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Biologically Potent Sulphonamide Imine Complexes of Organotin(IV): Synthesis, Spectroscopic Characterization and Biological Screening

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Coordination behaviour of a biologically potent sulphonamide-imine having N^N donor moiety towards the diorgano and triorganotin(IV) have been investigated. The unimolar and bimolar reactions of di-and triorgano-tin(IV) chlorides with monobasic bidentate imine resulted in the formation of colored solids, soluble in DMSO, DMF and MeOH, which have been characterized by elemental analyses, molecular weight determinations and conductance measurements. Structures of the resulting complexes have been proposed using IR, ^{1}H, ^{13}C and ^{119}Sn NMR spectral studies. All the complexes are monomeric in nature as indicated by their molecular weight determinations. Conductivity measurements in dry DMF show them to be non-electrolytes. The pathogenicity and virulence of certain microbial infections associated with ions of the complexes have been found to be potent and like broad spectrum antibiotics. These results made it desirable to delineate a comparison between the ligand and its metal complexes. Emphasis has been given to the nematicidal properties.

Keywords: Sulphonamide imine, Organotin(IV) complexes, Fungicidal, Bactericidal and Nematicidal activities

INTRODUCTION

Organotin compounds are toxic to a variety of microorganisms and find widespread applications in biocidal compositions. In the past few years organotin compounds of the types R_3SnX and R_2SnX_2 became well known as broad spectrum biocides, toxic additive in molluscicides, fungicides and other type of pesticides [1] and appeared in the literature [2-7] dealing with various aspects of applications.

The chemistry of organotin(IV) complexes (R_mSnX_n) were extensively studied due to their biological activities [8-11]. Thiazoles have been reported to exhibit significant antifungal, antibacterial, antileishmanial, anthelmintic and analgesic activity [12-13]. Thiazole and its derivatives exhibit significant chemotherapeutic activities [14]. Significant antitubercular activity has been reported in the case of 2,4-disubstituted thiazole derivatives [15]. Organotin compounds of several types have been found to possess significant biological and pharmacological activities [16-17] and are used as fungicides [18-19], bactericides and antitumour agents [20].

Encouraged by the above findings, during the course of the present investigations, synthesis of several new tin derivatives of monofunctional bidentate 2-acetylnaphthalene and its base are reported and biological and inorganic aspects of the resulting complexes have been studied and discussed in brief.

EXPERIMENTAL

All the chemicals and solvents were dried and purified by standard methods. The reactions were carried out under strictly anhydrous conditions.

Preparation of the Ligand

The ligand was prepared by the condensation of 2-acetylnaphthalene with sulphathiazole in 1:1 molar ratio by refluxing the reacting species for 5-6 hours in alcohol on a water bath (Scheme 1). On cooling, crystals of the imine

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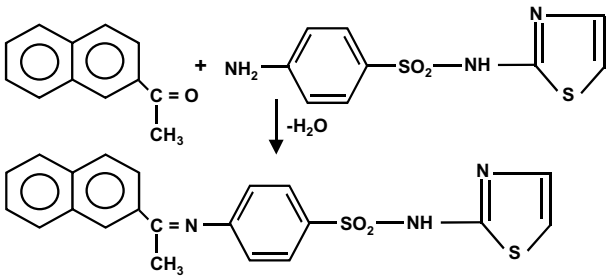


Scheme 1. Synthesis of the ligand.

separated out which were washed with ethanol, dried and recrystallized with acetone and dried *in vacuo*. The important physical and analytical are been given in Table 1.

Preparation of Tin(IV) Complexes

To a weighed amount of Ph_3SnCl (0.001 mol, 0.578 g), Ph_2SnCl_2 (0.001 mol, 0.617 g) or Me_2SnCl_2 (0.001 mol, 0.386 g), the sodium salt of the ligand (0.001 mol, 0.611, 0.732 or 0.717 g) in 1:1 molar ratio was added in dry methanol (30 ml).

For the 1:2 molar reaction, weighed amounts of Ph_2SnCl_2 (0.001 mol, 0.327 g) or Me_2SnCl_2 (0.001 mol, 0.224 g) were taken in dry methanol (30 ml). To these was added twice the amount of the sodium salt of the ligand (0.002 mol, 0.776 g or 0.833 g). The reaction mixture was refluxed for 10-14 h, during which period the white precipitate of sodium chloride separated out. The contents were cooled and the precipitate so formed was removed by filtration. The mother liquor was concentrated by removing the excess of solvent under reduced pressure and the resulting products were dried, repeatedly washed with Dry cyclohexane and methanol and finally dried *in vacuo* for 3-4 h. The physical properties and analytical data of the resulting complexes are given in Table 1.

