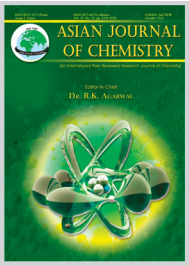
Asian Journal of Chemistry; Vol. 24, No. 10 (2012), 4763-4767

ASIAN JOURNAL OF CHEMISTRY

www.asianjournalofchemistry.co.in



Synthesis and Characterization of Dibutyltin(IV) and Triphenyltin(IV) Complexes Derivatives of 3-Methyl-4-nitrobenzoic and 4-Methyl-3-nitrobenzoic Acid and *in vitro* Cytotoxic Assay on Human Liver Hepatocellular Carcinoma Cells (HepG2)

YIP-FOO WIN[1,2,*], SIANG-GUAN TEOH[2,*], SIE-TIONG HA[1], SOON-HAN WAI[3] and EMAD YOUSIF[4]

[1]Department of Chemical Science, Faculty of Science, Universiti Tunku Abdul Rahman, Perak Campus, Jalan Universiti, Bandar Barat, 31900 Kampar, Perak, Malaysia
[2]School of Chemical Sciences, Universiti Sains Malaysia, 11800 Minden Penang, Pulau Pinang, Malaysia
[3]Department of Construction Management, Faculty of Engineering and Green Technology, Universiti Tunku Abdul Rahman, Perak Campus, Jalan Universiti, Bandar Barat, 31900 Kampar, Perak, Malaysia
[4]Department of Chemistry, College of Science, Al-Nahrain University, Baghdad, Iraq

*Corresponding author: Fax: +60 5 4661676; Tel: +60 5 4688888; E-mail: williamyfw@yahoo.com

(*Received*: 26 September 2011; *Accepted*: 14 May 2012) AJC-11487

In this study, two dibutyltin(IV) complexes which are obtained as organodistannoxane dimer with the general formulae of $\{[X\text{-}CH_3\text{-}Y\text{-}NO_2\text{-}C_6H_3COO(C_4H_9)_2Sn]_2O\}_2$ (X= *meta*, Y= *para* **1**; X= *para*, Y= *meta* **3**) and another two triphenyltin(IV) complexes (monomer) with the general formulae of $X\text{-}CH_3\text{-}Y\text{-}NO_2\text{-}C_6H_3COO(C_6H_5)_3Sn.CH_3OH$ (X= *meta*, Y= *para* **2**; X= *para*, Y= *meta* **4**) have been successfully synthesized. The infrared and NMR spectroscopy on the parent acids and their dibutyltin(IV) and triphenyltin(IV) complexes showed that the coordination took place *via* oxygen atoms from the carboxylate anions. Based on the spectroscopy studies indicated that one methanol molecule also take part in the coordination to tin(IV) atoms moiety in complexes **2** and **4** resulting the tin(IV) atoms moiety are five-coordinated in solid state and four-coordinated in solution state. From the preliminary *in vitro* cytotoxic assay, complexes **2** and **4** [triphenyltin(IV)] showed better activity compared to complexes **1** and **3** [diorganotin(IV)].

Key Words: Cytotoxic assay, Synthesis, Characterization, Dibutyltin(IV), Triphenyltin(IV), Complexes.

INTRODUCTION

The study of organotin(IV) complexes in term of chemistry has been well established and the study of its biological properties against bacterial, fungal and cancer cells line also have been expanded tremendously[1-5]. Based on literature review over the past 20 years, the structures of many different types of tri- and diorganotin(IV) carboxylate complexes have been reported and tested for their *in vitro* activities against a large array of tumor cell lines[1,6,7]. In fact and up to date, organotin(IV) carboxylate complexes are extensively studied due to its coordination geometries as well as structural diversity (monomer, dimeric, hexameric and oligomeric), which are attributed from the coordinating ligands[8-11].

