	03	2.80
5	$C_{52}H_{52}N_4O_8P_2S_8Pb$	15.10 (14.95)	45.51 (45.05)	3.79 (3.75)	4.08 (4.04)	03	2.83

4. APPLICATION

4.1 Antimicrobial Activity

Like their precursor macrocyclic complexes, the antifungal activity of alkylene dithiophosphate derivatives has been tested against three fungi, Aspergillus flavus, Fusarium oxysporum and Alternaria alternata The screening data for the average percentage inhibition of the fungi at 100, 125 and 200 ppm concentration are given in Table-4. The values obtained suggest that the alkylene dithiophosphate derivatives of macrocyclic complexes are more fungitoxic than their precursor macrocyclic complexes as well as the alkylene dithiophosphoric acids. Further the data also indicate that with the increase in the concentration, the fungitoxicity also increases.

The antibacterial activity against two bacteria, namely S.typhi and B.subtili, were tested by the inhibition zone technique[46]. The data obtained are presented in Table-5. The values suggest that the alkylene dithiophosphate derivatives of macrocyclic complexes are more antibacterial than their precursor macrocyclic complexes (PbL_1-PbL_5).

4.2 Magnetic Studies

Magnetic moments of the metal complexes calculated from the measured magnetic susceptibilities after employing diamagnetic corrections. The observed μ_{eff} values presented in Table 3. Suggest the octahedral geometry for nickel complexes. Study also shows paramagnetic nature of synthesized complexes.

4.3 Infrared Spectral Data

In the macrocyclic complexes, the four bands in the region 1680-1638(s), 1582-1516(m), 1272-1240(s) and 690-648(w) cm^{-1} have been ascribed to the amide I, amide II, amide III and amide IV in-plane deformation vibrations, respectively [47]. A broad band in the region 3189-3104 cm^{-1} has been assigned to the ν(N-H) vibration of the secondary amino group. These bands do not show any significant change from

Table 4. Antifungal Activity of Alkylene Dithiophosphate Derivatives of Macrocyclic Complexes of Pb(II) Average % of Inhibition After 72 h at 30 ± 2 °C

Sr. No	Complexes	*Aspergillus flavus*			*Fusarium oxysporum*			*Alternaria alternata*		
		100	**125**	**200**	**100**	**125**	**200**	**100**	**125**	**200**
1	Bavistin (Standard)	91	95	99	93	97	99	93	96	99
2	Bis –amino di phenyldisulphide	41	45	49	40	44	49	42	47	50
3	$PbCl_2.2H_2O$	20	24	28	22	26	30	26	30	34
4	$C_{38}H_{40}N_4O_8P_2S_8Pb$	74	78	82	72	76	83	71	75	79
5	$C_{42}H_{48}N_4O_8P_2S_8Pb$	78	82	84	74	79	83	72	79	86
6	$C_{46}H_{56}N_4O_8P_2S_8Pb$	76	79	85	75	80	86	76	80	84
7	$C_{44}H_{52}N_4O_8P_2S_8Pb$	74	78	82	74	79	84	76	80	86
8	$C_{52}H_{52}N_4O_8P_2S_8Pb$	79	83	86	77	81	85	76	80	85

Table 5. Antibacterial Activity of Alkylene Dithiophosphate Derivatives of Macrocyclic Complexes of Pb (II). Percentage Growth Inhibition After 24 Hours at 30 + 2 ^{0}C (Conc. in ppm)

Sr. No	Complexes	*Bacillus Subtili*		*Salmonella typhi*	
		500	**1000**	**500**	**1000**
1	Streptomycin (Standard)	97	99	97	99
2	Bis –amino di phenyldisulphide	22	26	21	25
3	$PbCl_2.2H_2O$	14	16	15	17
4	($C_{38}H_{40}N_4O_8P_2S_8Pb$)	30	32	28	31
5	($C_{42}H_{48}N_4O_8P_2S_8Pb$)	28	31	29	34
6	($C_{46}H_{56}N_4O_8P_2S_8Pb$)	32	36	31	36
7	($C_{44}H_{52}N_4O_8P_2S_8Pb$)	30	35	34	40
8	($C_{52}H_{52}N_4O_8P_2S_8Pb$)	30	35	31	37

