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Full Length Research Paper

Synthesis, characterization and cytotoxic assay on human liver carcinoma cells (HepG2) of organodistannoxane dimer complexes derived from alkylaminobenzoic acids

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A total number of five organodistannoxane dimer complexes derivative of alkylaminobenzoic acids have been successfully synthesized and characterized quantitatively and qualitatively. Results of the infrared spectroscopy of the acids and complexes showed that the coordination took place via oxygen atoms from the carboxylate group. This indicated that the carboxylate anions acted as mono- and bidentate ligand. From the ^{119}Sn NMR solution study, complexes 1 and 5 exhibited two well resolved signals, one lying in the range of five-coordinated geometry and the other in the range of six-coordinated geometry. Moreover, all the exo- and endo-cyclic tin atoms in complexes 2-4 were five-coordinated indicated that a pair of carboxylate anions was bonded to the tin atoms in a monodentate manner while the other two carboxylate anions were bonded to the tin atoms (endo and exo-cyclic tin atom) in a bridging bidentate manner. From the cytotoxic assay study, complex 1 revealed a significant result compared to complexes 2-5.

Key words: Organodistannoxane dimer, synthesis, characterization, cytotoxic assay.

INTRODUCTION

Although the first organotin(IV) compound was successfully isolated in 1850s, it did not gain any commercial significance in industrial applications until almost a hundred years later (Blunden et al., 1985). Since then, the study of organotin(IV) complexes received considerable attention due to the vast applications in industrial as well as its biological properties against bacterial, fungal and cancer cells line (Gielen et al., 2000; Mahmood et al., 2003, 2004; Xanthopoulou et al., 2008; Hanif et al., 2010; Win et al., 2010a; Win et al., 2010b). Up to date, there are many well-documented structures on complexes isolated as organodistannoxane dimer (Li et al., 2006; Win et al., 2008; Win et al., 2010c).

The core geometry of the organodistannoxane dimer complexes consists of a centrosymmetric planar Sn_2O_2 group bonded to the exo- and endocyclic tin(IV) atom moiety via the bridging oxygen atoms so that the oxygen atoms are tri-coordinated (Li et al., 2006; Win et al., 2008, 2010c).

In this paper, we report on the synthesis and structural characterization of organodistannoxane dimer derived from alkylaminobenzoic acids. Moreover, the *in vitro* cytotoxic assay on human liver carcinoma cells, HepG2 of the complexes was reported herein.

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MATERIALS AND METHODS

General and instrumental

All the reagents, starting materials as well as the solvents were purchased commercially and used without any further purification. The melting points were determined in an open capillary and were uncorrected. Elemental C, H and N analyses were carried out on a Perkin-Elmer 2400 CHN Elemental Analyzer. Tin was determined gravimetrically by igniting a known quantity of each complex to SnO_2. Infrared spectra were recorded using a Perkin-Elmer System 2000 FTIR Spectrophotometer as a KBr disc in the frequency range of 4000-400 cm^{-1}. The spectra for ^{1}H, ^{1}H-^{13}C HMQC and ^{119}Sn NMR were recorded on a Bruker AC-P 400 MHz FTNMR Spectrometer and ^{13}C NMR was recorded on a Bruker AC-P 300MHz FTNMR Spectrometer using deuterated $CDCl_3$ as the solvent and tetramethylsilane, TMS as the internal standard.

In vitro cytotoxic assay

The *in vitro* cytotoxic assay was carried out against human liver carcinoma cells line, HepG2. The cells were maintained in Eagle's minimum essential medium (MEM) supplemented with 2 mM of L-glutamine, 1 mM of sodium pyruvate, 0.1 mM of non-essential amino acid, 1.5 µg/mL sodium bicarbonate, 100 IU/mL penicillin and 100 µg/mL streptomycin. The cytotoxicity assay was determined using the microtitration 3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) assay (Mosmann, 1983; Ali et al., 2000). The assay of each concentration for each compound was performed in triplicate. The fraction of surviving cells was measured relative to the untreated cell population by measuring the absorbance values at 570 nm with a reference at 630 nm using an ELISA microplate reader (Bio Tek EL 340, USA) (Ali et al., 2000). Cytotoxicity was expressed as fifty percent cytotoxic dose (IC_{50}) that is the concentration causing 50% inhibition of cell growth with reference to the control (untreated cells). The IC_{50} and the S.E.M. (standard error of the mean) was determined using Probit Analysis (SPSS, version 12.0.1).