Analytical Methods and Physical Measurements

IR spectra were recorded as KBr pellets or Nujol Mulls on a FT-IR Spectrophotometer, model Magna FT-IR-50. ^1H NMR spectra were recorded on a Jeol FX 90Q Spectrometer in DMSO-d_6 using TMS as an internal standard. ^{13}C NMR and ^{119}Sn NMR spectra were recorded in methanol on this instrument. Nitrogen and sulphur were determined by the Kjeldahl's and Messenger's methods, respectively. Tin was estimated as tin oxide and molecular weights were determined by the Rast camphor method.

RESULTS AND DISCUSSION

The reactions of Me_2SnCl_2, Ph_2SnCl_2 and Ph_3SnCl with the sodium salts of sulphonamide-imine ligand (LH) has been carried out in 1:1 and 1:2 molar ratios resulting in the isolation of $Me_2SnCl(L)$, $Ph_2SnCl(L)$, $Ph_3Sn(L)$, $Me_2Sn(L)_2$ and $Ph_2Sn(L)_2$ solids. These are monomeric and possess sharp melting points. They are soluble in methanol, chloroform, DMF and DMSO.

Table 1. Physical Properties and Analytical Data of the Complexes

Complex	Colour	M.P.	Yield	Elemental Analysis (%)						
		(°C)	(%)	C Found (Calcd.)	H Found (Calcd.)	N Found (Calcd.)	S Found (Calcd.)	Sn Found (Calcd.)	Cl Found (Calcd.)	Mol. Wt. Found (Calcd.)
LH $C_{21}H_{17}N_3O_2S_2$	White	165-167	81	61.35 (61.89)	4.15 (4.19)	9.86 (10.31)	15.24 (15.73)	-	-	380 (407.49)
$Me_2SnCl(L)$ $C_{23}H_{22}N_3SnO_2S_2Cl$	Light Yellow	119-121	72	46.40 (46.76)	3.69 (3.75)	6.90 (7.11)	10.47 (10.85)	19.76 (20.09)	5.72 (6.00)	566 (590.70)
$Me_2Sn(L)_2$ $C_{42}H_{38}N_6SnO_4S_4$	Light Brown	91-93	76	52.07 (52.39)	3.94 (3.97)	8.41 (8.72)	13.00 (13.22)	12.01 (12.32)	-	933 (962.73)
$Ph_2SnCl(L)$ $C_{33}H_{26}N_3SnO_2S_2Cl$	Mastard Yellow	94-96	79	55.08 (55.44)	3.64 (3.66)	5.58 (5.87)	8.42 (8.97)	16.18 (16.60)	4.57 (4.95)	672 (714.84)
$Me_2Sn(L)_2$ $C_{54}H_{42}N_6SnO_4S_4$	Pitch	161-163	71	59.26 (59.72)	3.84 (3.89)	7.35 (7.73)	11.38 (11.81)	10.39 (10.93)	-	1049 (1085.87)
$Ph_3Sn(L)$ $C_{39}H_{31}N_3SnO_2S_2$	Light Brown	85-87	75	61.46 (61.92)	4.07 (4.13)	5.14 (5.55)	8.11 (8.47)	15.09 (15.68)	-	728 (756.40)

$R_2SnCl_2 + N^{\frown}NNa \rightarrow R_2SnCl(N^{\frown}N) + NaCl$
$R_2SnCl_2 + 2N^{\frown}NNa \rightarrow R_2SnCl(N^{\frown}N)_2 + 2NaCl$
$Ph_3SnCl + N^{\frown}NNa \rightarrow Ph_3Sn(N^{\frown}N) + NaCl$

(where $N^{\frown}N$ is the donor system of the sulphonamide imine ligand and R = Me or Ph).

Spectral Studies

UV-Vis spectra. The important UV-Vis spectral data of the ligand and its metal complexes are s ummarized in Table 2. As seen, in the electronic spectrum of the imine, a strong and due to >C = N chromophere was observed at 362 nm, shifted to the lower wavelength region in the spectra of the complexes. Such a shift in the n-π* band is probably due to the donation of the lone pair of electrons by the nitrogen of the ligand to the central metal atom. Two bands at 240-265 and 280-263 nm related to the ligand and its complexes assigned as K bands, π-π* transitions and B-bands, π-π* transitions with red shift and hypsochromic shift respectively.

