APPLIED ORGANOMETALLIC CHEMISTRY, VOL. 7, 407–412 (1993)

Diorganotin(IV) dipeptide complexes with potential antitumour activity

G Guli,* G Gennaro,* L Pellerito* and G C Stocco*†‡
* Dipartimento di Chimica Inorganica and † Facoltà di Farmacia, Università di Palermo, 90123 Palermo, Italy

A series of complexes of the dimethyltin(IV) moiety with dipeptides has been synthesized. The dipeptides are L-alanyl-L-histidine (H_2AlaHis), L-methionyl-L-methionine (H_2MetMet), L-glycyl-L-histidine (H_2GlyHis) and L-histidyl-L-glycine (H_2HisGly). The complexes have been characterized by IR and ^{119}Sn Mössbauer spectroscopy in the solid state and by ^{1}H NMR in CD_3OD and D_2O solutions. They consist of monomeric entities, with the tin atom arranged in a pentacoordinated trigonal bipyramidal structure. The dipeptides are coordinated via the amino group, deprotonated peptide nitrogen and carboxylate group. Neither imidazole in histidine-containing dipeptides, nor the thioether group in MetMet, is involved in bonding; they act as pendant arms on the outer surface of the complexes.

Keywords: Diorganotin(IV), dipeptide complexes, structure, IR spectra, Mössbauer spectra, ^{1}H NMR spectra

INTRODUCTION

The chemistry and biological properties of dipeptides and metal ions have been extensively reviewed.[1,2]

The histidine residue of peptides, besides being involved in metal complexation, appears to play a crucial role in peptide structures and folding; hence stabilization of the orientation of the DNA binding surface of the protein occurs, as in the case of 'Zinc finger' proteins.[3] It is tempting to suggest that simple complex molecules (i.e. complexes of dipeptides with metal ions) may specifically interact with biomolecules like DNA via a pendant arm such as the imidazole of histidine, which may be effective in DNA recognition.[4]

The title compounds could be tested for antitumor activity, analogous to that observed in the case of diorganotin(IV) glycylglycinates R_2SnGlyGly (R = Me, n-Bu, n-Oct, Ph).[5]

‡ Author to whom correspondence should be addressed, at Dipartimento di Chimica Inorganica, Via Archirafi 26, 90123 Palermo, Italy.

EXPERIMENTAL

The dipeptides used throughout this work are L-alanyl-L-histidine (H_2AlaHis), L-methionyl-L-methionine (H_2MetMet), L-glycyl-L-histidine (H_2GlyHis) and L-histidyl-L-glycine (H_2HisGly).

L-Dipeptides were purchased from Sigma Chemical Company (St Louis, Mo, USA) and used without further purification.

The solvent (methanol; MeOH) was dried by standard methods, and syntheses were carried out under exclusion of moisture.

Freshly prepared Me_2SnO was obtained by hydrolysis of Me_2SnCl_2 dissolved in water, by treatment with 25% (w/w) aqueous ammonia solution.

The compounds were synthesized by refluxing, for 4 hr, 2 mmol of Me_2SnO and 2 mmol of the appropriate dipeptide H_2L in 30–50 cm^3 of dry MeOH. The solvent was reduced in vacuum to a small volume (*ca* 5 cm^3) and the solids were precipitated by addition of diethyl ether. The white solids were collected on a sintered glass filter, washed with diethyl ether and small amounts of cold dry MeOH, and stored in a vacuum desiccator over P_4O_{10}. Solubility in MeOH for the Me_2SnL complexes was good, while it was distinctly less so in D_2O.

Elemental microanalyses were performed by Dipartimento di Chimica Organica e Industriale, University of Milan, and are reported along with molecular weights osmometrically determined for selected compounds, in Table 1.

Infrared spectra were recorded as split mulls on a Perkin–Elmer model 983G instrument (Table 2).