Preparation of sodium salts

The sodium salts of the acids were obtained by heating under reflux a 1:1 molar mixture of sodium hydroxide, NaOH (3 mmole) with the respective acids (3 mmole) in ethanol (50 mL) for two hours. After a few days, white precipitates were obtained.

Synthesis of complexes

Bis[2-(methylamino)benzoato]tetrabutyldistannoxane(IV) dimer, $[\{2\text{-}(NHCH_3)C_6H_4COO(C_4H_9)_2Sn\}_2O]_2$ (1): The title complex was obtained by heating under reflux a 1:2 molar mixture of dibutyltin(IV) oxide (0.75 g, 3 mmole) and 2-(methylamino)benzoic acid, 2-$(NHCH_3)C_6H_4COOH$ (0.91 g, 6 mmole) in methanol (50 mL) for two hours. A clear brown transparent solution was separated by filtration and kept in a bottle. After a few days, brown crystals (2.87 g, 61.2% yield) were collected. Melting point: 115.6-116.5 °C. Analysis for $C_{64}H_{104}N_4O_{10}Sn_4$: C, 49.27; H, 6.12; N, 3.58; Sn, 30.26%. Calculated for $C_{64}H_{104}N_4O_{10}Sn_4$: C, 49.14; H, 6.70; N, 3.58; Sn, 30.35%.

Bis[4-(methylamino)benzoato]tetrabutyldistannoxane(IV) dimer, $[\{4\text{-}(NHCH_3)C_6H_4COO(C_4H_9)_2Sn\}_2O]_2$ (2): Bis[4-(methylamino)benzoato]tetrabutyldistannoxane(IV) dimer was obtained by heating under reflux a 1:1 molar mixture of dibutyltin(IV) oxide (0.49 g, 2 mmole) and 4-(methylamino)benzoic acid, 4-$(NHCH_3)C_6H_4COOH$ (0.30 g, 2 mmole) in methanol (50 mL) for two hours. A clear yellow solution was collected by filtration and kept in a bottle. After five days, yellow crystals (2.40 g, 76.8% yield) were obtained. Melting point: 216.8-217.5°C. Analysis for $C_{64}H_{104}N_4O_{10}Sn_4$: C, 49.79; H, 6.45; N, 3.64; Sn, 29.83%. Calculated for $C_{64}H_{104}N_4O_{10}Sn_4$: C, 49.14; H, 6.70; N, 3.58; Sn, 30.35%.

Bis[4-(dimethylamino)benzoato]tetrabutyldistannoxane(IV) dimer, $[\{4\text{-}[N(CH_3)_2]C_6H_4COO(C_4H_9)_2Sn\}_2O]_2$ (3): Complex 3 was prepared by a similar method to those described for complex 2, except substituting 4-(methylamino)benzoic acid with 4-(dimethylamino)benzoic acid, 4-$[N(CH_3)_2]C_6H_4COOH$. Ethanol (50 mL) was used as solvent and the mixture was heated under reflux for three hours. A clear transparent solution was isolated by filtration and kept in a bottle. After five days, colourless crystals (2.10 g, 64.8% yield) were collected. Melting point: 226.7 to 227.3°C. Analysis for $C_{68}H_{112}N_4O_{10}Sn_4$: C, 50.60; H, 6.77; N, 3.43; Sn, 29.08%. Calculated for $C_{68}H_{112}N_4O_{10}Sn_4$: C, 50.40; H, 6.97; N, 3.46; Sn, 29.30%.

Bis[3-(dimethylamino)benzoato]tetrabutyldistannoxane(IV) dimer, $[\{3\text{-}[N(CH_3)_2]C_6H_4COO(C_4H_9)_2Sn\}_2O]_2$ (4): This title complex was prepared by similar method to those described for complex 2, except substituting with 3-(dimethylamino)benzoic acid, 3-$[N(CH_3)_2]C_6H_4COOH$ and the reaction was heating under reflux for three hours. After five days, brown crystals (2.14 g, 66.1% yield) were collected. M.p.: 137.3-138.2°C. Analysis for $C_{68}H_{112}N_4O_{10}Sn_4$: C, 50.62; H, 6.52; N, 3.44; Sn, 29.53%. Calculated for $C_{68}H_{112}N_4O_{10}Sn_4$: C, 50.40; H, 6.97; N, 3.46; Sn, 29.30%.