As part of our interest and research on organotin(IV) work, we have synthesized and characterized organotin(IV) carboxylate complexes derivatives of 3-methyl-4-nitrobenzoic acid, $3\text{-}CH_3\text{-}4\text{-}NO_2\text{-}C_6H_3COOH$ and 4-methyl-3-nitrobenzoic acid, $4\text{-}CH_3\text{-}3\text{-}NO_2\text{-}C_6H_3COOH$. In addition, the preliminary cytotoxic assay of the complexes were screened against human liver hepatocellular carcinoma cells, HepG2 and the results are reported herein.

EXPERIMENTAL

All the reagents and solvents were purchased commercially and used without any further purification. Infrared spectra were recorded using a Perkin-Elmer FTIR GX Spectrophotometer as KBr disc in the frequency range of 4000-400 cm^{-1}. The spectra for ^1H 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 300 MHz FTNMR spectrometer using deuterated $CDCl_3$ and d_6-DMSO as the solvent and tetramethylsilane, TMS as the internal standard. Elemental C, H and N analyses were carried out on a Fison EA 1108 CHNS-O analyzer. Tin was determined gravimetrically by igniting a known quantity of each complex to SnO_2. The melting points were determined in an open capillary and were uncorrected.

Preliminary *in vitro* cytotoxic assay: The *in vitro* cytotoxic assay was carried out against human liver hepatocellular carcinoma cells line, HepG2. The cells were maintained in

TABLE-1
SELECTED INFRARED DATA OF PARENT ACIDS, SODIUM SALTS AND COMPLEXES **1-4**

Compounds	Wavelength (cm^{-1})					
	ν(OH)	$\nu(COO)_{as}$	$\nu(COO)_{s}$	Δν	ν(Sn-O)	ν(Sn-O-Sn)/ν(O-Sn-O)
3-CH_3-4-NO_2-C_6H_3COOH	2827 - 2552	1682	1311	371	-	-
3-CH_3-4-NO_2-C_6H_3COONa	-	1635	1356	279	-	-
1	-	1635, 1523	1306, 1339	329, 183	469	635
2	-	1619	1340	279	449	-
4-CH_3-3-NO_2-C_6H_3COOH	2832 - 2546	1699	1319	380	-	-
4-CH_3-3-NO_2-C_6H_3COONa	-	1650	1354	296	-	-
3	-	1620, 1530	1341, 1407	179, 123	480	636
4	-	1641	1336	305	448	-

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[12,13]. 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)[13]. Cytotoxicity was expressed as fifty percent cytotoxic dose (IC_{50}), *i.e.* the concentration causing 50 % inhibition of cell growth with reference to the control (untreated cells). The IC_{50} and the SEM (standard error of the mean) was determined using Probit Analysis (SPSS, version 12.0.1).

Preparation of sodium salts and complexes: The sodium salts of the parent acids were obtained by heating under reflux a 1:1 molar mixture of sodium hydroxide and respective acid in ethanol (50 mL) for 2 h. After a few days, white precipitates were obtained.

Preparation of {[3-CH_3-4-NO_2-$C_6H_3COO(C_4H_9)_2$ $Sn]_2O\}_2$ (1): Complex **1** was prepared by heating under reflux a 1:1 molar mixture of dibutyltin(IV) oxide (0.49 g, 2 mmol) and 3-methyl-4-nitrobenzoic acid (0.36 g, 2 mmol) in acetone (50 mL) as solvent and the mixture was heated under reflux for 2 h. After few days, yellow crystals (0.77 g, 61 % yield) were collected. m.p.: 216.7-217.1 ºC. Analysis for $C_{64}H_{96}N_4O_{18}Sn_4$: C, 45.55; H, 5.93; N, 3.20; Sn, 16.95 %. Calculated for $C_{64}H_{96}N_4O_{18}Sn_4$: C, 45.64; H, 5.75; N, 3.33; Sn, 17.09 %.