^{119}Sn Mössbauer spectra (Table 3) were measured with a Laben 8001 multichannel analyzer, an MWE velocity transducer (Wissenschaftliche

0268–2605/93/060407–06 $08.00

Received 25 May 1993
Accepted 15 July 1993

Table 1 Analytical data for Me_2Sn–dipeptide complexes, Me_2SnL

Compound	M.p. (°C)	Analysis (%): Found (Calcd) C	H	N	MW[a]
$Me_2SnMetMet$	218–220	33.80 (33.74)	5.56 (5.66)	6.56 (6.56)	
$Me_2SnAlaHis$	180–185	35.60 (35.42)	5.12 (4.86)	14.77 (15.02)	
$Me_2SnGlyHis$	180d	33.87 (33.45)	4.60 (4.49)	15.12 (15.60)	346 (359)
$Me_2SnHisGly$	169d	32.94 (33.45)	4.31 (4.49)	15.27 (15.60)	363 (359)

[a] Osmometrically measured in methanol, at 25°C.

Elektronik GmbH, Munchen), an FG2 digital function generator (Wissenschaftliche Elektronik GmbH, Munchen) and an MA250 velocity transducer (Wissenschaftliche Elektronik GmbH, Munchen), moved at linear velocity, constant acceleration in a triangular waveform. A DN700 Oxford cryostat with DTC2 temperature controller was used to maintain the absorber samples (absorber concentration, 0.5–0.6 mg $^{119}Sn/cm^2$) at the temperature of liquid nitrogen.

^1H NMR spectra were recorded on a Bruker W80 instrument, operating at 80.1 MHz, using tetramethylsilane (TMS) or 3-(trimethylsilyl)-1-propane sulphonic acid sodium salt (DSS) as internal standards, and are reported in Table 4.

RESULTS AND DISCUSSION

$Me_2Sn(IV)$Dipeptide complexes in the solid state

For all the complexes, analytical data (Table 1) are consistent with a 1:1 molar ratio of Me_2Sn^{2+} moiety/dipeptide.

Table 2 Some relevant IR frequencies (cm^{-1}) for $Me_2Sn(IV)$–dipeptide complexes

Compound	ν(NH)	$\nu(NH_3^+)$	Amide I	$\nu_{as}(COO^-)$	Amide II	$\nu_{sym}(COO^-)$	$\Delta\nu$
AlaHis	3350 3299	2120	1660	1588	1530	1410	
$Me_2SnAlaHis$	3270 3094	—	1625	1596[a]	—	1405	191
GlyHis	3338 3190 3123	2040	1689	1586	1547	1386	
$Me_2SnGlyHis$	—[b] —[c] —[c]	—	1605[d]	1605[d]	—	1377	228
HisGly	3387 3340	2186	1657	1581	1550	1389	
$Me_2SnHisGly$	—[e] —[f]	—	1604[g]	1604[g]	—	1382	222
MetMet	3324 3211	—	1649	1572	1529	1414	
$Me_2SnMetMet$	3213 3115	—	1641	1603	—	1395	208

AlaHis

[a] Two vibrations, present in the spectrum of the ligand at 1610 and 1588 cm^{-1}, coalesce in a broad band at 1596 cm^{-1}.

GlyHis

[b] Broad band extending from 3300 to 3500 cm^{-1}.

[c] Broad band extending from 3000 to 3300 cm^{-1}.

[d] Two vibrations coalesce in broad band with a maximum at 1605 cm^{-1}, extending from 1580 to 1650 cm^{-1}.

HisGly

[e] Broad band extending from 3300 to 3500 cm^{-1}.

[f] Broad band extending from 3000 to 3300 cm^{-1}.

[g] Two vibrations coalesce in broad band with a maximum at 1604 cm^{-1}, extending from 1580 to 1650 cm^{-1}.

Table 3 ^{119}Sn Mössbauer parameters for $Me_2Sn(IV)$–dipeptide complexes at liquid-nitrogen temperature

Compound	δ[a] (mm s^{-1})	δE[b] (mm s^{-1})	Γ[c] (mm s^{-1})	C–Sn–C angle[d] (degrees)
$Me_2SnAlaHis$	1.25	3.20	1.09	133
$Me_2SnGlyHis$	1.17	2.92	1.05	124
$Me_2SnHisGly$	1.17	2.93	1.09	124
$Me_2SnMetMet$	1.16	2.94	1.03	125

[a] Isomer shift relative to room-temperature $Ca^{119}SnO_3$.
[b] Nuclear quadrupole splitting.
[c] Full width at half height of the resonant peaks, average.
[d] C–Sn–C angle values calculated for $Me_2Sn(IV)$–dipeptide complexes according to point-charge model.

The infrared spectra of the complexes (Table 2) afford a way of assessing which potential binding sites of the dipeptides are involved in complexation.

