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Model studies of trialkyltin–protein interactions: ^{13}C NMR analysis of solution equilibria of the complex between trimethyltin and methyl *N*-benzoyl-l-leucyl-l-histidinate

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^{13}C NMR spectra for the 1:1 complex between methyl *N*-benzoyl-l-leucyl-l-histidinate and the trimethyltin moiety in d-chloroform ($CDCl_3$), d_4-methanol (CD_3OD) dimethyl sulphoxide (DMSO) and d_6-DMSO/H_2O solvents are reported, and contrasted with those for the free ligand. The spectra are interpreted in terms of a variety of solution equilibria illustrating the nature of the interaction between the trimethyltin species and primarily the imidazole ring of the histidine residue. Evidence for the preferential stability of pentacoordinate solution structures about tin is presented.

Keywords: Organotin, histidine, ^{13}C NMR, equilibria, imidazole

INTRODUCTION

Organotin compounds are used extensively as biocides where selective toxicity towards different levels of life-form is used advantageously. Their species-specific potency varies primarily as a function of the number and types of organic ligand.[1] Additionally, human activity globally also gives rise to inorganic tin in the environment,[2,3] where biomethylation to toxic organotin has been demonstrated.[4] Recent attention has been focused on the environmental and ecological damage of organotin[5] and evidence of its rise up food chains has been the detection of organotins in humans.[6]

At the biomolecular level (mammalian), both trimethyl- and triethyltin are characteristically specific in their binding to relatively few tissue proteins.[7–10] Binding to mitochondrial ATPase[11] has been shown to inhibit function by blocking proton translocation through the membrane sector of this enzyme.[12] The role of histidine in trialkyltin (R_3Sn) binding has been implicated,[13] as has the requirement for some precise stereochemical geometry of ligands constituting the binding site within the tertiary structure of the enzyme.[8] Mössbauer studies of the trialkyltin–enzyme complex and competitive inhibition studies by intramolecularly pentacoordinate tin species[13] have suggested four-coordinate geometry about the bound tin, although a five-coordinate *cis* trigonal bipyramidal structure cannot be excluded.[14] The importance of hydrophobic interaction between the alkyl groups of R_3Sn and the region about the protein binding site has been suggested.[15]

In order to investigate this area further we report ^{13}C NMR studies on the solution equilibria for the interaction of trimethyltin with a histidine-only model binding site, namely the protected dipeptide methyl *N*-benzoyl-l-leucyl-l-histidinate. ^{13}C NMR studies on the solution equilibria of the free ligand model (isolated as a monohydrate) have been reported by the authors previously.[14]

EXPERIMENTAL PROCEDURE

The 1:1 complex **I** is formed simply by co-dissolving methyl *N*-benzoyl-l-leucyl-l-histidinate monohydrate[16] (209.2 mg, 0.518 mmol) and trimethyltin hydroxide (93.7 mg, equimolar) in anhydrous methanol (1 cm^3). After 24 h, the solution was reduced under vacuum at 0°C. The resulting solid was dissolved in the minimum of dichloromethane (CH_2Cl_2), filtered, and reduced under vacuum at 0°C to afford the off-white solid, complex **I** (233 mg, 82%). Analysis: Calc. for $C_{23}H_{34}N_4O_4Sn$: C, 50.29; H, 6.24; N, 10.20. Found:

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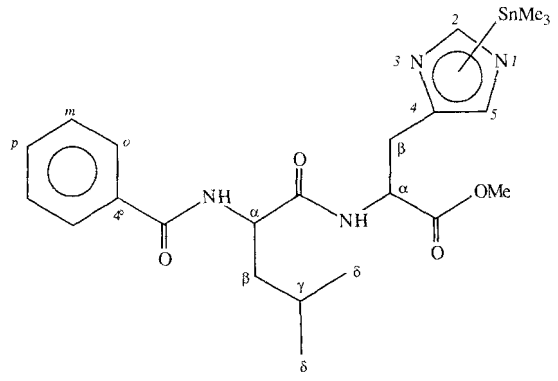

Figure 1 Atom identification nomenclature for complex **I** used in this paper.

C, 50.21; H, 6.41; N, 10.90%. The molecular framework of complex **I**, together with atom identification nomenclature is illustrated in Fig. 1.

^{13}C and H NMR spectra were recorded using a Bruker WM250 instrument and a Perkin–Elmer R32 respectively. Solutions were at 303°C and at a concentration of the order of 2×10^{-1} mol dm^{-3}. The number of hydrogen atoms directly bonded to the carbon atom associated with each particular resonance were determined by the DEPT technique. Data are relative to internal TMS. Microanalysis was obtained using a Perkin–Elmer 240B Elemental Analyser. Trimethyltin hydroxide was obtained from Ventron and used without further purification.

