numbering scheme for **2** (14 pages); a table of observed and calculated structure factors (15 pages). Ordering information is given on any current masthead page.

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Spectroscopic Evidence for Stacking and Electrostatic Interactions between Nucleoside 5′-Monophosphates and a Platinum DNA Intercalator, (2,2′-Bipyridine)(ethylenediamine)platinum(II), in Dilute Aqueous Solution

Sir:

Intercalative DNA binding by metal complexes involving planar aromatic rings is now well recognized and receives considerable attention in view of its relevance to the antitumor activity of drugs and usefulness for probing nucleic acid structures.[1] Lippard et al.[1-3] found that the platinum(II) complexes of aromatic amines such as $[Pt(terpy)Cl]^+$, $[Pt(phen)(en)]^{2+}$, and $[Pt(bpy)(en)]^{2+}$ bind to DNA by intercalation, whereas the complex of an aliphatic amine $[Pt(en)_2]^{2+}$ and the nonplanar complex $[Pt(py)_2(en)]^{2+}$ do not.[4] The X-ray diffraction patterns of intercalator–DNA complexes[3,5] and the crystal structure analyses of model intercalative complexes of AMP with $[Pt(terpy)Cl]^+$[6] and double-helical deoxycytidylyl-(3′–5′)-deoxyguanosine with $[Pt(terpy)(SCH_2CH_2OH)]^+$[7] revealed the existence and modes of the intercalative interactions. Intercalative binding is a crucial step for the sequence-specific DNA binding, and a number of investigations have been reported for various metal complex–DNA interactions, such as stereoselective intercalations by chiral complexes,[8] sequence-specific binding and cleavage by Fe(II)–bleomycin,[9] Fe(II)–EDTA linked to DNA-binding distamycin[10] or methidium[11] or *cis*-$[Pt(en)Cl_2]$ linked to acridine orange,[12] and cleavage by the Cu(I)–phenanthroline complex.[13]

Although these investigations reasonably indicate the importance of the stacking between the planar aromatic rings of DNA and the intercalators, information on the interaction with mononucleotides as DNA constituents would offer a further insight into the intercalator–DNA bond, where the base specificity, if

Chart I

$$Pt(bpy)(en) + L \underset{}{\overset{K}{\rightleftharpoons}} Pt(bpy)(en)\cdots L$$

L	AMP	GMP	CMP	UMP
Base	adenine	guanine	cytosine	uracil

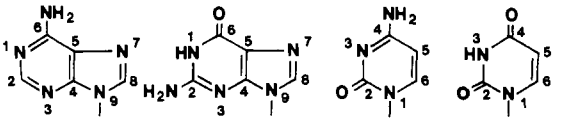

Table I. CD Spectral Data for Pt(bpy)(en)–L Systems in Water at ≥300 nm ([Pt(bpy)(en)] = 0.5 mM)

L	[L]/mM	pH	λ_{max}/nm ($\Delta\epsilon$/M(Pt)$^{-1}$ cm^{-1})
AMP	0.5	7.0	314 (+0.31)
	2.5	7.0	319 (+0.92)
	5.0	2.0	
	5.0	7.0	319 (+1.14)
GMP	0.5	7.0	320 (+0.26)
	2.5	7.0	319 (+0.50)
	5.0	2.0	319 (+0.12)
	5.0	7.0	319 (+0.69)
adenosine	0.5	7.0	
	5.0	7.0	326 (+0.15)
	5.0	10.0	326 (+0.12)

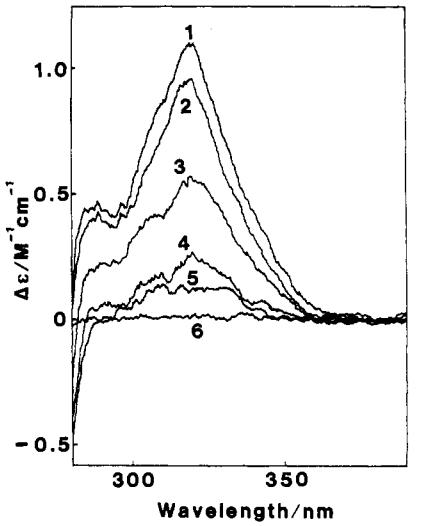

Figure 1. Ionic strength dependence of the CD spectrum due to the Pt(bpy)(en)–AMP interaction. The spectra were measured at room temperature with a JASCO J-500C spectropolarimeter for aqueous solutions (pH 6.8–7.7) containing 0.5 mM Pt(bpy)(en) and 5.0 mM AMP (disodium salt) at the following concentrations (M) of added $NaClO_4$: curve 1, 0; curve 2, 0.01; curve 3, 0.1; curve 4, 0.5; curve 5, 1.0; curve 6, baseline. The $\Delta\epsilon$ values are based on the concentration of Pt(bpy)(en).