Bis[4-(diethylamino)benzoato]tetrabutyldistannoxane(IV) dimer, $[\{4\text{-}[N(C_2H_5)_2]C_6H_4COO(C_4H_9)_2Sn\}_2O]_2$ (5): Complex 5 was obtained by heating under reflux a 1:1 molar mixture of dibutyltin(IV) oxide (0.49 g, 2 mM) and 4-(diethylamino)benzoic acid, 4-$[N(C_2H_5)_2]C_6H_4COOH$ (0.39 g, 2 mM) in ethanol (50 mL) for two hours. A clear transparent solution was isolated by filtration and kept in a bottle. After five days, colourless crystals (2.96 g, 85.3% yield) were collected. Melting point: 208.3 to 209.9°C. Analysis $C_{76}H_{128}N_4O_{10}Sn_4$: C, 52.78; H, 7.07; N, 3.19; Sn, 27.37%. Calculated $C_{76}H_{128}N_4O_{10}Sn_4$: C, 52.68; H, 7.45; N, 3.23; Sn, 27.41%.

RESULTS AND DISCUSSION

Physical and elemental analysis

In this study, complexes 1-5 derived of alkylaminobenzoic acids have been obtained in solid state. The micro-elemental analysis for C, H, N and Sn data obtained were in agreement with the predicted formula for complexes 1 to 5. Complexes 1 to 5 gave a sharp melting point indicated the isolation of fairly pure complexes. An outline of the proposed structure for complexes 1 to 5 is depicted in Figure 1.

Infrared and NMR spectral studies

The ν(O-H) bands which appeared in the range of 2899 to 2491 cm^{-1} for the parent acids were absent from the infrared spectra of the sodium salts and complexes 1 to 5, indicating the deprotonation and coordination of the carboxylate anions to the tin atom moiety (Table 1). The