RESULTS AND DISCUSSION

The ^{13}C NMR spectrum for complex **I** in $CDCl_3$ solution is reproduced in Fig. 2 and the full listing of chemical shift data presented in Table 1. For comparison, the data for the free ligand, isolated as a monohydrate whose integrity persists in $CDCl_3$ solution, are also listed. As previously reported,[16] the ^{13}C NMR spectrum for the free ligand in $CDCl_3$ is best interpreted in terms of two sets of equilibria. The first is constituted by *cis/trans* conformational isomerization about the amide bond, whilst the second is an equilibrium between two structural isomers. These latter are composed, on the one hand, of a freely rotating imidazole ring species (displaying dynamic tautomeric isomerism in its protonation at positions N-1 and N-3) and the other a more rigidly fixed molecular structure embodying the imidazole ring species protonated at N-1 and hydrogen-bonded at N-3 to the molecule of water in the monohydrate.

For the complex **I** in $CDCl_3$ solution, the $Sn\underline{C}H_3$ resonance is observed as a single resonance at -1.20 ppm with unresolved coupling $^1J(^{119/117}Sn-^{13}C) = 475 \pm 10$ Hz. This coupling constant indicates five-coordination about tin.[17] The unresolved nature of the coupling strongly indicates some dynamic exchange in bonding about tin, although between two almost identical structural species. Examination of the proton NMR spectrum yields the single $Sn\underline{C}H_3$ resonance at 0.50 ppm with unresolved coupling $^2J(^{119/117}Sn-C-^1H) = 63 \pm 1$ Hz, corroborating the five-coordinate structure about tin,[18–20] and further supporting dynamic exchange between two almost equivalent magnetic environments about tin. [Note: ^1H NMR indicates that no racemization occurs upon complexation. This is true for all solution spectra discussed in this paper. ^1H NMR has been found clearly to indicate racemization (in terms of α-CH coupling and duplication of particular resonances), an adventitious discovery during our various synthetic approaches to free-ligand synthesis.]

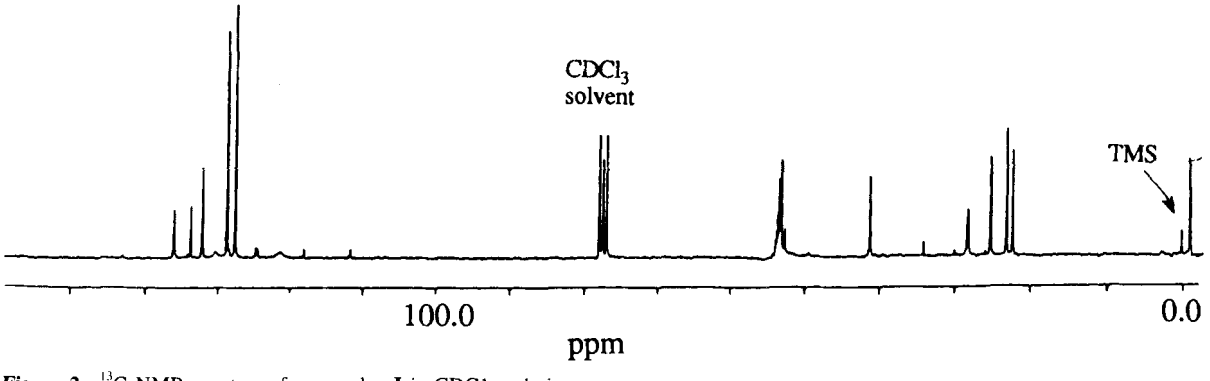

Figure 2 ^{13}C NMR spectrum for complex **I** in $CDCl_3$ solution.

Table 1 ^{13}C NMR chemical shift data for complex **I** in $CDCl_3$ and d_4-methanol solution, (in ppm with resonance intensities in parentheses)

Carbon species	Solvent/solute system		
	$CDCl_3$/free ligand	$CDCl_3$/Sn complex	d_4-methanol/Sn complex
$SnCH_3$		−1.20 (10.2)	−2.79 (13.5)
δ-CH_3 (Leu)	21.91 (17.7) 22.86 (15.4)	22.17 (11.4) 22.94 (13.2)	22.14 (15.0) 23.36 (13.8)
γ-CH (Leu)	24.86 (14.2)	25.05 (10.8)	26.07 (12.5)
β-CH_2 (His)	28.29 (4.7) 28.45 (1.6)	28.02 (5.1)	29.70 (8.4)
β-CH_2 (Leu) (*cis*-amide)	33.84 (2.5)	33.99 (1.8)	34.72 (1.2)
β-CH_2 (Leu) (*trans*-amide)	40.93 (10.2)	40.99 (8.4)	41.69 (11.6)
OCH_3	52.46 (11.1)	52.56 (3.3)	52.77 (11.6)
CH(α,α′)	52.79 (13.0) 52.86 (9.5)	53.01 (10.2) 53.22 (8.1)	53.76 (10.6) 53.94 (11.2) 55.17 (0.9)
o-Benzoyl *cis*-amide	110.98 (1.6)	111.70 (0.9)	112.01 (1.1)
4′ Benzoyl (*cis*-amide)	117.49 (0.6)	118.05 (0.9)	121.0 (0.1)
Imidazole C-5	118.46 (1.6) 119.16[a] (2.0)	121.3[b] (0.6)	118.96 (2.8)
m-,*p*-Benzoyl (*cis*-amide)	124.01 (1.7) 124.77 (1.9)	124.41 (0.9) 124.69 (0.9)	125.43 (1.3) 126.05 (1.1)
o-,*m*-Benzoyl (*trans*-amide)	127.32 (26.5) 128.45 (26.1)	127.45 (25.8) 128.59 (23.7)	128.52 (26.4) 129.50 (25.1)
Imidazole C-4	130.73[a] (1.4) 133.71 (2.0)	130.1[b] (0.6)	133.84 (1.4)
p-Benzoyl (*trans*-amide)	131.82 (9.5)	132.01 (9.3)	132.81 (12.2)
4′ Benzoyl (*trans*-amide)	133.47 (6.5)	133.57 (5.7)	135.22 (3.9
Imidazole C-2	135.27[c] (1.9)	135.85 (5.1)	136.37 (6.2)
CO_2Me	167.97 (2.4) 168.06 (9.0)	168.09 (6.3)	170.17 (4.1)
NHCO (amide)	171.35 (8.6) 171.79 (2.1)	171.27 (5.3)	172.96 (4.5) 174.23 (0.8)
NHCO (peptide)	172.79 (8.2)	172.61 (6.0)	174.84 (4.8) 176.56 (0.6)