Preparation of 3-CH_3-4-NO_2-$C_6H_3COO(C_6H_5)_3$ Sn.CH_3OH (2): Complex **2** was obtained by heating under reflux a 1:1 molar mixture of triphenyltin(IV) hydroxide (0.73 g, 2 mmol) and 3-methyl-4-nitrobenzoic acid (0.36 g, 2 mmol) in a mixture of methanol/ethanol (1:1, 60 mL) for 2 h. After few days, yellow solids (0.77 g, 68.8 % yield) were collected. m.p.: 95.3-96.1 ºC. Analysis for $C_{27}H_{25}N_1O_5Sn$: C, 57.42; H, 3.92; N, 2.45; Sn, 20.33 %. Calcd. for $C_{27}H_{25}N_1O_5Sn$: C, 57.68; H, 4.48; N, 2.49; Sn, 21.11 %.

Preparation of {[4-CH_3-3-NO_2-$C_6H_3COO(C_4H_9)_2$ $Sn]_2O\}_2$ (3): This complex was prepared by a similar method to those described for complex **1**, except substituting 3-methyl-4-nitrobenzoic acid with 4-methyl-3-nitrobenzoic acid. Acetone (50 mL) was used as solvent and the mixture was heated under reflux for 3 h. A clear yellow transparent solution was isolated by filtration and kept in a bottle. After few days, yellow crystals (0.90 g, 71.3 % yield) were collected. m.p.: 209.3-209.9 ºC. Analysis for $C_{64}H_{96}N_4O_{18}Sn_4$: C, 45.46; H, 5.52; N, 3.25; Sn, 16.99 %. Calcd. for $C_{64}H_{96}N_4O_{18}Sn_4$: C, 45.64; H, 5.75; N, 3.33; Sn, 17.69 %.

Preparation of 4-CH_3-3-NO_2-$C_6H_3COO(C_6H_5)_3$ Sn.CH_3OH (4): Complex **4** was prepared by a similar method to those described for complex **2**, except replacing 3-methyl-4-nitrobenzoic acid with 4-methyl-3-nitrobenzoic acid and methanol (50 mL) was utilized in the synthesis. After few days, yellow solids (0.68 g, 60.7 % yield) were collected. m.p.: 133.7-134.3 ºC. Analysis for $C_{27}H_{25}N_1O_5Sn$: C, 57.80; H, 4.01; N, 2.43; Sn, 20.82 %. Calcd. for $C_{27}H_{25}N_1O_5Sn$: C, 57.68; H, 4.48; N, 2.49; Sn, 21.11 %.

RESULTS AND DISCUSSION

Complexes **1-4** have been obtained in solid state and gave a sharp melting point indicating the isolation of fairly pure complexes. In addition, the micro-elemental analysis for C, H, N and Sn data obtained were in agreement with the predicted formula for complexes **1-4** further supported the proposed structure of the respective complexes. An outline of the proposed reaction and structure of complexes **1-4** are depicted in Fig. 1 for further clarification.

The characteristic infrared absorption frequencies (cm^{-1}) and assignments of important absorption bands of the parent acids, sodium salts and complexes **1-4** are listed in Table-1. The ν(O-H) bands of the parent acids were absent in the infrared spectra of sodium salts and complexes **1-4** showed the deprotonation and coordination of the carboxylate anion. Complexes **1-4** revealed that the $\nu(COO)_{as}$ was shifted to a lower wavelength number compared to the parent acids which signify that the coordination took place *via* the oxygen atoms of the carboxylate anion. From the infrared spectra of complexes **1** and **3**, two Δν values were observed and the Δν values were either comparable or lower than the Δν of the sodium salts of the respective parent acids, indicating that the carboxylate anions were bonded to the tin(IV) atom in a bidentate mode[14]. As a result, two tin(IV) atoms were five-coordinated while another two tin(IV) atoms were six-coordinated in complexes **1** and **3**.