Vibrations associated with terminal NH_3^+ groups[6] in the ligands (2000–2200 cm^{-1}) are missing in the complex compounds and appear as $\nu(NH_2)$ vibration at 3100–3300 cm^{-1}.

Frequencies associated with carboxylate groups are shifted upon coordination: $\nu_{as}(COO^-)$ is shifted to higher and $\nu_{sym}(COO^-)$ to lower frequency with respect to the free ligands. The values of $\Delta\nu = [\nu_{as}(COO^-) - \nu_{sym}(COO^-)]$ are in the range 190–230 cm^{-1}, suggesting that the carboxylate groups are coordinating in a monodentate fashion.[7]

The $\nu(CO_{pept})$ (amide I band) is shifted to lower frequencies, while the amide II band, which involves the peptide group N–H deformation,[7] disappears.

The infrared data suggest that dimethyltin(IV) is bonded to the terminal amino and carboxylate groups and to deprotonated peptide nitrogen.

^{199}Sn Mössbauer data are reported in Table 3. All of the complexes give a two-line quadrupole spectrum, with full width at half height of the resonant peak values, Γ (mm s^{-1}), which strongly support the occurrence of only one tin environment. The isomer shift values, δ (mm s^{-1}), are typical of diorganotin derivatives.[9,10]

The experimental nuclear quadrupole splitting parameters, ΔE (mm s^{-1}), indicate a trigonal bypiramidal arrangement around the tin atom.[9,10] ΔE values range from 3.20 mm s^{-1} for $Me_2SnAlaHis$ to 2.92 mm s^{-1} for $Me_2SnGlyHis$.

These findings have been tentatively attributed mainly to C–Sn–C angle variations occurring in the complexes. Mössbauer data have been rationalized through a literal version of a point-charge model calculation of ΔE to give C–Sn–C values of the individual R_2SnL species (Table 3).[11–14]

The partial quadrupole splitting values, p.q.s. (mm s^{-1}), employed in the calculations are:

$$\{Alk\}^{tbe} = -1.13 \qquad \{NH_2\}^{tba} = +0.01$$
$$\{N_{pept}\}^{tbe} = -0.30 \qquad \{COO^-\}^{tba} = +0.075$$

Regular trigonal bipyramidal structures are considered, except for C–Sn–C angles that are allowed to vary.

The dimethyltin(IV) moiety and deprotonated peptide nitrogen lie on the equatorial plane, while the amino and carboxylate groups occupy apical positions (Fig. 1). This is consistent with the X-ray molecular structures for $Me_2SnAlaHis$ and $Me_2SnMetMet$ which have been determined and

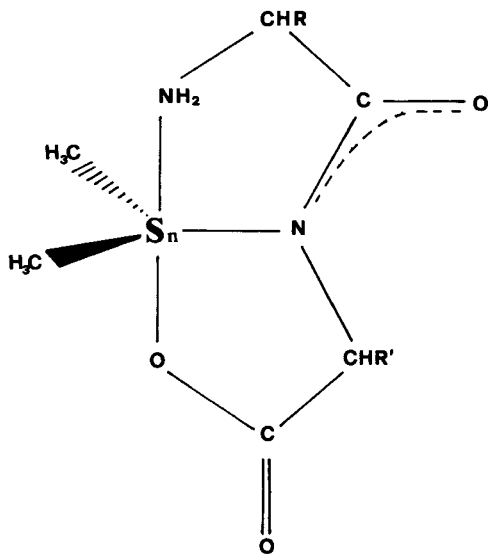
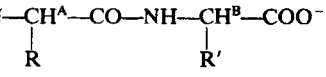


Figure 1 Structure proposed for the dimethyltin(IV)–dipeptide complexes.