their parent macrocyclic complexes. Two bands present in the region 1072-1040 cm^{-1} and 888-840 cm^{-1} may be assigned to (P)-O-C and P-O-(C) stretching vibrations, respectively[48].The band present between 999-954 cm^{-1} may be attributed to the ring vibrations of dioxaphospholanes and dioxaphosphorinanes respectively, which are probably coupled with C-C stretching vibrations[49,50]. A weak band present in the region 570-538 cm^{-1} has been attributed to P-S symmetric and asymmetric vibrations. A strong band observed in the region 728-680 cm^{-1}, which also appears in ammonium alkylene dithiophosphate at around the same region, is attributed to the P=S moiety. This indicates the unidentate behavior of the dithiophosphate moieties[51,52]. The presence of sharp and weak bands in the region 483-418 cm^{-1} and 364-320cm^{-1} have been assigned to ν(Pb-N) and ν(Pb-S) vibrations, respectively [7,8,53].

4.4 ^{1}H NMR Spectral Data

The structure of alkylene dithiophosphate derivatives of macrocyclic complexes of Pb(II) have been further confirmed by recording the ^{1}H NMR using DMSO-d_6 as a solvent and TMS as an internal standard. In addition to the protons appear in the parent macrocyclic complexes, the additional protons of alkylene dithiophosphate moieties appear in the spectra. The methyl protons of tetramethyl ethylene moiety, butylene dithiophosphate moiety and neo-pentylene dithiophosphate moiety appeared in the range δ 1.36 to 1.59 ppm. The protons of methylene and methine moieties appear in the range δ 3.8 to 4.9 ppm. The broad singlet observed between δ 8.09 to 8.36 ppm has been assigned the proton of–C(O)NH- group. The protons of -CH_2- group of malonic acid appear as a singlet in the range, δ 3.09 to 3.42 ppm. The methylene protons of -CH2-CH2- group of succinic acid appear as a singlet in the range of δ 3.04 to 3.26 ppm. The protons of α-C atoms of glutaric acid moiety were observed as a multiplate δ 1.78 ppm and δ 1.81 ppm. The protons of β-C atoms of the above moiety appeared as a multiplet δ 1.95 ppm. The protons of α-C atoms of adipic acid moiety appeared between δ 1.81 ppm. The protons of β-C atoms appear in the range δ 1.70 to 1.82. Aromatic protons of bis-(2-aminophenyl)disulphide moiety were observed as a multiplate in the range δ 7.14 to 7.60 ppm. The values are in the expected region.[54,55].

4.5 ^{13}C NMR Spectral Data

Alkylene dithiophosphate moieties spectral data shows The carbons of –CH_3- group of tetramethylene, butylene and neopentylene dithiophosphate moieties appear in the range δ 12.14 to 14.12 ppm. The carbons of methylene and mithine moieties appear in the range δ 38.64 to 43.16 ppm. The carbon of -CH_2- group of malonic acid moiety lies in the range δ 30.86 to 34.29 ppm. The carbons of –CH_2-CH_2- moiety appear in the range δ 27.52 to 31.64 ppm. The α-carbon of glutaric acid moiety were observed in the range δ 29.76 to 30.14 ppm and the β carbons of the above moiety appear in the range δ 24.90 to 27.20 ppm respectively Signals observed at δ 170.08 to 174.62 ppm have been assigned to the carbons of >C=O group. The signals of the carbons of –C(O)NH- group appear in the range δ 80.09 to 83.48 ppm. The carbons of phenyl group of bis-(2-aminophenyl) disulphide moiety appeared in the range δ 70.49 to 74.86 ppm. The values are in the expected range [54,55].

5. CONCLUSION

The object of invention Mixed Ligand Macrocyclic Complexes of Pb (II) shows the anti-microbial activity. A process for preparation of Mixed Ligand Macrocyclic Complexes of Pb (II) under the microwave method very fast with high yield compare to the thermal process. Magnetic studies indicate the paramagnetic nature of the complexes. lead ion square-planar geometry and each alkylene Di thiophosphate moiety occupies the axial positions binding the central lead ion in a unidentate manner. The IR spectra show the bonding of the metal ion through -N and -X donor atoms of the two ligands. The biological study shows that complexes are less active against Aspergilus flavus, Fusrium oxisporum, Alternaria alternata than compared with the standard antibacterial compound Bavistin, the complexes show modest activity. The antibacterial activity of the Pb(II) complexes shows that Pb(II) complexes were less active than the free ligand against *Bacillus subtili* and *Salmonella typhi*.

CONSENT

It is not applicable.

ETHICAL APPROVAL

We conducted our research after obtaining proper IEC approval.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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