Note: All resonance intensity values are normalized both within and between Tables 1 and 2, i.e. all six spectra are directly comparable in terms of peak intensity data.

[a], Broadened. [b], Very broadened. [c], Coincident peaks, one broadened and one sharp.

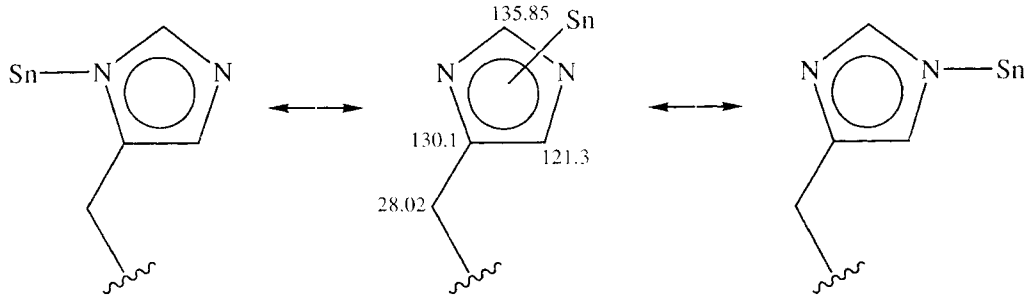

Figure 3 Chemical shift assignments for the coordination equilibrium of complex **I** in $CDCl_3$ solution.

The fourth coordination at tin is clearly by an imidazole ring nitrogen, which gives rise to the spectrum's most interesting features. In contrast to the limited changes in ^{13}C resonances for most of the ligand carbon atoms upon complexation in both shift position and intensity (except CO_2Me), the resonances associated with the carbon atoms of the histidine side chain alter markedly. These resonance features may be rationalized by the coordination equilibrium illustrated in Fig. 3. The equilibrium is fast on the NMR timescale. This equilibrium satisfactorily accounts for the massive line broadening of the C-4 and C-5 resonances, brought about by the pronounced change in the magnetic environment of each by chemical exchange. The C-4 resonance shifts to higher field at 130.1 ppm and the C-5 resonance to lower field at 121.3 ppm, compared with the free ligand. The magnetic environment about the C-2 carbon remains reasonably equivalent, thus yielding a sharp and intense resonance at 135.85 ppm shifted rather less upon complexation, by 0.58 ppm to lower field. It might be imagined that the imidazole ring species is a spinning ligand, in order to accommodate alternate N-atom coordination to tin. The β-$\underline{C}H_2$(His) resonance also exhibits a slight but noticeable broadening. Although this is not very pronounced, the resonance is shifted to higher field at 28.02 ppm, in contrast to the general trend to slightly lower field exhibited by all other aliphatic carbons. However, in the proton NMR the β-C$\underline{H}_2$(His) proton resonance is not seen to shift upon complexation at 3.20 ppm and the coupling to the αCH(His) proton is uninterupted with $J = 4.5$ Hz. Upon complexation the H-5 singlet moves from 6.38 to 6.74 ppm. Owing to the crowded aromatic region in which the H-2 resonates, unambiguous assignments are less easy, but the H-2 proton seems to persist as a singlet at 7.33 ppm.

The identity of the fifth coordination to tin may only be implied. Worthy of note is the anomalous decrease in intensity of the $CO_2\underline{C}H_3$ species upon complexation at 52.56 ppm (presumably as a consequence of changes in tumbling and therefore relaxation lifetimes of the carbon nucleus). Additionally the ester carbonyl resonance moves slightly to lower field upon complexation, in contrast to the slight shifts to higher field in the cases of the peptide and amide carbonyls, although this effect is by no means pronounced. The $CO_2C\underline{H}_3$ proton signal remains unaltered upon complexation, a singlet at 3.75 ppm. On the basis of peak intensities, the *cis/trans* equilibrium about the amide bond moves towards the already predominant *trans* rotamer upon complexation.