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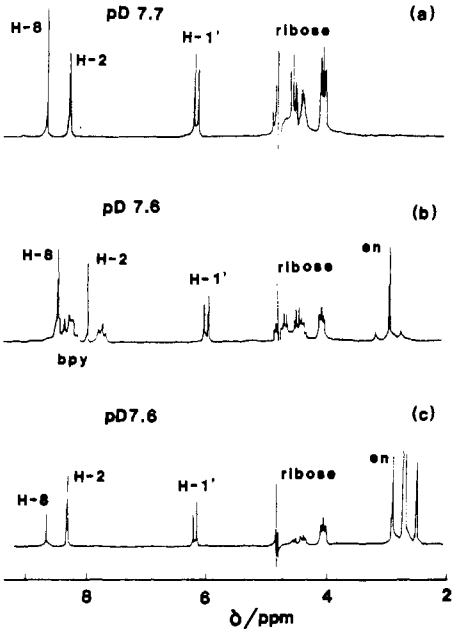

Figure 2. ^{1}H NMR spectra of AMP (a), 1:1 Pt(bpy)(en)–AMP (b), and 1:1 Pt(en)$_2$–AMP (c).[15]

any, at low intercalator:base-pair ratios and individual contributions by the base and phosphate moieties remain uncertain. In the course of studies on noncovalent interactions involving metal complexes, we observed that a DNA intercalator Pt(bpy)(en) (charges are omitted hereafter) effectively binds with uncoordinated nucleoside 5′-monophosphates L (Chart I) in dilute aqueous solution through the bpy–base stacking and Pt(II)–phosphate electrostatic interactions to give an induced circular dichroism (CD) peak and/or cause upfield shifts of the ^{1}H and ^{31}P NMR signals due to the ring current effect[14] of coordinated bpy.

The Pt(bpy)(en)–L systems (L = AMP, GMP, or adenosine) in dilute aqueous solution (Pt(bpy)(en), 0.5 mM) exhibited a positive CD peak at 310–330 nm at neutral pH (Table I). Since none of the nucleotides showed CD peaks in this region under the conditions employed, the result indicates that the D-ribose moiety is located close enough to the coordination plane to induce the CD peak in the bpy absorption band, which undergoes small intensity changes in the presence of L at 300–350 nm. The CD magnitudes increased with the nucleotide:complex ratios (1:1–10:1) and the pH values (2–7) of the solution. Dependence of the magnitudes on the ionic strength of solution (Figure 1) clearly shows that there is a definite electrostatic interaction between the positively charged complex and the negatively charged phosphate group of L. The systems with L = adenosine exhibited the CD peak at a high concentration of L and those with D-ribose phosphate added in place of L were not CD active at ≥300 nm. Under comparable conditions, it was difficult to detect distinct CD bands for the systems with the pyrimidine nucleotides. The aromatic ring stacking is established by the ^{1}H NMR spectra, which revealed that in the presence of Pt(bpy)(en) the purine or pyrimidine proton signals and the ribose proton signal suffer upfield shifts (positive $\Delta\delta$ values) ascribable to the ring current effect[14] of coordinated bpy (Figure 2).[15] The observed upfield shifts are attributed to the stacking interaction, because Pt(en)$_2$

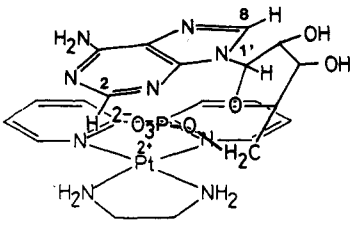

Figure 3. Tentative mode of the Pt(bpy)(en)–AMP interaction in solution.