IR spectra. Several significant changes with respect to the ligand are observed in the corresponding metal complexes. A sharp band at 1637 cm^{-1} due to ν(>C = N) is shifted to the lower frequency (ca 15 cm^{-1}) in the complexes indicating the coordination of the ligand through nitrogen atom of the azomethine group. Free ligand displays absorption bands at 3130-3420 and 1610 cm^{-1} assigned to ν(N-H) [21] and δ(N-H) [21], respectively.

Two medium to sharp intensity bands observed in the far IR region of the tin complexes [22,23] at 402-412 and 353-367 cm^{-1} are assigned to ν(Sn-N) and ν(Sn-Cl) modes, respectively, which are not observed in the spectrum of the ligand. The medium to sharp intensity bands are observed at 595 and 525 cm^{-1}, which may be assigned to the asymmetric and symmetric modes of Sn-C stretching vibrations respectively.

One strong to medium intensity band appeared in the spectra of the complexes in the region 1230-1180 cm^{-1} which can be assigned to ($Sn-CH_3$) stretching vibrations.

The proposed structure is also supported by the comparatively low δ(^{119}Sn) value of the triphenyl tin complexes. A new band observed ~275 cm^{-1} may be assigned to ν(Sn-Ph). All IR data are shown in Table 3.

^{1}H NMR spectra. The proton NMR spectra of the ligand and its corresponding metal complexes were recorded in DMSO-d_6. The spectrum of the ligand exhibits signals due to –CH aromatic protons, the –NH of the sulphathiazole and -CH_3 of 2-acetylnaphthalene. The

Table 2. Important UV-Vis Spectral Data of the Ligand and Its Metal Complexes

Group	Ligand	$Me_2SnCl(L)$	$Me_2Sn(L)_2$	$Ph_2SnCl(L)$	$Ph_2Sn(L)_2$	$Ph_3Sn(L)$
n-π* λmax/nm >C=N	362	350	353	341	346	343
π-π* λmax/nm C_6H_5 ring	240	251	254	257	260	265
π-π* λmax/nm >C=N	280	276	273	270	268	263

Table 3. Important IR Absorption Bands (in cm^{-1}) of the Ligand and Its Metal Complexes

Compound	ν(NH)	ν(C=N)	δ(N-H)	ν(Sn←N)	ν(M-Cl)
Ligand	3130-3420m	1637 vs	1610 w	-	-
$Me_2SnCl(L)$	-	1626 vs	-	407w	353 m
$Me_2Sn(L)_2$	-	1629 vs	-	412w	-
$Ph_2SnCl(L)$	-	1627 vs	-	402w	367 m
$Ph_2Sn(L_2)$	-	1622 vs	-	404w	-
$Ph_3Sn(L)$	-	1624 vs	-	405w	-

M = medium, vs = very strong, w = weak

disappearance of the -NH signal of imine in the organometallic derivatives indicates coordination of the azomethine nitrogen to the metal atom. Further, in the spectra of the complexes, a downfield shift in the position of the aromatic protons also indicated the coordination of the azomethine nitrogen to the metal atom resulting in the formation of a coordinate linkage (M←N).

The ^{1}H NMR spectrum of the ligand exhibits NH protons at δ 10.66 ppm and C(CH_3) = N protons at δ 2.13 ppm.

The additional signals in the region δ 1.04-1.13 ppm are due to (CH_3Sn) and ($C6H_5$-Sn) groups, respectively. Data are shown in Table 4.

^{13}C NMR spectra. The conclusions drawn from the IR and ^{1}H NMR spectra are concurrent with ^{13}C spectral data regarding the authenticity of the proposed structures. The considerable shifts in the position of the carbon atoms adjacent to the azomethine nitrogen further support the proposed, coordination in these complexes.

^{119}Sn NMR spectra. ^{119}Sn NMR spectra recorded for the di and triorganotin(IV) derivatives exhibit sharp signals at δ-115.13-155.54 ppm and δ-335.41-362.30 ppm strongly support the penta- and hexa- coordination around the tin atom. Quantitatively δ(^{119}Sn) values depend on the coordination number [24] and the nature of the ligand, as well as on the ligand bite [25].

Thus on the basis of the results discussed and the analytical as well as the spectral data, suitable trigonal bipyramidal geometry for penta-coordinated state and octahedral geometry for hexa-coordinated state have been suggested for the 1:1 and 1:2 metal compounds (Fig. 1).