In summary, the ^{13}C NMR spectrum of the 1:1 complex indicates monomolecular five-coordination to tin, with the imidazole ring nitrogens indulging in dynamic equilibrium on the NMR timescale in mode of coordination to tin, exchanging between N-1 and N-3 coordination. The fifth coordination is thought to be by the O-atom from the ester carbonyl.

O-atom ester carbonyl coordination to tin centres is normally considered to be weak. To test the hypothesis as to the fifth coordination, the ^{13}C NMR of complex **I** in d_4-methanol as solvent was recorded, and the spectrum is illustrated in Fig. 4 with a full listing of chemical shift data presented in Table 1. This solvent may be considered to compete successfully with ester carbonyl coordination to tin. The spectrum indicates this to be so, with full recovery in resonance intensity associated with the $CO_2\underline{C}H_3$ nucleus at 52.77 ppm. Complexation behaviour in this solvent also displays additional features of interest, not least a preferential mode of imidazole ring nitrogen coordination to tin. For d_4-methanol as solvent, only one $Sn\underline{C}H_3$ species is observed at -2.79 ppm, possessed of clearly resolv-

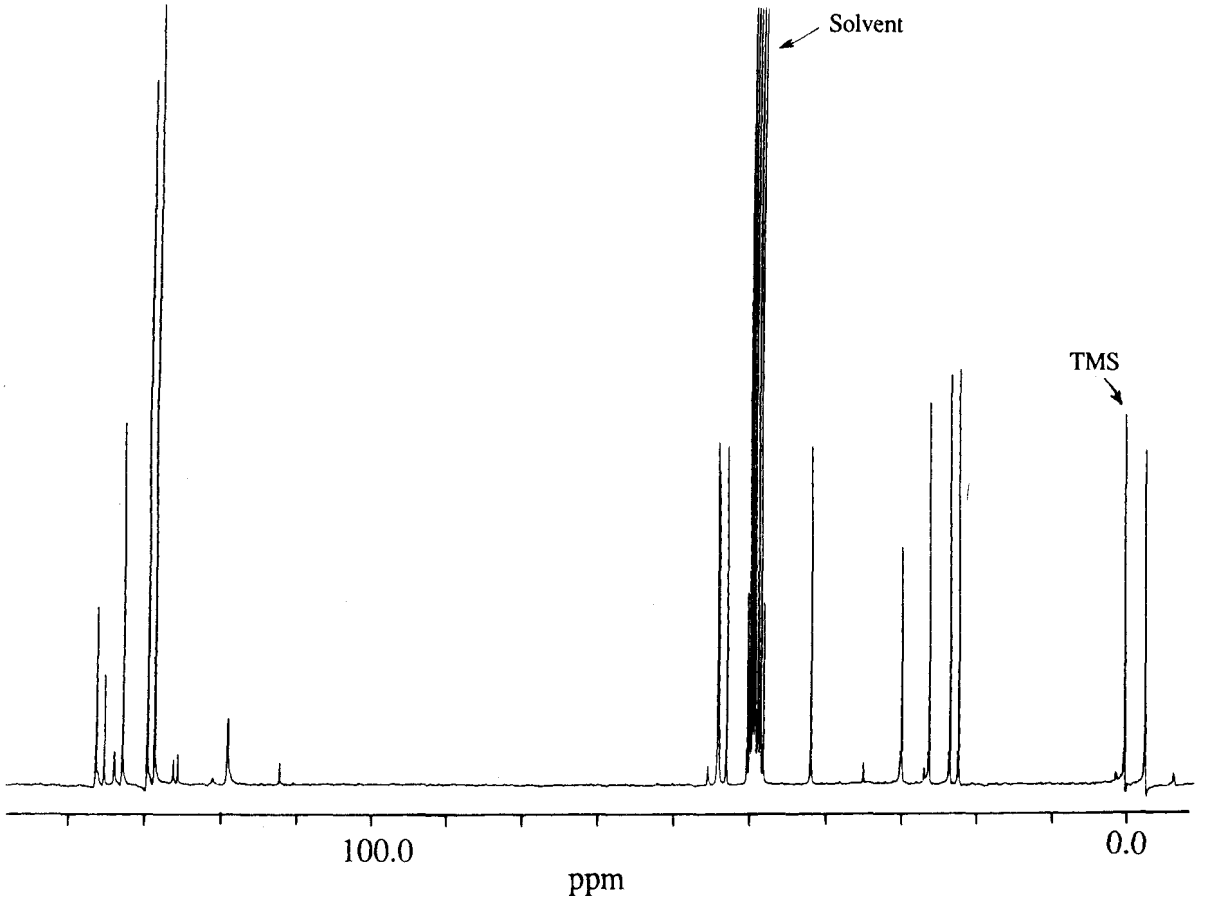

Figure 4 ^{13}C NMR spectrum for complex **I** in $DOCD_3$ solution.

ed satellites with coupling constants $^1J(^{119}Sn-^{13}C)$ = 493.1 Hz and $^1J(^{117}Sn-^{13}C)$ = 471.2 Hz. These values indicate five-coordination about tin. The clear resolution of ^{119}Sn and ^{117}Sn satellites and the expected ratio of their respective coupling constants of 1.046 indicates that for methanol as solvent there exists only one $Sn\underline{C}H_3$ species on the NMR timescale, i.e. no pronounced dynamic ligand exchange at tin operates, in contrast to the case of $CDCl_3$ as solvent.