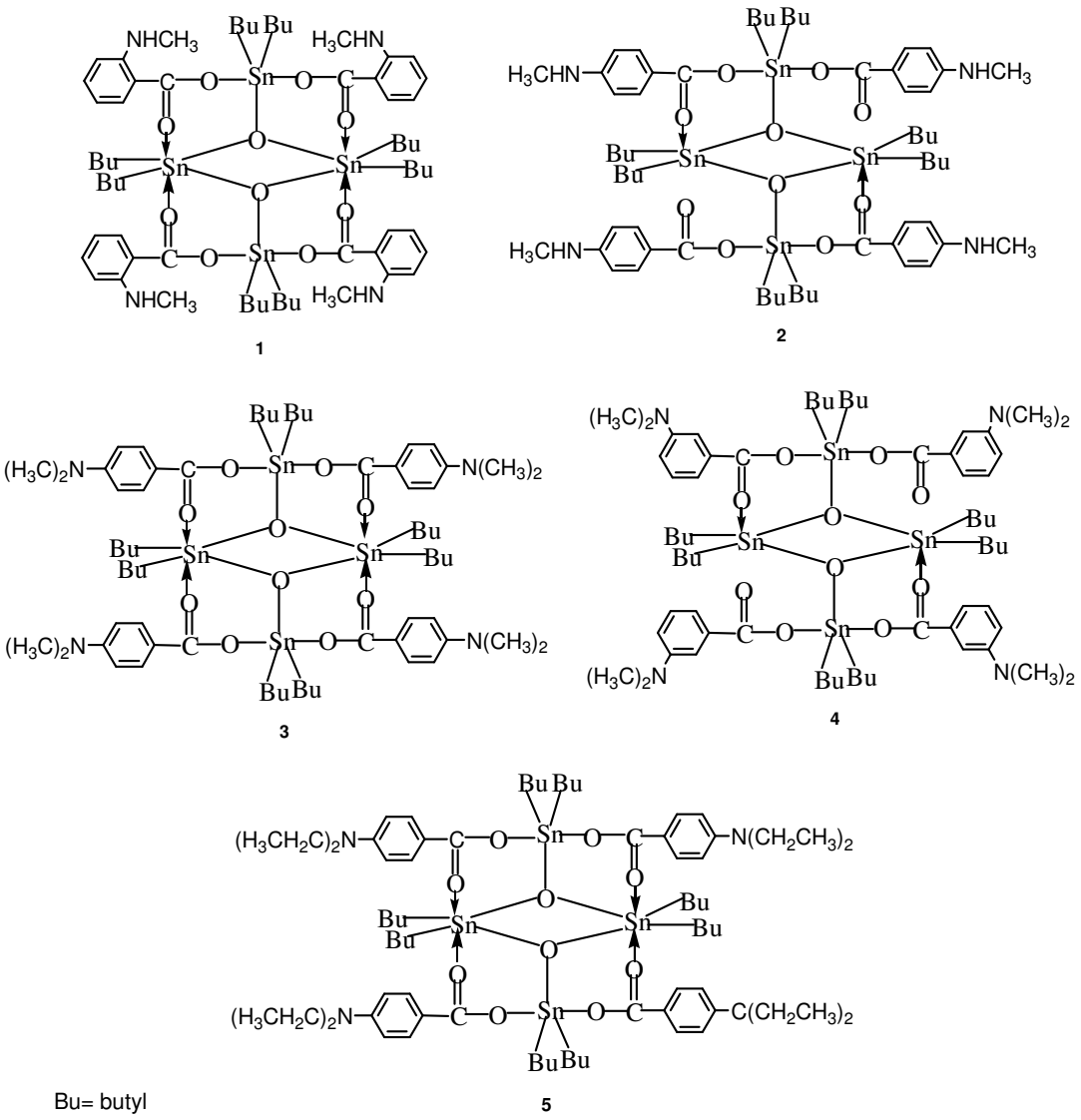


Figure 1. The proposed structure for complexes 1 to 5.

infrared spectra of complexes 1 to 5 revealed that the $\nu(COO)_{as}$ was shifted to a lower wave number compared to the parent acids signifying that the coordination took place via the oxygen atoms of the carboxylate anion. Generally, the $\Delta\nu = [\nu(COO)_{as} - \nu(COO)_s]$ value is used to determine the bonding properties of carboxylate anion to tin atom in organotin(IV) carboxylate complexes (Sandhu and Verma, 1987). Two $\Delta\nu$ values for organodistannoxane dimer type complexes indicate that the carboxylate anions were coordinated to the tin atom moiety in either a monodentate or bidentate manner (Win *et al.*, 2008). From the infrared spectra of complexes 1 to 3; two $\Delta\nu$ values (242 and 126 cm^{-1} for complex 1, 278 and 201 cm^{-1} for complex 2 and 277 and 192 cm^{-1} for complex 3) were observed. For complexes 1 and 3, the $\Delta\nu$ values were either comparable or lower than the $\Delta\nu$ of the sodium salt of the respective acids, indicating that the carboxylate anions were bonded to the tin atom in a bidentate mode (Win et al., 2008). As a result, the two tin atoms exhibited distorted trigonal bipyramid geometry while the other two tin atoms exhibited a distorted octahedral geometry in complexes 1 and 3. Complex 5 also revealed similarities to complexes 1 and 3. For complex 2, the first $\Delta\nu$ value (278 cm^{-1}) was larger than the $\Delta\nu$ value of the sodium salt while the second $\Delta\nu$ value (201 cm^{-1}) was comparable to the sodium salt (194 cm^{-1}). As a result, each tin atom exhibited a distorted trigonal bipyramid geometry in complex 2. Complex 4 was also isolated as bulky organodistannoxane dimer types and was found to be similar to complex 2. Further evidence for the coordination to Sn via O atoms was revealed by the presence of the ν(Sn-O-Sn) stretching bands in the

Table 1. Selected infrared data of organic acids and complexes 1 to 5.

