Inorganica Chimica Acta, 91 (1984) 129–135

The Interaction of Aquated Platinum(II) Compounds with Purine Mononucleotides

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Received June 1, 1983

The interactions of aquated bifunctional cis-*diammineplatinum(II) and 2,2,N,N-tetramethylpropanediamineplatinum(II) and monofunctional diethylenetriamineplatinum(II) with 5'-GMP, 5'-IMP and 3'-GMP were investigated by* ^{1}*H-NMR. Reaction of these aquated platinum compounds with a stoichiometric amount of the mononucleotide yields 1:1 Pt–nucleotide complexes as reaction products. The bifunctional platinum compounds containing one mononucleotide may react further with additional mononucleotides or with chloride to form 1:2 Pt–nucleotide complexes, or 1:1 Pt–nucleotide complexes in which a chloride is coordinated to platinum. Competition binding experiments show that both aquated* cis-*diammineplatinum and diethylenetriamineplatinum(II) bind preferentially to 5'-GMP when allowed to react with a mixture of 5'-GMP and 3'-GMP.*

Introduction

The interaction of *cis*-platinum compounds with DNA appears to be essential for their activity as antitumor agents [1, 2]. The primary targets for attack by *cis*-$Pt(NH_3)_2Cl_2$(*cis*-A_2PtCl_2) are most likely the N7 atoms of the guanine base [3, 4]. A specific bifunctional binding only possible for *cis*-A_2PtCl_2 could explain its activity in comparison with inactive platinum compounds like *trans*-A_2PtCl_2 and the monofunctional compound (dienPtCl)Cl, which are also able to bind to DNA, both *in vitro* [5] and *in vivo* [6, 7]. As a specific binding mode for *cis*-A_2PtCl_2 intrastrand crosslinks between two bases in DNA *via cis*-A_2PtCl_2 have been suggested [8, 9] and there is evidence that such crosslinks occur in oligonucleotides [10–16] and DNA's with defined sequences treated with *cis*-A_2PtCl_2 *in vitro* [17, 18].

Alternatively, chelate formation by binding of *cis*-A_2PtCl_2 to the N7 and O6 of a single guanine base has been proposed as the cytotoxic lesion [19]. A direct Pt–O6 interaction is not very likely, and has been rejected for geometric reasons [20]. Hydrogen-bonding interactions between O6 and H_2O or NH_3 ligand of platinum bound to N7 may, however, occur [21, 22]. Such interactions may also occur for *trans*-A_2Pt and dienPt bound to guanine N7.

The binding of a bifunctional platinum compound to DNA probably occurs in two stages. After an initial monofunctional binding, which may result in a local structural perturbation of the DNA, the second, crosslinking, reaction can take place. The study of the first interaction of platinum compounds with nucleotides, *i.e.* the formation of 1:1 Pt–nucleotide compounds has so far received little attention.

In an earlier study [23] we showed that *cis*-A_2Pt reacts faster with 5'-GMP or 5'-dGMP than does *trans*-A_2Pt (GMP = guanosine monophosphate). Reaction of the first 5'-GMP with *cis*-A_2Pt is fast and yields the 1:1 product *cis*-A_2Pt(5'-GMP)(H_2O).

In this study the interaction of several platinum compounds (see Fig. 1) with the mononucleotides 5'-GMP, 5'-IMP (IMP = inosine monophosphate; see Fig. 2) and 3'-GMP was followed by ^{1}H NMR to obtain information about the formation of 1:1 and 1:2 Pt–nucleotide compounds and their properties. Some competition experiments to study the relative binding preferences of platinum compounds and mononucleotides have also been performed, in order to determine the factors that are important for the binding of platinum compounds to mononucleotides as models for platinum binding to DNA.

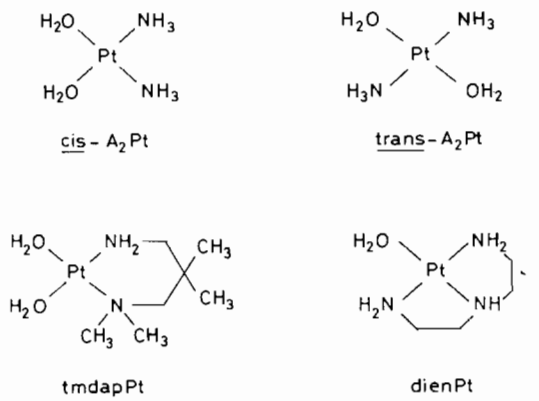

Fig. 1. Schematic structure of the aquated forms of the platinum compounds used in this study. CH_2 protons have been omitted for clarity.