Table 4 ^{1}HNMR data of dimethyltin(IV)–dipeptides



Compound	A	B	C	D	E	Me	δ(CH$_3$Sn) (ppm)	|^2J(^{1}H–^{119}Sn)| (Hz)	Solvent
H$_2$AlaHis[a]	3.50q	4.20t	3.15m	6.52s	7.45s	1.40d			D$_2$O[b]
R = Me									
R′ = CH$_2^C$—C=CH^D—NH—CH^E=N)									
H$_2$AlaHis	3.66q	4.45t	3.12m	7.00s	7.82s	1.49d			CD$_3$OD[c]
Me$_2$SnAlaHis	3.59q	4.40t	—[d]	6.77s	7.57s	1.52d	0.69s	77.1	CD$_3$OD[c]
							0.34s	80.7	
Me$_2$SnAlaHis	3.71q	4.42t	3.29m	6.87	7.82s	1.43d	0.71s	80.7	D$_2$O[b]
							0.27s	80.1	
	—[e]	—[e]	—[e]	7.04s	7.97s	—[e]	0.70s	—[e]	
H$_2$GlyHis	3.69s	4.49t	3.03m	6.99s	7.84s				D$_2$O[b]
R = H^A									
R′ = CH$_2^C$—C=CH^D—NH—CH^E=N)									
H$_2$GlyHis	3.65s	4.55t	3.12m	6.91s	7.68s				CD$_3$OD[c]
Me$_2$SnGlyHis	3.53d	4.44t	—[d]	7.12s	8.36s		0.73	80.8	CD$_3$OD[c]
							0.45s	80.4	
Me$_2$SnGlyHis	3.68d	4.59t	3.33m	7.17s	8.35s		0.84s	81.7	D$_2$O[b]
							0.43s	81.2	
	3.91s	—[e]	—[e]	7.29s	8.47s		0.83s	—[e]	
H$_2$HisGly	4.02t	3.82s	3.17m	7.06s	7.85s				CD$_3$OD[c]
R = CH$_2^C$—C=CD^D—NH—CH^E=N)									
R′ = H^B									
Me$_2$SnHisGly[f]	4.04t	3.82s	3.22m	7.10s	7. 96s		0.72s	78	CD$_3$OD[c]
							0.45s	82	
H$_2$MetMet	4.15t	4.30dd	2.07m	2.58m		2.15s			D$_2$O[b]
R = CH$_2^C$—CH$_2^D$—S—Me						2.12s			
R′ = CH$_2^{C'}$—CH$_2^{D'}$—S—Me									
HMetMet$^-$	3.53t	4.29dd	1.98m	2.59m		2.12s			D$_2$O[b] + NaOD
						2.11s			
Me$_2$SnMetMet	3.62dd	4.32t	2.19m	2.63m		2.17s	0.77s	79.6	CD$_3$OD[c]
						2.11s	0.89s	79.5	
Me$_2$SnMetMet	3.85dd	4.42t	2.29m	2.58m		2.23s	0.87s	79.9	D$_2$O[b]
						2.18s	1.01s	81.2	
	—[e]	4.23t	—[e]	—[e]		—[e]	0.89s	84.9	

Abbreviations: s, singlet; d, doublet; dd, doublet of doublets; t, triplet; q, quartet; m, multiplet.

[a] From Ref. 14.

[b] Internal reference DSS.

[c] Internal reference TMS.

[d] Obscured by the multiplet of non-deuterated CD$_3$OD, at 3.35 ppm.

[e] Signals associated with the hydrolyzed species, present in low concentration due to the limited solubility of the complex in D$_2$O, partially overlapped with those of the parent complex.

[f] Not recorded in D$_2$O, owing to the low solubility of the complex.

are reported elsewhere.[14] The same general pentacoordinated structure is reported for several R$_2$Sn(IV) complexes of dipeptides.[15–19]

According to the point-charge formalism, C–Sn–C angles have been extracted (Table 3). On the other hand, as far as Me$_2$SnAlaHis and Me$_2$SnMetMet are concerned, predicted ΔE values, calculated using experimental C–Sn–C angles[14] (143.9° and 132° respectively), differ less than the allowed ± 0.4 mm s^{-1} proposed for the idealized structures, used for the above-mentioned calculations.[11]

It is noteworthy that in the X-ray structure of the Et$_2$SnGlyHis complex,[20] two different molecules appear: one in which tin is pentacoordinated in the above-mentioned fashion, whilst in the

other molecule the tin(IV) atom attains hexacoordination through bonding with N_{amino}, $O_{carboxylate}$ and αC(Et) atoms in the equatorial plane and $N_{peptide}$ (deprotonated) and N_{imide} (imidazole group of histidine of the neighboring molecule) in the apical positions. This finding prompted us to determine the molecular weights of $Me_2SnGlyHis$ and $Me_2SnHisGly$ in dry MeOH (see Table 1); these gave us evidence that no dimeric species were present in either case. For $Me_2SnAlaHis$ and $Me_2SnMetMet$ the X-ray structure determination[14] rules out such interactions; hence the imidazole and thioether are acting as pendant arms on the exterior surface of the complexes.