For complexes **2** and **4,** which were derivatives of triphenyltin(IV) carboxylate, Δν below 200 cm^{-1} would be expected for bridging or chelating carboxylates, but greater than 200 cm^{-1} for the monodentate bonding carboxylate anions[15]. Based on the data in Table-1, the carboxylate anions in complexes **2** and **4** would be expected to bond to the tin(IV)

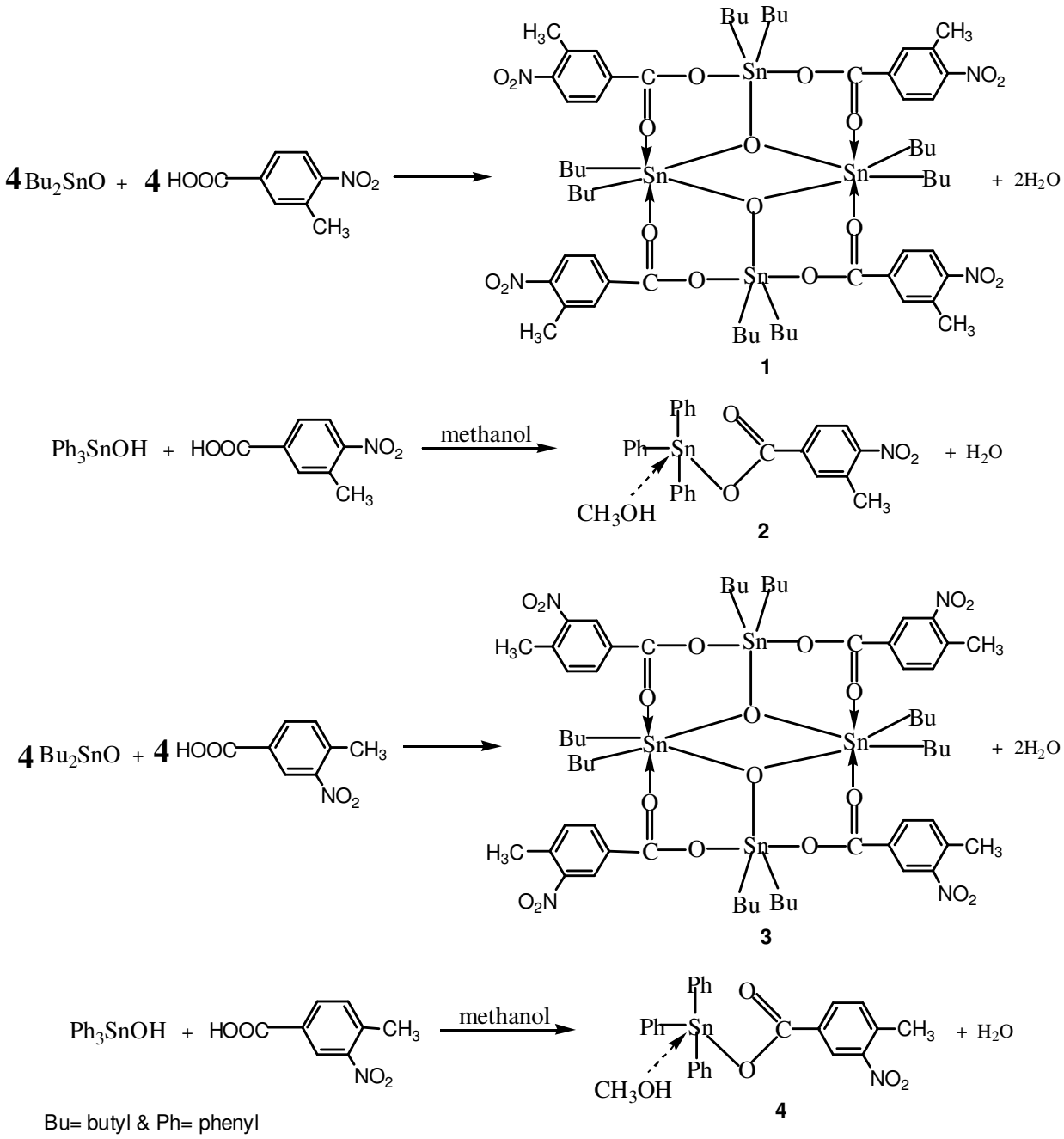


Fig. 1. Proposed reactions and structure of complexes **1-4**

atom in monodentate manner since the Δν above 200 cm^{-1}. Moreover, based on the micro-elemental analysis and proposed structure of complexes **2** and **4**, it was believed that one methanol molecule has taken part in the coordination to the tin(IV) atom. As a result, the overall coordination number of complexes **2** and **4** were five-coordinated and similar to the structure reported[16].