Compounds	Wavelength (cm^{-1})						
	ν(OH)	$\nu(COO)_{as}$	$\nu(COO)_{s}$	Δν	ν(Sn-O)	ν(Sn-O-Sn)	ν(Sn-C)
2-$(NHCH_3)C_6H_4COOH$	2823 - 2491	1660	1334	326	-	-	-
2-$(NHCH_3)C_6H_4COONa$		1610	1389	221	-	-	-
1		1618 1519	1376 1393	242 126	437	630	526
4-$(NHCH_3)C_6H_4COOH$	2888 - 2551	1670	1318	352	-	-	-
4-$(NHCH_3)C_6H_4COONa$	-	1615	1421	194	-	-	-
2	-	1610 1592	1332 1391	278 201	481	618	553
4-$[N(CH_3)_2]C_6H_4COOH$	2807 - 2558	1668	1319	349	-	-	-
4-$[N(CH_3)_2]C_6H_4COONa$	-	1614	1383	231	-	-	-
3	-	1618 1605	1341 1413	277 192	483	600	551
3-$[N(CH_3)_2]C_6H_4COOH$	2885 - 2544	1677	1360	317	-	-	-
3-$[N(CH_3)_2]C_6H_4COONa$	-	1569	1387	182	-	-	-
4	-	1595 1573	1330 1368	265 205	420	635	572
4-$[N(C_2H_5)_2]C_6H_4COOH$	2899 - 2545	1663	1357	306	-	-	-
4-$[N(C_2H_5)_2]C_6H_4COONa$	-	1604	1356	248	-	-	-
5	-	1604 1576	1350 1394	254 182	468	634	548

$\Delta\nu = [\nu(COO)_{as} - \nu(COO)_{s}]$.

range of 635-600 cm^{-1} in the spectra of complexes 1 to 5 (Win et al., 2008).

The ^1H NMR spectra of complexes 1 to 5 revealed similarities to their parent acids such as the occurring of the -alkylamino groups and benzene rings. These signals did not show any significant shifting indicating that the benzene and –alkylamino groups is not involved in the coordination to tin atom moiety in the complexes (Table 2). Complexes 1 to 5 consisted of dibutyl groups (distannoxane dimer types) and found in the upfield region in the NMR spectra. Theoretically, the butyl groups should exhibit four signals corresponding to the protons, with multiplicities of triplet, sextet, quintet and triplet with integration values of 3:2:2:2, respectively. However, these complexes only exhibited three sets of signals in the range of 0.84 to 0.96 ppm (CH_3, triplet or multiplet), 1.31 to 1.50 ppm (CH_2, multiplet) and 1.58 to 1.79 ppm (CH_2, multiplet) respectively, due to the methylene protons having very similar environment causing their signals to overlap with each other in the ^1H NMR spectra (Danish et al., 1995; Win et al., 2008). Based on the integration values, the number of butyls protons in complexes 1 to 5 was in accordance with the number of protons proposed.

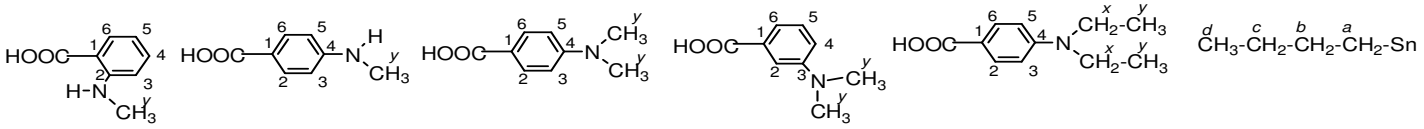
The formation of the complexes was evident from the δ(COO) values in the ^{13}C NMR spectra. All the complexes exhibited a δ(COO) signal in the range of 173.44 to 175.55 ppm. The chemical shift of the δ(COO) signal in each complex was shifted downfield compared to that of their respective parent acids indicating the participation of the carboxylate anions in the coordination to the tin(IV) atom. This phenomenon resulted from the decrease of the electron density in the carboxylate anions upon coordinated to the tin atom moiety during complex formation. Complexes 1 to 5 derivatives of the organodistannoxane dimer type exhibited two sets of butyl signals in the ^{13}C NMR spectra. These two sets of butyl signals were attributed to the butyl groups linked to the exo- and endo-cyclic tin atoms respectively (Danish et al., 1995; Win et al., 2008). Overall, the methyl and methylene carbon signals of butyls groups of complexes 1 to 5 were found in the range of 14.04 to 14.09 and 25.87 to 30.99 ppm. Generally, the ^1H and ^{13}C NMR spectra of the complexes obtained are found to exhibit no additional resonance and thus reflect the purity of the complexes.