With regard to the ^{13}C resonances associated with the histidine side chain, and in particular the imidazole ring species, only three sharp resonances are observed. This, together with the clearly resolved satellites for $Sn\underline{C}H_3$, identifies a specific preferential mode of ring nitrogen coordination to tin. The suggested coordination solution structure is presented in Fig. 5. The C-2 resonance at 136.37 ppm is sharp and notably intense, as are those of the C-4 and C-5 atoms at 133.84 and 118.96 ppm respectively, the relative intensity of the three resonances being as expected for this ring system, i.e. C-2 > C-5 > C-4. Also the β-$\underline{C}H_2$(His) resonance is sharp and notably intense at 29.70 ppm. The suggested mode of coordination illustrated in Fig. 5, i.e. N-1 atom coordination to tin, cannot be considered unequivocal. However, it is considered to be the firmest rationalization of all resonance positions, given the magnetic environment for ring carbons

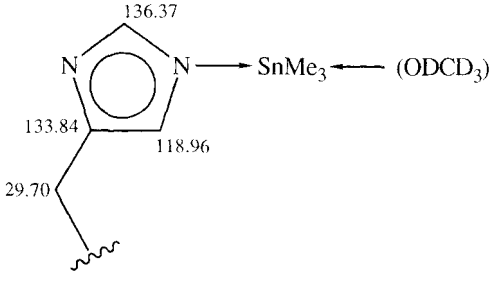

Figure 5 Chemical shift assignments for the coordination isomer of complex **I** dominant in methanol solutions.

engendered by studies on *N*-methyl derivatives of histidine side chains of small peptides.[21,22]

The shift behaviour and relative intensities of the quaternary benzene ring carbons for both *cis* and *trans* amide bond rotamers is worthy of note, and is presumably a feature of methanol as solvent. Also the appearance of minor resonances in the α-CH and carbonyl regions suggests a minor degree of *cis/trans* isomerism about the peptide bond for methanol as solvent.

In summary, for d_4-methanol as solvent, ligand complexation to the trimethyltin moiety is via the N-1 atom of the imidazole ring. The structure about tin is five-coordinate, presumably *trans* trigonal bipyramidal, with O-atom coordination by one molecule of solvent providing the fifth ligand. The complex does not exhibit dynamic equilibrium features on the NMR timescale.

For the biological medium in which trialkyltin complexation to protein structures occurs we must consider a solvent environment which is essentially aqueous. In such a medium it will be the competitive role of the trialkyltin binding site with respect to an aqueous R_3Sn species which determines the magnitude of binding affinities. In this study, neither the free ligand nor the complex **I** are soluble in aqueous solution. However, in order to investigate the likely consequences of such interactions, the ^{13}C NMR spectra for the complex were recorded for d_6-DMSO and d_6-DMSO/H_2O as solvents. The ^{13}C NMR spectrum for complex **I** in pure d_6-DMSO solution is reproduced in Fig. 6 with full listing of chemical shift data reported in Table 2, together with those for the free ligand in this solvent, reported earlier.[16]

Only one $Sn\underline{C}H_3$ species is observed on the NMR timescale at +0.21 ppm with clearly resolved satellites yielding $^1J(^{119}Sn-^{13}C)$ = 535.5 Hz and $^1J(^{117}Sn-^{13}C)$ = 511.4 Hz, indicative of five-coordination. Similarly, the proton spectrum shows one only $SnC\underline{H}_3$ resonance at +0.40 ppm with clearly resolved satellites yielding $^2J(^{119}Sn-C-^1H)$ = 65.7 Hz and $^2J(^{117}Sn-C-^1H)$ = 62.7 Hz. As such, the equilibrium magnetic environment about tin appears to persist, despite evidence for alternate N-atom coordination to tin (*vide infra*), on the NMR timescale. In the proton NMR spectrum, the resonances associated with the ligand aromatic protons are drawn out over a larger range, thus enabling unambiguous assignment.