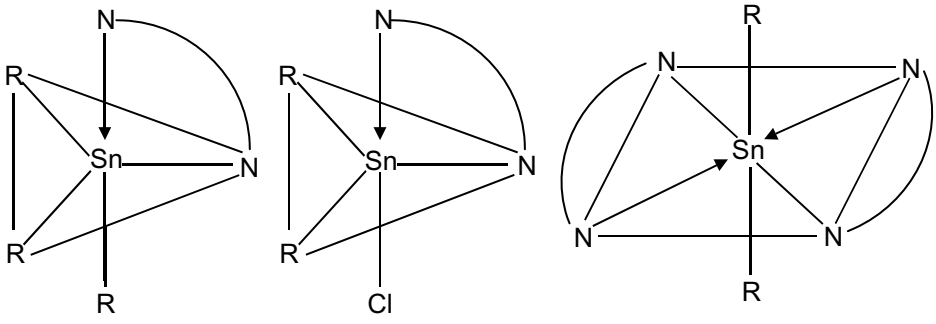
BIOCIDAL SCREENING

The ligand and its metal complexes have been screened against antibacterial and antifungal activities *in vitro*.

Table 4. ^{1}H NMR and ^{119}Sn NMR Spectral Data (δ, ppm) of the Ligand and Its Complexes

Compound	CH_3	M-CH_3/C_6H_5	NH	Aromatic Protons	^{119}Sn NMR
Ligand	2.13 (s,3H)	-	10.66 (br, 1H)	8.18-6.95 (m)	-
$Me_2SnCl(L)$	2.22 (s,3H)	1.13 (br,6H)	-	8.26-7.70 (m)	δ-115.13
$Me_2Sn(L)_2$	2.19 (s,6H)	1.04 (br,6H)	-	8.65-7.15 (m)	δ-362.30
$Ph_2SnCl(L)$	2.18 (s,3H)	8.19 (br, 10H)	-	8.43-6.95 (m)	δ-120.65
$Ph_2Sn(L)_2$	2.17 (s,6H)	8.28 (br,10H)	-	8.49-7.14 (m)	δ-335.41
$Ph_3Sn(L)$	2.15 (s,3H)	8.09 (br,15H)	-	8.45-6.94 (m)	δ-155.54

s = singlet, br = broad, m = multiplet



R = Me or Ph

Fig. 1. Proposed structures for the complexes.

Antifungal Activity (Hanging Drop Method)

This method, also known as spore germination method [26], this includes the deposition of chemicals on a slide, evaporation to dryness and addition of a drop of water containing spores of the test fungus. Under this method, we placed a drop of spore suspension on a clear slide and determined the number of spores in the drop under low power magnification and adjusted the suspension to obtain nearly ten spores per microsocpic field. These slides were incubated for 12 hours at 25 ^{o}C in petridishes working as moisture chambers. Slides were put in such a position that spore drops keep hanging. After incubation, spore germination was counted. Each treatment was replicated three times. In this technique, total number of spores, number of germinated spores and number of ungerminated spores were counted. Lethal Dose (LD_{50}) values have been calculated to compare the activity of the ligand with its chelates by plotting graphs between the concentration of ligand/complexes with the number of germinated spores.

The organisms selected for this method are: *Alternaria alternata* and *Helminthosporium gramineum*.

On the basis of the results, we concluded that comparatively, complexes are more active on the *Helminthosporium gramineum* than *Alternaria alternata* (Table 5).

Agar Plate Technique [27]

Bioefficacies of the synthesized compounds were checked *in vitro*. The *in vitro* antifungal activities of the ligand and its complexes have been evaluated against several fungi by the agar plate technique. The compounds were directly mixed with the medium in different concentrations. Controls were also run and three replicates were used in each case. The linear growth of the fungus was obtained by measuring the diameter of the fungal colony after four days. The amount of growth inhibition in each of the replicate was calculated by Eq_n (1):

$$\text{Percentage inhibition} = (C\text{-}T) \times 100/C \qquad (1)$$

where C is the diameter of the colony on the control plate and T is the diameter of the fungal colony on the test plate

Table 5. Comparison of LD_{50} Values of Ligand with Its Complexes Obtained from Antifungal Activity by Hanging Drop Method

Compound	LD_{50} value in *Alternaria alternata*	LD_{50} values in *Helminthosporium gramineum*
Ligand	135	70
$Me_2SnCl(L)$	120	55
$Me_2Sn(L)_2$	90	40
$Ph_2SnCl(L)$	100	50
$Ph_2Sn(L)_2$	80	35
$Ph_3Sn(L)$	95	45

Table 6. Average Percentage Inhibition after 96 h

Compound	Aspergillus niger			Macrophomina phaseolina			Fusarium oxysporum			Alternaria alternatea		
Conc. (ppm)	25	50	100	25	50	100	25	50	100	25	50	100
Ligand	36	47	66	36	48	65	42	55	62	44	56	64
$Me_2SnCl(L)$	42	55	70	45	56	69	43	59	70	45	59	71
$Me_2Sn(L)_2$	52	63	82	53	62	84	48	62	80	50	59	79
$Ph_2SnCl(L)$	47	58	80	46	61	83	44	59	77	49	56	76
$Ph_2Sn(L)_2$	50	62	85	52	65	87	48	63	82	53	62	86
$Ph_3Sn(L)$	49	58	81	49	66	82	47	61	79	52	60	78

(Table 6).