Me_2Sn–dipeptide complexes in solution

^1H NMR spectra for Me_2Sn(IV)–dipeptide complexes were measured both in CD_3OD and D_2O (Table 4). The spectra in D_2O were intended to provide an insight into the stability of the complexes in water. For histidine-containing peptides, assignments were made according to Ref. 21. The following information can be gained from the spectra.

(a) In CD_3OD solution the complexes exist in the form of undissociated monomers, since single sets of peaks for the individual protons [CH_3(Ala), CH_3(Met), CH_2 and CH] appear. The shifts which are detected, relative to the free dipeptide, give a way of assessing which binding sites are involved and are indicative of a structure in solution which is similar (*vide infra*) to that of solid-state complexes.

It is noteworthy that the chirality of the complexes around the tin atom is responsible for the appearance of two magnetically non-equivalent (CH_3(Sn) signals, besides giving an indication that pentacoordination is maintained in CD_3OD solution ($|^2J(^1H–^{119}Sn)|$ coupling constant values).[22]

(b) In D_2O solution the signals are generally doubled, which is consistent with a partial dissociation (*ca* 10%) of the dipeptide complex.[23] For instance, in histidine-containing derivatives (see Table 4) the signals due to the ligand in the hydrolyzed species occur downfield relative to those of the parent complex.

For spectra of $Me_2SnMetMet$ in D_2O solution, assignments were made according to the literature[24] and by monitoring the shifts caused by addition of NaOD to the free ligand (see Table 4). Addition of NaOD in a slight excess, relative to the molar amount of MetMet, caused the shift of the H^A signal from 4.15 to 3.53 ppm. Metallation by the organometallic moiety showed a downfield shift of both H^A and H^B signals of the ligand in its anionic form, while those relative to the thioether methyl groups appear nearly unshifted, which rules out any involvement of sulfur in bonding.

For all complexes, the additional peak which is present in the 0–1 ppm region, due to the methyl groups of the organometallic moiety of the hydrolyzed species, is not split into two signals, which gives support to the view that upon hydrolysis the apical COO^- and NH_2 terminal groups have been removed from tin, with water molecules presumably taking their place, and bonding through $N_{peptide}$ is preserved.[23]

The fact that in D_2O solution the structure of the parent tridentate complex is preserved, is confirmed by the constancy of $|^2J(^1H–^{119}Sn)|$ values, at least for the complex $Me_2SnMetMet$ where the solubility in D_2O was high enough to detect ^2J values.

In order to determine C–Sn–C angle values for the organometallic moiety in solutions, Lockhart's relationship between C–Sn–C bond angles and $|^2J(^1H–^{119}Sn)|$ was applied.[22] The results are: $Me_2SnAlaHis$, 129°; $Me_2SnGlyHis$, 131°; $Me_2SnHisGly$, 131°; $Me_2SnMetMet$, 130°.

In general, the calculated C–Sn–C angles do not differ greatly (within 7° or less) from those calculated for the solid complexes by use of the literal point-charge model. A good agreement is found in the case of $Me_2SnMetMet$ where the angle determined by X-ray diffraction (132°) compares favorably with the 130° angle calculated for the solution species. By contrast, in the case of $Me_2SnAlaHis$, where a C–Sn–C angle of 143.9° was determined from the X-ray diffraction study,[14] the same angle appears to be reduced to 129° by calculation.

In conclusion, it appears that dimethyltin(IV) moieties do not interact with imidazole or thioether groups in the context of dipeptide complexes which present the usual pentacoordination. There may be some subtle stereochemical influence exerted by the aliphatic group, if the diethyltin derivative of GlyHis does show such intermolecular interaction.[20] It should be stressed that, in the complexes under investigation containing a histidine residue, the imidazole group, not being involved in coordinating the organometallic ion in the solid or in solution, might interact

with DNA in an intercalative way and/or via hydrogen bonding; this, ultimately, could increase the affinity of the complexes toward the nucleic acid and their ability to inhibit replication. Simultaneous intercalation and covalent coordination to DNA has been demonstrated in some classes of DNA-binding drugs such as psoralens,[25] and it is also a preferred mode of binding in covalent complexes formed on DNA by *cis*-DDP and ethidium bromide.[26] Experiments designed to test the intercalative ability and antitumor activity of the complexes toward DNA are in progress.