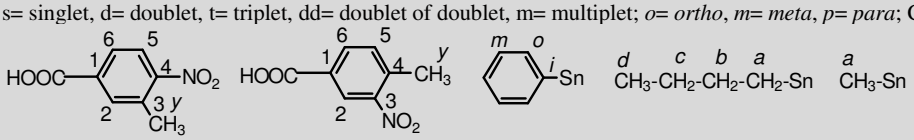
The ^{1}H NMR spectra of complexes **1-4** exhibited similarities to their parent acids and the spectral data of complexes **1-4** are summarized in Table-2. In the upfield regions of the ^{1}H NMR spectra of the complexes **1** and **3** showed that the signal of the butyl protons in the range of 0.77-1.87 ppm respectively. For complexes **2** and **4**, the resonances appeared as two well separated sets of multiplets in the regions centering around δ ≈ 7.51 and 7.85 ppm (downfield) with the integration values of 9:6 respectively ascribed to the aromatic protons of the phenyl group[17]. Based on the ^{1}H NMR spectral studies of complexes **2** and **4**, the proton resonances originating from the methanol molecule occured around δ ≈ 3.48 ppm and based on the integration, only one methanol molecule was present in complexes **2** and **4**. The methanol molecule (polar solvent) was believed to coordinate to the tin(IV) atom during the formation of the complexes.

Evidence of the formation of the complexes is displayed in the ^{13}C NMR spectra and the data of parent acids and complexes **1-4** are summarized in Table-3. The ^{13}C NMR spectra of complexes **1-4** showed the δ(COO) signal shifted to the downfield region, which is lower compared to that of the parent acids indicating the carboxylate anion is bonded to tin(IV) atom. Complexes **1** and **3** exhibited two sets of signals

TABLE-2
^{1}H NMR DATA OF PARENT ACIDS AND COMPLEXES **1-4**

Compounds	Chemical Shift, δ(ppm)			
	Benzene	Methyl	Sn-R (R= Bu and Ph)	CH_3OH
3-CH_3-4-NO_2-C_6H_3COOH (d_6-DMSO)	7.94 (dd, 1.4 Hz, 8.4 Hz, 1H) H6 8.02 (d, 4.5 Hz, 1H) H5 8.05 (s, 1H) H2	2.54 (s, 3H) H*y*	-	-
1 ($CDCl_3$)	7.99 (s, 12H) H2, H5 and H6	2.70 (s, 12H) H*y*	0.81 (t, 6.7 Hz, 12H) H*d* 0.89 (t, 7.1 Hz, 12H) H*d* 1.31-1.45 *(m, 16H) H*c* 1.65-1.75 *(m, 32H) H*a* and H*b*	-
2 ($CDCl_3$)	7.99 (d, 8.4 Hz, 1H) H5 8.12 (d, 7.9 Hz, 1H) H6 8.16 (s, 1H) H2	2.65 (s, 3H) H*y*	7.54-7.58 *(m, 9H) H*m* and H*p* 7.86-7.91 *(m, 6H) H*o*	3.49 (s, 3H)
4-CH_3-3-NO_2-C_6H_3COOH (d_6-DMSO)	7.58 (d, 7.8 Hz, 1H) H5 8.07 (d, 7.9 Hz, 1H) H6 8.36 (s, 1H) H2	2.55 (s, 3H) H*y*	-	-
3 ($CDCl_3$)	7.52 (d, 6.9 Hz, 4H) H5 8.21 (d, 7.6 Hz, 4H) H6 8.61 (s, 4H) H2	2.72 (s, 12H) H*y*	0.77 (t, 6.9 Hz, 12H) H*d* 0.89 (t, 7.2 Hz, 12H) H*d* 1.28-1.45 *(m, 16H) H*c* 1.63-1.78 *(m, 32H) H*a* and H*b*	-
4 ($CDCl_3$)	7.38 (d, 7.9 Hz, 1H) H5 8.20 (d, 7.9 Hz, 1H) H6 8.68 (s, 1H) H2	2.62 (s, 3H) H*y*	7.48-7.51 *(m, 9H) H*m* and H*p* 7.71-7.88 *(m, 6H) H*o*	3.47 (s, 3H)