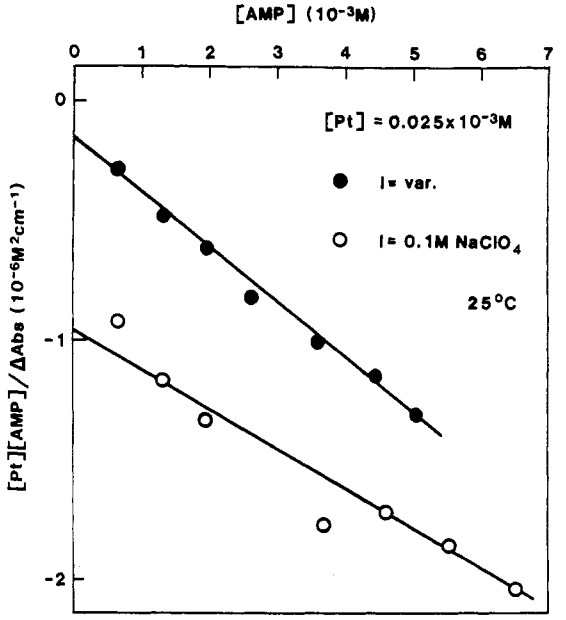

Figure 4. Benesi–Hildebrand plots for the Pt(bpy)(en)–AMP system according to [Pt][AMP]/Δ(abs) = $(1/\epsilon')$ [AMP] + $(1/\epsilon')$ $1/K$, where [Pt] = concentration of Pt(bpy)(en), [AMP] = concentration of monomeric AMP, ϵ' = difference between the molar absorptivity of the Pt(bpy)(en)–AMP complex (ϵ_c) and the sum of the molar absorptivities of Pt(bpy)(en) (ϵ_{Pt}) and AMP (ϵ_{AMP}) = $\epsilon_c - \epsilon_{Pt} - \epsilon_{AMP}$, and Δ(abs) = absorbance of the difference spectrum.

did not affect the signals for AMP in accordance with its inability to intercalate to DNA.[1-3] The H-2 signal of AMP, which suffered the largest shift, suggests that H-2 is located just above the aromatic rings of the complex. In line with the trend observed with the CD spectra, the $\Delta\delta$ values for adenosine and the pyrimidine nucleotides were smaller than those for the purine nucleotides.[15] The ^{31}P NMR signal for 1 M and 20 mM AMP observed at 4.34 ppm relative to the signal of 85% H_3PO_4 shifted upfield by 0.03 ppm in the presence of Pt(bpy)(en) due to the ring current effect,[16] which implies close contact between the Pt(II) center and the phosphate group and supports the CD spectral results indicating the electrostatic interaction between them. Although space-filling molecular models suggest hydrogen bonds between the phosphate oxygens and the amino groups of coordinated en, it may be negligible under the present conditions because no indication of AMP···en association due to the bonding has been given by the ^{1}H NMR spectra in the absence of Pt(II).[17] On the basis of the NMR and CD spectral data and the diagram for long-range shielding by the benzene ring,[14] we may propose a tentative mode of interaction between Pt(bpy)(en) and AMP as shown in Figure 3.

From the preliminary experiments using the difference spectra at 300–350 nm, the stability constant log K for the stacked species

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(15) The ^{1}H NMR spectra were measured with a JEOL FX-100 NMR spectrometer at 100 MHz in FT mode for 20 mM solutions (pD 7.6–8.9) of L, Pt(bpy)(en), Pt(en)$_2$, 1:1 Pt(bpy)(en)–L, and 1:1 Pt(en)$_2$–AMP with sodium 2-(trimethylsilyl)propionate (TSP) as internal standard (accuracy, 0.01 ppm). The $\Delta\delta$ values in ppm for the nucleotides and adenosine are as follows: AMP, 0.29 (H-2), 0.15 (H-8), 0.20 (H-1′); GMP, 0.14 (H-8), 0.16 (H-1′); CMP, 0.18 (H-5), 0.16 (H-6); 0.10 (H-1′); UMP, 0.02 (H-5), 0.05 (H-6), 0.04 (H-1′); adenosine, 0.11 (H-2), 0.03 (H-8), 0.04 (H-1′).

(16) The ^{31}P NMR spectra were obtained with a JEOL FX-200 NMR spectrometer at 80.76 MHz in FT mode (digital resolution, 0.6 Hz) for AMP (1 M and 20 mM solutions) and 1:1 Pt(bpy)(en)–AMP (20 mM) with 85 % H_3PO_4 as external standard.