For diorganotin(IV) carboxylate complexes, the $\delta(^{119}Sn)$ value for five-coordinated complexes between -90 to -190 ppm and for six-coordinated complexes between -210 to -400 ppm (Holeček et al., 1986). In the ^{119}Sn NMR spectra and the data in Table 3, complexes 1 and 5 exhibited two well resolved signals, one lying in the range of a five-coordinated geometry and the other in the range of a six-coordinated geometry, indicating that the carboxylate anions remained in a bidentate manner in the coordination to the tin atom. From Table 3, the exo- and endo-cyclic tin atoms in complexes 2 to 4 were five-coordinated respectively. This indicated that a pair of carboxylate anions was bonded to the tin atom in a monodentate manner while the other two carboxylate anions were bonded to the tin atoms (endo and exo-cyclic tin atom) in a bridging bidentate manner. As a

Table 2. ^{1}H NMR data of organic acids and complexes 1 to 5.

Compounds	Chemical Shift, δ(ppm)		
	Benzene	Amino (N-R, R= $NHCH_3$, $N(CH_3)_2$ and $N(CH_2CH_3)_2$	Sn- Bu
2-$(NHCH_3)C_6H_4COOH$ ($CDCl_3$)	6.66 (t, 7.4 Hz, 1H) H5; 6.71 (d, 8.2 Hz, 1H) H3 7.46 (t, 7.4 Hz, 1H) H4; 8.01 (d, 7.7 Hz, 1H) H6	2.96 (s, 3H) H*y*	-
1 ($CDCl_3$)	6.66 (t, 6.7 Hz, 4H) H5; 6.70 (d, 8.2 Hz, 4H) H3 7.43 (t, 7.4 Hz, 4H) H4; 7.92 (d, 7.2 Hz, 4H) H6	2.97 (s, 12H) H*y*	0.84-0.96 *(m, 24H) H*d* 1.32-1.50 *(m, 16H) H*c* 1.63-1.79 *(m, 32H) H*a* and H*b*
4-$(NHCH_3)C_6H_4COOH$ ($CDCl_3$)	6.59 (d, 8.8 Hz, 2H) H3 and H5; 7.97 (d, 8.9 Hz, 2H) H2 and H6	2.93 (s, 3H) H*y*	-
2 ($CDCl_3$)	6.61 (s, 8H) H3 and H5; 7.92 (s, 8H) H2 and H6	2.92 (s, 12H) H*y*	0.87-0.90 *(m, 24H) H*d* 1.31-1.44 *(m, 16H) H*c* 1.65-1.75 *(m, 32H) H*a* and H*b*
4-$[N(CH_3)_2]C_6H_4COOH$ ($CDCl_3$)	6.72 (d, 8.9 Hz, 2H) H3 and H5; 8.00 (d, 9.0 Hz, 2H) H2 and H6	3.09 (s, 6H) H*y*	-
3 ($CDCl_3$)	6.71 (s, 8H) H3 and H5; 7.96 (s, 8H) H2 and H6	3.07 (s, 24H) H*y*	0.87-0.91 *(m, 24H) H*d* 1.38-1.42 *(m, 16H) H*c* 1.58-1.75 *(m, 32H) H*a* and H*b*
3-$[N(CH_3)_2]C_6H_4COOH$ ($CDCl_3$)	7.00 (d, 9.3 Hz, 1H) H4; 7.35 (t, 8.1 Hz, 1H) H5 7.54 (d, 7.8 Hz, 2H) H2 and H6	3.03 (s, 6H) H*y*	-
4 ($CDCl_3$)	6.94 (d, 6.8 Hz, 4H) H4; 7.34 (t, 7.3 Hz, 4H) H5 7.43 (d, 7.5 Hz, 8H) H2 and H6	3.04 (s, 24H) H*y*	0.81-0.93 *(m, 24H) H*d* 1.32-1.45 *(m, 16H) H*c* 1.71-1.78 *(m, 32H) H*a* and H*b*
4-$[N(C_2H_5)_2]C_6H_4COOH$ ($CDCl_3$)	6.66 (d, 9.1 Hz, 2H) H3 and H5; 7.97 (d, 9.1 Hz, 2H) H2 and H6	1.22 (t, 7.1 Hz, 6H) H*y* 3.44 (q, 7.1 Hz, 4H) H*x*	-
5 ($CDCl_3$)	6.66 (d, 6.9 Hz, 8H) H3 and H5; 7.91 (d, 7.3 Hz, 8H) H2 and H6	1.23 (t, 6.5 Hz, 24H) H*y* 3.44 (q, 6.8 Hz, 16H) H*x*	0.88 (t, 7.3 Hz, 24H) H*d* 1.37-1.41 *(m, 16H) H*c* 1.57-1.74 *(m, 32H) H*a* and H*b*