s= singlet, d= doublet, t= triplet, dd= doublet of doublet, m= multiplet; *o*= *ortho*, *m*= *meta*, *p*= *para*; Coupling constant= Hz, *= overlap



corresponding to the butyl groups linked to the exo- and endocyclic tin(IV) atom respectively in the upfield region in the range of 13.87-30.70 ppm[18]. Complexes **2** and **4** revealed the chemical shifts of the $\delta(^{13}C)_{ipso}$ at 138.31 and 138.22 ppm respectively indicative of a four-coordinated tin(IV) atom and exhibited distorted tetrahedral geometry[19-21].

The $\delta(^{119}Sn)$ value of the five-coordinated diorganotin(IV) complexes fall in the range between -90 to -190 ppm and the six-coordinated complexes between -210 to -400 ppm[22]. Based on the ^{119}Sn NMR spectra, all the tin(IV) atoms in complexes **1** and **3** were five-coordinated and each exhibited a distorted trigonal bipyramidal geometry. However, based on the infrared spectral studies, the tin(IV) atoms of complexes **1** and **3** are either a five- or six-coordinated but the ^{119}Sn NMR spectral studies concluded that all the tin(IV) atoms in complexes **1** and **3** were five-coordinated. This is due to the disassociation of the bidentate bonds upon dilution during the preparation of the NMR sample in solution form.

Complexes **2** and **4** showed that the $\delta(^{119}Sn)$ values at -103.30 and -100.11 ppm which lie in the range of -40 to -120 ppm [for triphenyltin(IV) complexes], hence, indicating that the tin(IV) atoms in complexes **2** and **4** are four-coordinated and possessed a distorted tetrahedral geometry[19,20]. It is believed that methanol molecule was disassociated upon dilution during the NMR solution preparation resultant the tin(IV) atom in complexes **2** and **4** remained four-coordinated. This phenomenon may due to the coordinating bond of the carboxylate anions are stronger than methanol (dative bond).

Preliminary *in vitro* cytotoxic assay: The preliminary *in vitro* cytotoxic assay of parent acids and complexes **1-4** are given in Table-4. Based on the data given in Table-4, it was found that both the parent acids are inactive against HepG2 cell lines. In addition, complexes **2** (0.111 mg/mL) and **4** (0.061 mg/mL) are more active compared to complexes **1** (0.162 mg/mL) and **3** (0.522 mg/mL). This is due to complexes **1** and **3** were obtained as organodistannoxane dimer type (bulky molecules) hence the aid of ligands on organotin(IV) to the receptor (active) sites of the cells was inhibited. In general, complexes **2** and **4** were derivatives of triorganotin(IV), which is more active compared to the diorganotin(IV)[23-25]. In addition, in solution form, complexes **2** and **4** were four-coordinated and exhibited distorted tetrahedral geometry (sp^3) causing them to be more active[18]. However, their activities were lower compared to the vincristine sulphate (reference drug).

Conclusion: Complexes **1- 4** have been successfully synthesized. The structural as well as the coordination number of tin(IV) moieties of complexes **1-4** have been successfully characterized quantitatively and qualitatively. Based on the preliminary *in vitro* cytotoxic assay on human liver hepatocellular carcinoma cells (HepG2), complexes **2** and **4** [triphenyltin(IV)] showed better activity compared to complexes **1** and **3** [diorganotin(IV)] but lower activity compared to the reference drug.

ACKNOWLEDGEMENTS

The authors thank Universiti Tunku Abdul Rahman (UTAR) for the UTAR Research Fund (Project No. IPSR/ RMC/ UTARRF/C1-11/C07) and Universiti Sains Malaysia (USM) for financial support as well as technical assistance and facilities.