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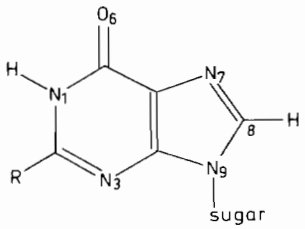

Fig. 2. Purine ring numbering system; R = NH_2(GMP) or R = H(IMP).

Experimental

The mononucleotides used were commercially available as their disodium salts. *Cis*-A_2PtX_2, tmdapPtX_2 and (dienPtX)X (X = I) were prepared by published methods [24–26] (tmdap = 2,2,N-tetramethyl-1,3-propanediamine, dien = diethylenetriamine). Solutions of the aquated platinum compounds were prepared from the iodides by stirring suspensions of these compounds with 1.95 equivalents of $AgNO_3$ in D_2O for one hour and filtering off the AgI precipitate.

Aquated *cis*-A_2Pt may be represented as *cis*-$A_2Pt(H_2O)_2^{2+}$. At neutral pH this compound is in equilibrium with the species *cis*-$A_2Pt(H_2O)(OH)^+$ and *cis*-$A_2Pt(OH)_2$ [19]. The pK_a values are reported to be 5.6 and 7.3, so the species dominating at neutral pH is *cis*-$A_2Pt(H_2O)(OH)^+$. Under neutral conditions formation of non-reactive hydroxy-bridged dimers may occur, especially in concentrated solutions [19, 27]. Therefore, stock solutions of the aquated *cis*-platinum compounds were kept unbuffered (pH 3–4) and were used within 24 hours after preparation.

Due to the good solubility of the aquated platinum compounds in water, experiments with these compounds were usually performed in 5 mm diameter NMR tubes at a concentration of 20 mM for the nucleotide. ^1H NMR spectra were recorded on a JEOL PS-100 spectrometer, operating in the Fourier-transform mode. The solutions were not buffered. After the reaction had taken place, the pH* was usually between 6 and 7. No essential differences were found between reactions performed in buffered and unbuffered solutions, although the reactions in buffered solutions appeared to be slightly slower [28]. As internal chemical shift reference tetramethylammoniumnitrate (TMA) was used. The reported chemical shifts are referenced on the TSP scale (TMA: 3.18 ppm downfield from TSP; TSP = sodium 2,2,3,3-tetradeutero-3(trimethylsilyl)propionic acid). No attempt was made to isolate the platinum compounds as crystalline solids.

Results

General

Under the conditions used, platinum coordination at the N7 atoms of the guanine bases is expected [29–31]. This is confirmed by the downfield shifts observed for the H8 resonances [4, 10–15, 22, 32, 33]. Unfortunately, the satellites due to coupling of these protons with the ^{195}Pt nucleus were not clearly observed, for reasons described by Chottard *et al.* [9, 34]. With 5′-IMP as nucleotide binding at N7 is also expected [21, 35, 36]. However, in some experiments small amounts of impurities were observed; probably these were compounds in which the N1 atom of the ligand is bound to platinum.

The chemical shifts, especially those of the H8 resonances, are slightly temperature dependent. They decrease with an increasing temperature, as also found by other workers [37]. A concentration dependence is also likely. Furthermore, because spectra were obtained from approximately neutral solutions, slight variations in the pH may give small differences in the chemical shift values, due to the effect of the phosphate protonation (pK_a about 6.0) [33]. Therefore, the accuracy of the chemical shifts under the indicated conditions is estimated to be only about 0.02 ppm. The charge of the compounds that were obtained is not indicated. At the pH used, most of the phosphate groups will be double deprotonated, and the bases are, therefore, expected to be uncharged. ^1H NMR chemical shifts of the base protons and H1′ protons of the platinum compounds are listed in Table I.

Formation of Reaction Products

Reaction of the aquated platinum compounds *cis*-A_2Pt and tmdapPt with one equivalent of 5′-GMP is very fast, even at 5 °C. This reaction is much faster than the reaction of aquated *trans*-A_2Pt with 5′-GMP [23]. Within half an hour the 1:1 products *cis*-A_2Pt(5′-GMP)(H_2O) and tmdapPt(5′-GMP)(H_2O) are formed at 5 °C. The presence of a 1:1 product is inferred from the fact that subsequent addition of a second equivalent of 5′-GMP leads to formation of the bis(nucleotide)-platinum compounds. This second reaction, however, is much slower. Binding of the first 5′-GMP to tmdapPt gives a product with only one H8 resonance. This is most likely due to binding of the first 5′-GMP to the least sterically hindered site of the tmdapPt molecule, *i.e.* close to the NH_2 group. Binding of the second molecule of 5′-GMP to tmdapPt(5′-GMP)(H_2O) is very slow, probably due to steric hindrance by the $N(CH_3)_2$ group of the tmdap ligand. Hydrogen bonding between coordinated H_2O and the O6 atom of GMP could also stabilize the 1:1 adduct, however. In the compound tmdapPt(5′-GMP)$_2$ four sets of resonances should be expected for the 5′-GMP ligands [38]. In fact, only