Antibacterial Activity

Determination of the antibacterial activity was carried out by the paper-disc plate method [28]. All the paper with a diameter of 5 mm were soaked in these solutions. These discs were placed on the appropriate medium (peptone, beaf extract, NaCl and agar-agar) previously seeded with organisms in petri dishes and stored in an incubator at 30 ± 1 °C. The inhibition zone thus formed around each disc was measured (in mm) after 24 h (Table 7).

Nematicidal Activity

Root knots, incited by *Meloidogyne spp.,* are responsible for serious yield losses in rice, pulses, compounds were dissolved in methanol; paper discs of Whatman No. 1 vegetables, sugarcane, potato, sugarbeet, tobacco and in some fruit crops, in India. Although control of root-knot nematodes have been demonstrated by cultural and physical methods as also by using resistant varieties, chemical control continues to remain the mainstay in their management [29]. The systemic and non fumigant nematicides entered the market as potential nematicides having granular formulation, low phyto-toxicity, systemic action and their ability to kill nematodes at very low doses [29]. *Meloidogyne incognita* produce golls on the roots of many host plants and responsible for 45 percent yield loss in brinjal [30].

In experiment, the solutions of test compounds in methanol of 25, 50 and 100 ppm concentrations were prepared. Eggmasses from heavily infected brinjal roots were washed under running water. Step by step procedure was adopted for obtaining the pure quantities of *Meloidogyne incognita* eggs, viz., cutting the clean root, addition of 1% NaOCl solution, shaking it and then sieving through 150 and 400 mesh sieves [31]. Hatching of nematodes in simple water were counted 230 eggs per egg mass. It is called control plate and then we treated egg mass with different concentration of chemical solution and each experiment was repeated three times. The temperature range was fixed at 30 $\pm$ 2 ^{o}C. In the end we observed the comparative results and concluded that in increasing chemical treatment, minimum hatching and other side in control plate maximum hatching were recorded (Table 8).

Table 7. Diameter of Inhibition Zone (mm) after 24 h

Compound	E-coli (-)		Klebsiella aerogenous (-)		Pseudomonas cepacicola (-)		Staphylococcus aureus (+)	
Conc. (ppm)	500	1000	500	1000	500	1000	500	1000
Ligand	7	10	7	11	11	13	11	14
$Me_2SnCl(L)$	8	11	9	12	13	16	13	15
$Me_2Sn(L)_2$	11	13	13	15	18	18	15	17
$Ph_2SnCl(L)$	11	13	11	14	15	17	15	17
$Ph_2Sn(L)_2$	14	16	14	17	17	19	17	19
$Ph_3Sn(L)$	12	16	12	16	16	18	16	18

Table 8. Hatching (%) of *Meloidogyne Incognita* in Different ppm (Solutions)

Compound	25	50	100
Ligand	22	17	13
$Me_2SnCl(L)$	18	14	nil
$Me_2Sn(L)_2$	15	11	nil
$Ph_2SnCl(L)$	16	13	
$Ph_2Sn(L)_2$	12	9	
$Ph_3Sn(L)$	14	12	nil

All the complexes along with the parent ligand have been tested and the results revealed that activity is increased on complexation.

CONCLUSION

Knowledge of the mechanism of the action of compound is important from a purely scientific point of view. Here we have distinguished three different methods by which complexes can exert their action.

1. The effect of resonating structure [32], such as potentially reactive groupings. If toxicity is depended on one or more chemical reactions, then any molecule which would increase the rate of chemical reactions must, perforce, enhance toxicity.

2. The introduction of a lipophilic substituent, either aryl or alkyl, often conferred toxicity as did the substitution of polar groups [33].

3. Complexes having amido groups or reactive halogen atoms tend to hydrolyse to form compounds, which have modified activity spectrum. The halogen replaced by hydroxyl ion and as a result of slight alkaline pH the increase in activity was observed.

The microbial activity of the complexes and ligand showed that the former are more active than its parent ligand. The data in Tables IV-VII reveal that $Ph_2Sn(L)_2$ was found to be more toxic than the other complexes, and the hexa-coordinated complexes display better results than the penta-coordinated complexes.

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