s= singlet, d= doublet, t= triplet, q= quartet, m= multiplet; Coupling constant= Hz, *= overlap



result, all the tin atoms in complexes 2 to 4 were five-coordinated and exhibited a distorted trigonal bipyramid geometry. The structure elucidation (Figure 1) based on the infrared spectrum of complex 3, indicated that the carboxylate anions were bonded to the tin atom in a bidentate manner resulting in a pair of tin atoms being six-coordinated while the other pair of tin atoms were five-coordinated. However all the tin atoms in complex 3 were five-coordinated in ^{119}Sn solution due to the disassociation of a pair of bidentate bonding carboxylate

Table 3. ^{119}Sn and ^{13}C NMR data of organic acids and complexes 1-5.

Compounds	Chemical Shift (ppm)				
	^{119}Sn	Benzene	Amino [N-R, R= $NHCH_3$, $N(CH_3)_2$ and $N(CH_2CH_3)_2$]	Sn- Bu	COO
2-$(NHCH_3)C_6H_4COOH$ ($CDCl_3$)	-	109.04 (C1), 111.32 (C3), 114.97 (C5), 133.01 (C6), 136.10 (C4), 153.05 (C2)	29.97 (C*y*)	-	174.57
1 ($CDCl_3$)	-202.58 -214.13	110.91 (C1), 113.59 (C3), 114.46 (C5), 133.11 (C6), 134.35 (C4), 152.44 (C2)	30.03 (C*y*)	14.12 (C*d*), 25.87 (C*c*), 26.80 (C*c*), 27.26 (C*a*), 27.89 (C*a*), 28.23 (C*b*), 28.66 (C*b*)	175.55
4-$(NHCH_3)C_6H_4COOH$ ($CDCl_3$)	-	111.51 (C3 and C5), 117.61 (C1) 132.61 (C2 and C6), 153.89 (C4)	30.50 (C*y*)	-	172.44
2 ($CDCl_3$)	-199.79 -202.16	111.38 (C3 and C5), 122.15 (C1) 132.29 (C2 and C6), 152.68 (C4)	30.70 (C*y*)	14.06 (C*d*), 26.80 (C*c*), 27.14 (C*c*), 27.23 (C*a*), 27.34 (C*a*), 27.89 (C*b*), 28.19 (C*b*)	173.45
4-$[N(CH_3)_2]C_6H_4COOH$ ($CDCl_3$)	-	111.29 (C3 and C5), 116.61 (C1) 132.44 (C2 and C6), 154.08 (C4)	40.57 (C*y*)	-	172.64
3 ($CDCl_3$)	-199.33 -201.84	111.08 (C3 and C5), 120.99 (C1) 132.06 (C2 and C6), 153.25 (C4)	40.57 (C*y*)	14.08 (C*d*), 26.46 (C*a*), 27.15 (C*a*), 27.26 (C*b*), 27.38 (C*b*), 27.90 (C*c*), 28.21 (C*c*)	173.54
3-$[N(CH_3)_2]C_6H_4COOH$ ($CDCl_3$)	-	114.11 (C2), 118.06 (C4), 118.69 (C6), 129.53 (C5), 130.37 (C1), 150.87 (C3)	40.97 (C*y*)	-	173.29
4 ($CDCl_3$)	-195.35 -207.95	114.29 (C2), 116.42 (C4), 118.58 (C6), 129.11 (C5), 131.52 (C1), 150.90 (C3)	41.09 (C*y*)	14.04 (C*d*), 26.84 (C*c*), 27.26 (C*c*), 27.96 (C*b*), 28.24 (C*b*), 29.06 (C*a*), 30.99 (C*a*)	173.79
4-$[N(C_2H_5)_2]C_6H_4COOH$ ($CDCl_3$)	-	110.54 (C3 and C5), 115.45 (C1), 132.73 (C2 and C6), 151.96 (C4)	12.88 (C*y*) 44.93 (C*x*)	-	173.02
5 ($CDCl_3$)	-171.65 -221.43	110.43 (C3 and C5), 120.03 (C1), 132.36 (C2 and C6), 150.78 (C4)	12.95 (C*y*) 44.87 (C*x*)	14.09 (C*d*), 26.83 (C*c*), 27.13 (C*c*), 27.27 (C*b*), 27.42 (C*b*), 27.90 (C*a*), 28.45 (C*a*)	173.44