The complexes $Me_2SnMetMet$, where the usual coordination by the amino and carboxylate groups and deprotonated peptide nitrogen, with no involvement of sulfur atoms is shown, offers clear evidence of the strikingly different binding ability of sulfur as a thioether (MetMet) relative to thiols, which are the main binding groups both with metallothioneins and glutathione.[2]

Acknowledgement Financial support by the Ministero per l'Università e la Ricerca Scientifica e Tecnologica, Rome, is gratefully acknowledged.

REFERENCES

1. Sundberg, R J and Martin, R B *Chem. Rev.*, 1974, 74: 471
2. Sigel, H and Martin, R B *Chem. Rev.*, 1982, 82: 385
3. Jasanoff, A and Weiss, M A *Biochemistry*, 1993, 32: 1423
4. Sundquist, W I and Lippard, S J *Coord. Chem. Rev.*, 1990, 100: 293
5. Barbieri, R, Pellerito, L, Ruisi, G, Lo Giudice, M T, Huber, F and Atassi, G *Inorg. Chim. Acta*, 1982, 66: 139, and references therein
6. Silverstein, R M and Clayton Bassler, G *Spectrometric Identification of Organic Compounds*, 2nd edn, John Wiley & Sons, New York, chapter 3, p 97
7. Deacon, G B and Phillips, R J *Coord. Chem. Rev.*, 1980, 33: 227
8. Parker, F S *Applications of Infrared Spectroscopy in Biochemistry, Biology and Medicine*, Plenum Press, New York, 1971
9. Bancroft, G M and Platt, R H *Adv. Inorg. Chem. Radiochem.*, 1972, 15: 59
10. Parish, R V Structure and bonding in tin compounds. In: *Mössbauer Spectroscopy Applied to Inorganic Chemistry*, vol 1, Long, G J (ed), Plenum Press, New York, 1984, p 527
11. Clark, M G, Maddock, A G and Platt, R H *J. Chem. Soc., Dalton Trans.*, 1972: 281
12. Bancroft, G M, Kumar Das, V G, Sham, T K and Clark, M G *J. Chem. Soc., Dalton Trans.*, 1976, 643
13. Barbieri, R, Silvestri, A, Huber, F and Hager, C D *Canad. J. Spectrosc.*, 1981, 26: 194
14. Stocco, G, Guli, G and Valle, G *Acta Cryst.*, 1992, C48: 2116
15. Preut, H, Mundus, B, Huber, F and Barbieri, R *Acta Cryst.*, 1986, C42: 536
16. Huber, F, Haupt, H J, Preut, H, Barbieri, R and Lo Giudice, M T *Z. Anorg. Allg. Chem.*, 1977, 432: 51
17. Mundus-Glowachki, B, Huber, F, Preut, H, Barbieri, R and Ruisi, G *Acta Cryst.*, 1986, C42: 536
18. Mundus-Glowachki, B, Huber, F, Preut, H, Ruisi, G and Barbieri, R *Appl. Organomet. Chem.*, 1992, 6: 83
19. Vornefeld, M, Huber, F, Preut, H, Ruisi, G and Barbieri, R *Appl. Organomet. Chem.*, 1992, 6: 1
20. Preut, H, Vornfeld, M and Huber, F *Acta Crystl., 1991*, C47: 264
21. Rabenstein, D L, Daignault, S A, Isab, A A, Arnold, A P and Shoukry, M M *J. Am. Chem. Soc.*, 1985, 107: 6435
22. Lockart, T P and Manders, W F *Inorg. Chem.*, 1986, 25: 892
23. Ruisi, G, Silvestri, A, Lo Giudice, M T, Barbieri, R, Atassi, G, Huber, F, Kratz, K and Lamartina, L *J. Inorg. Biochem.*, 1985, 25: 229
24. Appleton, T G, Connor, J W and Hall, J R *Inorg. Chem.*, 1988, 27: 130
25. Cimino, G D, Gamper, H B, Isaacs, S T and Hearst, J E *Annu. Rev. Biochem.*, 1985, 54: 1151
26. Malinge, J M, Schwartz, A and Leng, M, *Nucleic Acids Res.*, 1987, 15: 1179