REFERENCES

1. M. Gielen, M. Biesemans, D.d. Vos and R. Willem, *J. Inorg. Biochem.*, **79**, 139 (2000).
2. S. Mahmood, S. Ali, M.H. Bhatti, M. Mazhar, K. Shahid, K.M. Khan and G.M. Maharvi, *Turk. J. Chem.*, **28**, 17 (2004).
3. M. Hanif, M. Hussain, S. Ali, M.H. Bhatti, M.S. Ahmed, B. Mirza and H. Stockli-Evans, *Polyhedron*, **29**, 613 (2010).

TABLE-3
^{119}Sn AND ^{13}C NMR DATA OF PARENT ACIDS AND COMPLEXES **1-4**

Compounds	Chemical Shift (ppm)					
	^{119}Sn	Benzene	Sn-R (R= Me, Bu and Ph) $^{n}J(^{119}Sn-^{13}C)$ (n=1, 2, 3 and 4)	Methyl	CH_3OH	COO
3-CH_3-4-NO_2-C_6H_3COOH (d_6-DMSO)	-	125.46 (C5), 128.94 (C6), 133.70 (C3), 134.36 (C2), 135.42 (C1), 152.32 (C4)	-	19.95 (C*y*)	-	166.72
1 ($CDCl_3$)	-206.97 -208.09	124.87 (C5), 128.57 (C6), 133.61 (C3), 134.49 (C2), 137.36 (C1), 151.94 (C4)	13.87 (C*d*), 13.96 (C*d*), 27.08 (C*c*), 27.13 (C*c*), 27.88 (C*b*), 28.21 (C*b*), 29.09 (C*a*), 30.70 (C*a*)	20.47 (C*y*)	-	171.38
2 ($CDCl_3$)	-103.30	124.75 (C5), 129.39 (C6), 133.56 (C3), 135.28 (C2), 138.05 (C1), 152.12 (C4)	138.31 (^{1}J= 633.6 Hz) (C*i*), 137.31 (^{2}J= 48.2 Hz) (C*o*), 129.09 (^{3}J= 63.7 Hz) (C*m*), 130.84 (^{4}J= 12.3 Hz) (C*p*)	20.31 (C*y*)	51.06	170.98
4-CH_3-3-NO_2-C_6H_3COOH (d_6-DMSO)	-	125.79 (C2), 130.87 (C1), 134.18 (C5 & C6), 138.49 (4), 149.57 (C3)	-	20.54 (C*y*)	-	166.32
3 ($CDCl_3$)	-207.39 -211.19	126.37 (C2), 132.99 (C1), 133.37 (C5), 134.38 (C6), 138.03 (C4), 149.54 (C3)	13.87 (C*d*), 14.01 (C*d*), 26.34 (C*c*), 27.14 (C*c*), 28.18 (C*b*), 28.43 (C*b*), 29.18 (C*a*), 30.56 (C*a*)	20.95 (C*y*)	-	170.97
4 ($CDCl_3$)	-100.11	127.12 (C2), 130.53 (C1), 133.13 (C5), 134.80 (C6), 137.97 (C4), 149.58 (C3)	138.22 (C*i*), 137.32 (^{2}J= 49.1 Hz) (C*o*) 129.48 (^{3}J= 65.1 Hz) (C*m*), 130.82 (^{4}J= 13.3 Hz) (C*p*)	20.91 (C*y*)	-	170.70

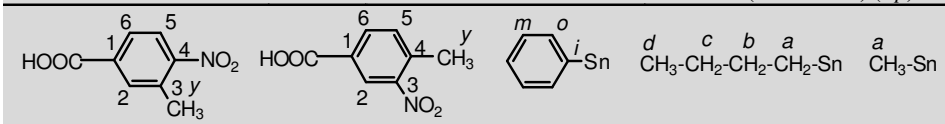


TABLE-4
CYTOTOXIC ASSAYS, IC_{50} VALUE OF PARENT ACIDS AND COMPLEXES **1-4**

Complexes	IC_{50} (μg/mL)
	Human liver hepatocellular carcinoma cells, HepG2
3-CH_3-4-NO_2-C_6H_3COOH	Inactive (start at 1.0)
1	0.161 ± 0.013
2	0.111 ± 0.011
4-CH_3-3-NO_2-C_6H_3COOH	Inactive (start at 1.0)
3	0.522 ± 0.018
4	0.061 ± 0.008
Vincristine sulphate (reference)	0.042 ± 0.031