(17) The ^{1}H NMR chemical shifts (ppm) for 26 mM AMP at pD 7.8 (8.24 (H-2), 8.61 (H-8), 6.13 (H-1′)) and 26 mM en at pD 11.1 (2.69) remained unchanged for 26 mM 1:1 AMP–en at pD 11.0 (8.23 (H-2), 8.61 (H-8), 6.13 (H-1′), 2.70 (en)).

in the Pt(bpy)(en)–AMP system was calculated to be 2.95 ± 0.10 (I varies) and 2.19 ± 0.08 (I = 0.1 M $NaClO_4$) at 25 °C from the Benesi–Hildebrand plots (Figure 4). These values are significantly higher than the values for self-stacking of AMP (0.3)[18] and the bpy–AMP stacking (1.41).[19] The ΔG value for the Pt(bpy)(en)–AMP stacking is calculated to be −16.8 kJ mol^{-1} (I varies), which is in contrast with the small value of −6 kJ mol^{-1} for the stacking between the benzene rings of phenylalanine.[20] Since the CD spectral magnitude decreases with the increase of the ionic strength (Figure 1), the observed stability difference due to the ionic strength may be indicative of the contribution of the Pt(II)···phosphate interaction.

In conclusion, the Pt(II) complex–nucleotide bonding involves both stacking and the electrostatic interaction, and the latter serves to orient the ribose group near the Pt(II) center. Further studies on the modes and stabilities of the interactions are in progress.

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Registry No. Pt(bpy)(en)$^{2+}$, 24972-61-0; AMP, 61-19-8; GMP, 58-61-7; adenosine, 58-61-7; CMP, 63-37-6; UMP, 58-97-9.

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Articles

Contribution from the Departament de Quimica Inorgànica, Universitat Autònoma de Barcelona, Bellaterra, Barcelona, Spain, and Department of Chemistry, The University of Michigan, Ann Arbor, Michigan 48109

Rules for Predicting the ^{11}B NMR Spectra of *closo*-Boranes and *closo*-Heteroboranes

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The ^{11}B NMR pattern of *closo*-boranes and *closo*-heteroboranes can be predicted by utilizing five parameters, which we term antipodal, rhomboidal, butterfly, neighbor, and symmetrical neighbor effects. The application of the rules depends on the symmetry of the parent $B_nH_n^{2-}$ borane, and the magnitudes of the various shifts are functions of the coordination number and framework electron contribution of the atoms that cause them. In substituted B–R *closo*-boranes and -heteroboranes, the magnitude of the effects depends primarily on the electronegativity of the R substituent.

Introduction

Progress in the general area of boron chemistry has depended to a very large extent on information derived from ^{11}B NMR spectroscopy. Not only is it possible to determine if new species have been formed by comparison of observed chemical shifts with data in the literature[1,2] but the boron NMR pattern displayed reveals, in many cases, important information about the structure. However, distinctions between possible structures on the basis of NMR results so far have depended to a very great extent on symmetry considerations.

Inasmuch as boron chemical shifts are dominated by the contribution of the paramagnetic term (σ_p),[3,4] assignment of absorptions in the ^{11}B NMR spectrum to specific borons in the molecule is a difficult process. Theoretical predictions require extensive calculations involving a number of approximations. Chemical assignment by deuteration, which to date has been the most reliable method, is time consuming and not always straightforward. Recently, the use of two dimensional (2D) ^{11}B–^{11}B (COSY) NMR has been shown to be helpful in favorable cases;[5] still, the chemical shift position of some groups of atoms have to be known a priori. Consequently, the availability of rules, even though they may be strictly empirical, that can relate the position of a peak in the ^{11}B NMR spectrum to a particular boron in a cluster would be very valuable. Some regularities in NMR spectra that depend on the antipodal effect,[6] the coordination number,[7] and the position of the hydrogen bridges have already been noted.[8]

The paramagnetic term (σ_p) of the nucleus of interest depends on the molecular orbitals, which in turn are dependent on the symmetry of the molecule. This leads us to believe that by symmetry considerations alone, it should be possible to predict for a specific cluster not only the ^{11}B NMR pattern but also the order of its components.[9] In general, the geometry of a closo compound can be predicted once the number of atoms in the cluster is known;[10] however, this is not necessarily true for nido, arachno, and hypho species since they have "extra" hydrogens to be located.[8] For that reason, closo species constitute the most favorable of the four types of clusters that could be used to demonstrate our approach. In this paper, we present some effects we have observed in the closo clusters, which we believe will be valuable in assigning

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