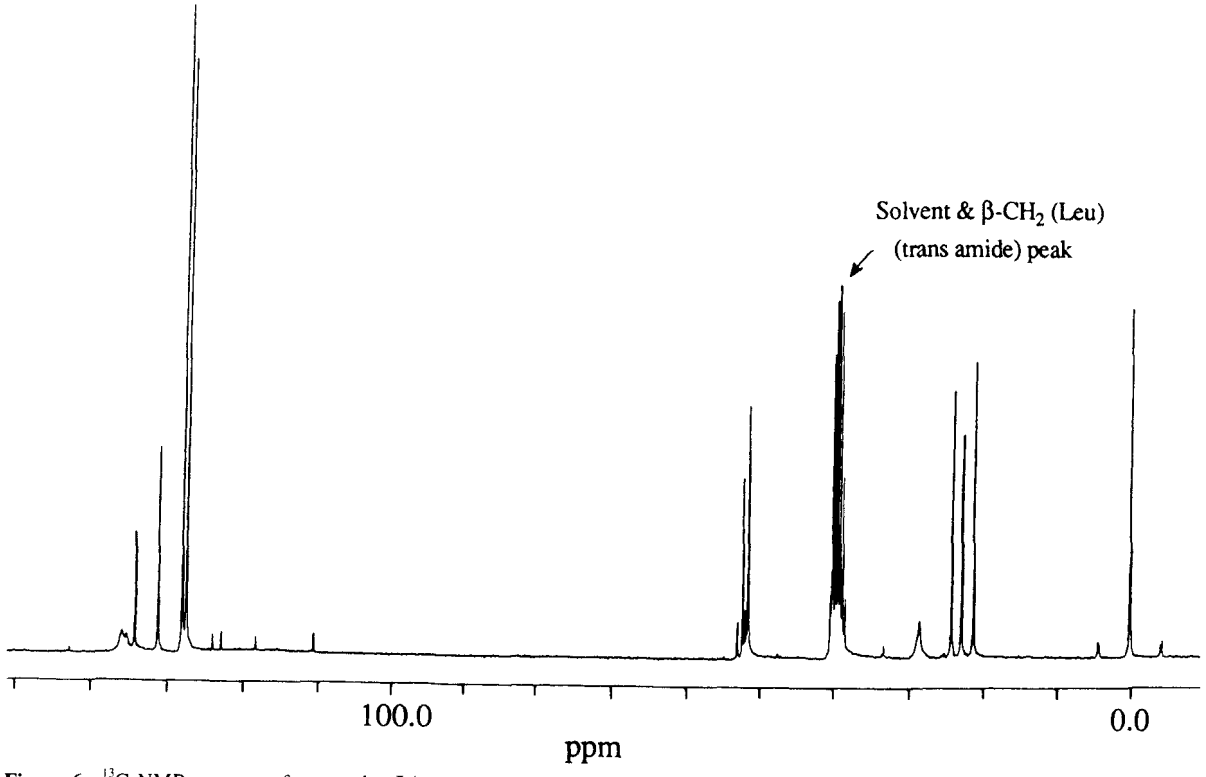

Figure 6 ^{13}C NMR spectrum for complex **I** in pure DMSO solution.

Table 2 ^{13}C NMR chemical shift data for complex **I** in pure d_6-DMSO and d_6-DMSO/H_2O solution (in ppm with resonance intensities in parentheses)

Carbon species	Solvent/solute system		
	d_6-DMSO/free ligand	d_6-DMSO/Sn complex	d_6-DMSO/H_2O)/Sn complex
$SnCH_3$		0.21 (15.0)	0.45 (22.4)
δ-CH_3 (Leu)	21.50 (12.7)	21.44 (12.8)	22.15 (13.9)
	22.97 (11.8)	22.98 (9.8)	23.56 (12.0)
γ-CH (Leu)	24.44 (12.3)	24.36 (11.5)	25.24 (9.9)
β-CH_2 (His)	28.44 (6.8)		29.04 (4.7)
	28.61 (1.2)	28.62[a] (1.3)	29.51 (1.4)
β-CH_2 (Leu) (*cis*-amide)	33.34 (0.8)	33.30 (0.5)	34.02 (0.5)
β-CH_2 (Leu) (*trans*-amide)	40.24 (9.8)	40.33[b]	40.88 (8.2)
OCH_3	51.78 (15.0)[c]	51.67 (10.7)	52.97 (9.9)
CH(α,α′)	51.78	51.59 (8.8)	52.83 (8.3)
	52.38 (9.4)	52.35 (7.6)	54.36 (1.6)
		53.06 (1.5)	
o-Benzoyl *cis*-amide	110.19 (0.9)	110.72 (0.9)	111.96 (0.5)
Imidazole C-5	116.64 (3.9)	Broadened into baseline; almost disappears	118.04[a] (1.5)
			119.84[a] (0.5)
4′ Benzoyl (*cis*-amide)	118.77 (0.7)	118.36 (0.6)	118.83 (0.8)
m-,*p*-Benzoyl	123.73 (0.6)	122.87 (0.9)	124.80 (0.5)
(*cis*-amide)	125.67 (0.7)	123.98 (0.8)	125.40 (0.9)
o-,*m*-Benzoyl	127.61 (25.3)	127.51 (25.1)	128.15 (25.1)
(*trans*-amide)	128.17 (28.2)	128.07 (28.1)	129.29 (24.4)
p-Benzoyl (*trans*-amide)	131.29 (8.9)	131.18 (9.1)	132.61 (8.8)
Imidazole C-4	132.92 (2.4)	Not observed	133.40[a] (1.3)
4′ Benzoyl (*trans*-amide)	134.26 (7.3)	134.18 (5.4)	134.31 (7.7
Imidazole C-2	134.75 (4.0)	135.21 (0.7)	135.53 (1.4)
		135.81 (0.9)	136.00 (4.8)
CO_2Me	166.62 (7.1)	166.39 (7.5)	168.33 (6.9)
NHCO (amide)	171.71 (8.2)	171.44 (2.5)	172.62 (6.0)
	171.83 (1.1)	171.71 (6.2)	172.94 (2.3)
NHCO (peptide)	172.46 (7.1)	172.27 (7.8)	173.80 (7.8)
		173.86 (0.9)	175.21 (1.4)

Note: All resonance intensity values are normalized both within and between Tables 1 and 2, i.e. all six spectra may be directly compared in terms of peak intensity data.