IC_{50} (μg/mL) = 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

4. Y.-F. Win, S.-G. Teoh, T.-S. Tengku-Muhammad, Y. Sivasothy and S.-T. Ha, *Am. J. Applied Sci.*, **7**, 301 (2010).
5. Y.-F. Win, S.-G. Teoh, T.-S. Tengku-Muhammad, Y. Sivasothy and S.-T. Ha, *Aust. J. Basic and Appl. Sci.*, **4(6)**, 1383 (2010).
6. R. Willem, A. Bouhdid, B. Mahieu, L. Ghys, M. Biesmans, E.R.T. Tiekink, D.d. Vos and M. Gielen, *J. Organomet. Chem.*, **531**, 151 (1997).
7. D. Zuo, T. Jiang, H. Guan, K. Wang, X. Qi and Z. Shi, *Molecules*, **6**, 647 (2001).
8. R. Zhang, J. Sun and C. Ma, *J. Organomet. Chem.*, **690**, 4366 (2005).
9. Y.F. Win, S.G. Teoh, J.B.J. Teh, H.K. Fun and L. Zakaria, *Acta Cryst.*, **E63**, m323 (2007).
10. Y.F. Win, S.G. Teoh, P. Ibrahim, S.L. Ng and H.K. Fun, *Acta Cryst.*, **E63**, m667 (2007).
11. Y.F. Win, S.G. Teoh, S.T. Ha, R. Kia and H.K. Fun, *Acta Cryst.*, **E64**, m1530 (2008).
12. T. Mosmann, *J. Immunol. Methods.*, **65**, 55 (1983).
13. A.M. Ali, N.H. Ismail, M.M. Mackeen, L.S. Yazan, S.M. Mohamed, A.S.H. Ho and N.H. Lajis, *Pharmaceut. Biol.*, **38**, 298 (2000).
14. G.K. Sandhu and S.P. Verma, *Polyhedron*, **6**, 587 (1987).
15. L.L. Yeap and S.G. Teoh, *J. Coord. Chem.*, **56**, 701 (2003).
16. F.W. Yip, S.G. Teoh, B.M. Yamin and S.W. Ng, *Acta Cryst.*, **E66**, m1164 (2010).
17. A. Sau and R.R. Holmes, *J. Organomet. Chem.*, **217**, 157 (1981).
18. M. Danish, H.G. Alt, A. Badshah, S. Ali, M. Mazhar and N. Islam, *J. Organomet. Chem.*, **486**, 51 (1995).
19. J. Holecek, K. Handlír, M. Nádvornik and A. Lycka, *J. Organomet. Chem.*, **258**, 147 (1983).
20. J. Holecek, M. Nadvorník, K. Handlír and A. Lycka, *J. Organomet. Chem.*, **241**, 177 (1983).
21. T.S.B. Baul, S. Dhar, S.M. Pyke, E.R.T. Tiekink, E. Rivarola, R. Butcher and F.E. Smith, *J. Organomet. Chem.*, **633**, 7 (2001).
22. J. Holecek, M. Nádvornik, K. Handlír and A. Lycka, *J. Organomet. Chem.*, **315**, 299 (1986).
23. W. Rehman, M.K. Baloch and A. Badshah, *J. Braz. Chem. Soc.*, **4**, 827 (2005).
24. K. Shahid, S. Shahzadi, S. Ali and M. Mazhar, *Bull. Korean Chem. Soc.*, **27**, 44 (2006).
25. M.S. Ahmad, M. Hussain, M. Hanif, S. Ali and B. Mirza, *Molecules*, **12**, 2348 (2007).