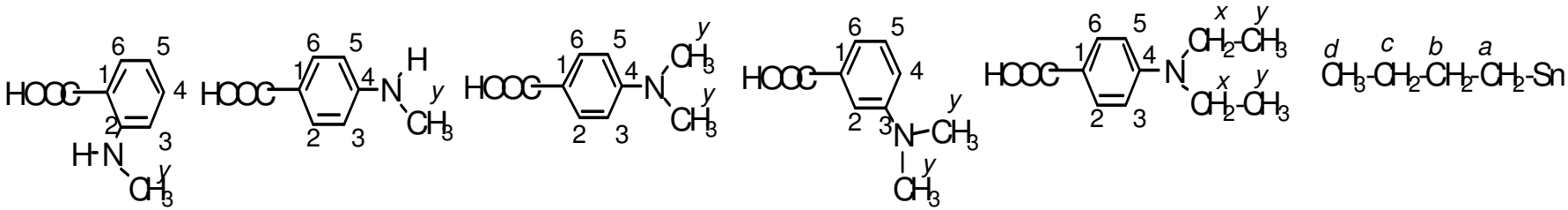


Table 4. Cytotoxicity assays, IC_{50} of organic acids and complexes 1 to 5.

Complexes	IC_{50} (µg/mL)
	Human liver hepatocellular carcinoma cells, HepG2
2-$(NHCH_3)C_6H_4COOH$	0.322 ± 0.018
4-$(NHCH_3)C_6H_4COOH$	0.821 ± 0.039
4-$[N(CH_3)_2]C_6H_4COOH$	Inactive (start at 1.0)
3-$[N(CH_3)_2]C_6H_4COOH$	Inactive (start at 1.0)
4-$[N(C_2H_5)_2]C_6H_4COOH$	Inactive (start at 1.0)
1	0.210 ± 0.014
2	0.344 ± 0.014
3	0.501 ± 0.021
4	0.541 ± 0.027
5	0.722 ± 0.036
Vincristine sulphate	0.042 ± 0.013

IC_{50}(µg/mL)= the concentration that yields 50% inhibition of the cell compared with untreated control. The cytotoxicity values are expressed as mean ± S.E.M. from the triplicate. Reference drug= Vincristine sulphate.

anions upon dilution.

In vitro cytotoxic assay

The IC_{50} values for the acids and complexes 1 to 5 are given in Table 4. From Table 4, all the parent acids were found to be inactive against HepG2 cell lines except 2-(methylamino)benzoic and 4-(methylamino)benzoic acid showed some activity towards the cell line with IC_{50} values of 0.322 and 0.821 µg/mL respectively. This maybe due to the presence of the substituted amino group at the *ortho* position of the benzene ring [2-(methylamino)benzoic acid] which causes an increase in the polarity of the compounds. In addition, the acids with the substituted amino groups at the *meta* and *para* position of the benzene ring with the increasing of the -alkylamino group resulting the compounds to be more bulky. This in turn will decrease the transportation ability towards the targeted cell as the major hypothesis in this screening study. Complexes 1-5 derivative of dibutyltin(IV) distannoxane dimer type showed that the increase in the IC_{50} values from 0.210 to 0.722 µg/mL indicated a decrease in the cytotoxicity activity for the respective complexes due to the increase in the bulkiness of the complexes. This observation is similar to the results found for their parent acids respectively. As a result, the additional of -alkylamino group increase the bulkiest of the complexes and decrease the transportation ability of the complexes towards the targeted cell.

Conclusion

Complexes 1 to 5 have been successfully synthesized and obtained as an organodistannoxane dimer type. The structural as well as the coordination number of tin moieties of complexes 1 to 5 have been successfully characterized quantitatively and qualitatively. Based on the *in vitro* cytotoxic screening activity, complex 1 showed significant activity compared to complexes 2 to 5 but its activity is lower than the reference drugs.

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