For the free-ligand spectrum above, the minor tautomers of imidazole have been omitted for clarity (see Ref. 10).

[a], Broadened. [b], Accurate measurement of intensity precluded by overlapping solvent peaks. [c], Distinguished by DEPT.

The H-5 proton suffers massive line broadening and reduction in peak height intensity upon complexation, and is observed as a broad hump in the region 6.8 ± 0.1 ppm, as compared with the sharp singlet at 6.96 ppm for the free ligand. The H-2 proton resonance shifts upfield from 7.64 to 7.38 ppm upon complexation with minor broadening. ^{1}H NMR resonances show no other changes upon complexation, including that associated with β-C$\underline{H}_2$(His) which remains unchanged at 3.00 ppm (J = 6 Hz). The massive line broadening of the H-5 resonance indicates a dynamic equilibrium between two magnetically inequivalent environments of the H-5 atom by chemical exchange. Similar features have been noted for histidine imidazole ring nitrogen coordination to transition metals in DMSO soltuion.[23] No evidence for H-2 proton acidity was manifest.[24]

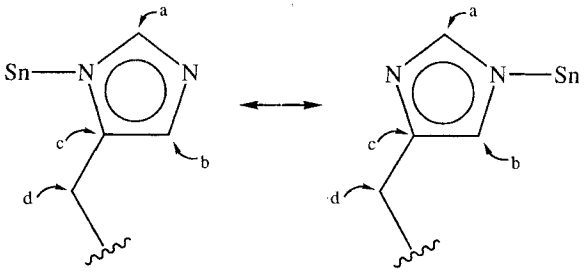

Figure 7 Chemical shift assignments for the coordination equilibrium of complex **I** in pure DMSO solution. (a) C-2 resonances, broad at 135.21 and 135.81 ppm. (b) C-5 resonance broadened into the baseline in the region 113–121 ppm. (c) C-4 resonance not observed. (d) β-CH_2(His) resonance broad at 28.62 ppm.

With regard to the ^{13}C NMR spectrum for complex **I** in pure d_6-DMSO solution, a number of features present themselves. These include: (a) the considerable broadening of the β-C$\underline{H}_2$(His) resonance at 28.62 ppm and concomitant reduction in intensity (unique in our experience); (b) the broadening of the imidazole C-5 resonance almost completely into the baseline in the region 113–121 ppm; (c) the disappearance of the resonances associated with the C-4 carbon species altogether; and (d) the appearance of two very broadened C-2 resonances, both of reduced intensity. These features may be successfully rationalized by the equilibrium displayed in Fig. 7.

As with d_4-methanol as solvent, some suggestion of minor *cis/trans* isomerization about the peptide bond is evident. All other features of the spectrum change little upon complexation as compared with the free ligand in the same solvent (in which the molecule of water of hydration is cleaved).[16] The fifth coordination at tin is presumably by solvent.

The consequences of the addition of water to the

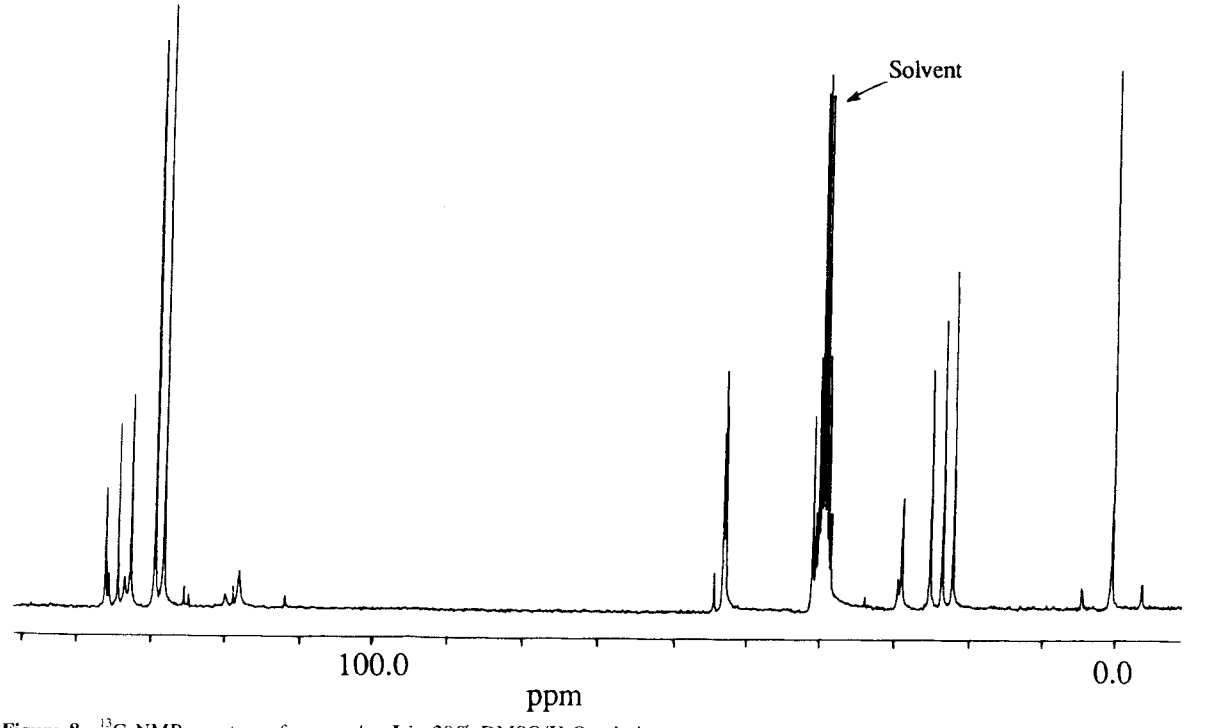

Figure 8 ^{13}C NMR spectrum for complex **I** in 20% DMSO/H_2O solution.

DMSO solution (20% water/d_6-DMSO by volume) are illustrated in Fig. 8 and full chemical shift data for this spectrum presented in Table 2. The addition of water would appear to alter the rate of exchange between coordination isomers, and also to protonate the N-atom remote to N-atom coordination to tin, which itself presumably becomes an hydroxylated species.

Once again, only one $Sn\underline{C}H_3$ species is observed on the NMR timescale with a single resonance at +0.45 ppm possessing clearly resolved satellites with coupling constants $^1J(^{119}Sn-^{13}C)$ = 517.2 Hz and $^1J(^{117}Sn-^{13}C)$ = 494.4 Hz. The resonances associated with the histidine side chain now split into two groups, attributed to the two structures given in Fig. 9. However, it must be stressed that owing to the similarities in the two sets of resonances, specific identification of one set with the particular coordination species (i.e. via N-1, Fig. 9b, or via N-3, Fig. 9a) is interchangeable as illustrated in Fig. 9. That two sets exist is witnessed by relative peak intensities, and that one preferential mode of coordination is favoured relates to this feature. The two structures and their associated resonances are the most rational comparisons to the protonated forms of *N*-methyl derivatives of simple peptides.[21,22] For each structure the intensity ratios fall in the order C-2 > C-5 > C-4, which presumably accounts for why C-4 in structure (a), as illustrated in Fig. 9, is not observed. In contrast to pure d_6-DMSO as solvent, the resonances associated with the β-$\underline{C}H_2$(His) species are now resolved.

The fact that in aqueous/d_6-DMSO solution, the N-atom on the imidazole ring remote to tin coordination is protonated, is most probably a reflection of the fact that the pK_a for the coordinated imidazole ring is intermediate between the pK_a of water and methanol (which latter solvent was not observed to protonate the ring).

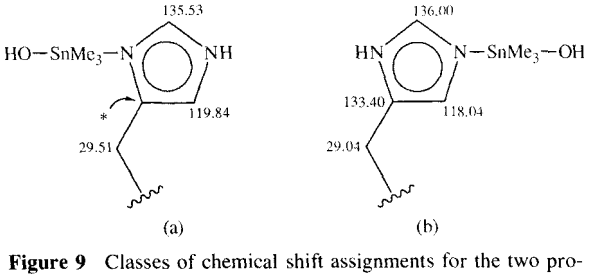

Figure 9 Classes of chemical shift assignments for the two proposed isomers of complex **I** in DMSO/H_2O solution (*expected to be weak and possibly occluded by overlapping intense phenyl C-atom resonances).

CONCLUSIONS

The complexation behaviour of trimethyltin with histidine residues in solution has been demonstrated to be complex. It would appear that ^{13}C NMR is a more useful technique than ^1H NMR as an investigative tool, with the former indicating features not apparent in the latter.

That coordination to tin by the imidazole moiety of histidine leaves the tin centre still sufficiently Lewis-acidic to in nature allow further coordination by even such weak donors as ester carbonyl, suggests that four-coordination is disfavoured in general. In this respect, i.e. the requirement for a fifth donor in addition to coordination by histidine in solution, the system would appear to parallel features previously noted for these complexes in the solid state.[25] It would seem that the stereochemical geometries of the ligands to tin, additionally, dictate the kinetics of equilibria phenomena (i.e. binding affinities). It might be noted that variance of the alkyl group ligands on tin and its net result on the Lewis acidity of the tin centre (inductive effect) will play a role in addition to any hydrophobic interaction, thus determining the specific requirements upon the nature of the binding site of the enzyme. It is evident that the required ligand geometry of binding site must be such as to compete with water for coordination to the tin centre in an aqueous environment.

These factors reflect the specificity for organotins in forming stable complexes with relatively few proteins and their selective biocidal potencies to differing animal species.

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