TABLE I. ^{1}H NMR Chemical Shifts of the Base and H1′ Protons of Some Nucleotides and of their Platinum–Nucleotide Compounds[a]. First Order Coupling Constants $J_{H1'-H2'}$ are Given in Parentheses.

Compound	L = 5′-GMP		L = 5′-IMP			L = 3′-GMP		Temp.
	H8	H1′	H8	H2	H1′	H8	H1′	
L	8.22	5.92 (6.2)	8.55	8.20	6.14 (5.9)	8.01	5.93 (5.6)	5 °C
cis-A_2PtL_2	8.70	5.85 (3.2)	9.13	8.14	6.10 (3.9)	8.48	5.90 (2.5)	20 °C
cis-A_2PtLCl	8.62	5.99 (5.1)	9.02	8.28	6.20 (4.6)			20 °C
cis-$A_2PtL(H_2O)$	8.89	6.02 (1.7)	9.25	8.30	6.25 (2.2)			5 °C
cis-A_2PtLL'	8.75	5.84 (3.4)	9.07	8.12	6.09 (2.7)			22 °C
tmdapPtL$_2$	8.53 8.37 8.36	5.83						20 °C
tmdapPtLCl	8.49	5.96 (5.1)	8.88	8.29	6.18 (4.6)			20 °C
tmdapPtL(H_2O)	8.85	6.04 (2.7)	9.18	8.31	6.26 (3.2)			5 °C
dienPtL	8.86	6.00 (3.9)				8.47	5.96 (3.4)	5 °C

[a]Chemical shifts in ppm relative to TSP; coupling constants in Hertz. Spectra were obtained from D_2O solutions at neutral pH (6.0–7.0); the temperature is indicated in the Table.

three resonances are observed, probably because two of the four resonances merge. The same reactions were also observed for 5′-IMP. As an example the reactions of 5′-IMP with *cis*-A_2Pt are schematically depicted in Fig. 3.

Reaction of 3′-GMP with one equivalent of *cis*-A_2-Pt leads to rapid formation of an insoluble white precipitate in the NMR tube, which could not be studied by NMR. This product, however, reacts further with additional 3′-GMP to yield the soluble product *cis*-$A_2Pt(3'\text{-GMP})_2$ (see Table I).

Reaction of aquated *cis*-platinum compounds with 5′-GMP in a 1:1 ratio yields almost pure *cis*-Pt(5′-GMP)(H_2O) compounds. These products react further with added chloride. Reaction of these compounds with a five-fold excess of NaCl takes several hours at room temperature to produce the compounds *cis*-A_2-Pt(5′-GMP)Cl or tmdapPt(5′-GMP)Cl. Compounds with 5′-IMP react similarly with excess chloride.

The availability of stable *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$ allows the synthesis of mixed platinum-nucleotide compounds. Reaction of *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$ with one equivalent of 5′-IMP yields the mixed compound *cis*-$A_2Pt(5'\text{-GMP})(5'\text{-IMP})$. A synthesis starting with *cis*-$A_2Pt(5'\text{-IMP})(H_2O)$ yields the same compound upon reaction with 5′-GMP. The chemical shifts of this mixed compound are slightly different from those of *cis*-$A_2Pt(5'\text{-GMP})_2$ and *cis*-$A_2Pt(5'\text{-IMP})_2$, as is seen from NMR spectra of mixtures of these compounds (see Table I). Reaction between aquated dien-Pt and 5′-GMP or 3′-GMP yields dienPt(5′-GMP) or dienPt(3′-GMP) within half an hour at 5 °C (see Table I).

Competition Experiments

Competition binding experiments of platinum compounds with mixtures of 5′-nucleoside monophosphates have already been performed [4]. From these experiments it became clear that *cis*-A_2Pt has a strong preference for binding to the guanine N7. In these experiments, however, no distinction was made between binding of the first and of the second nucleotide to platinum.

To a mixture of equal amounts of 5′-GMP, 5′-AMP and 5′-CMP one equivalent of aquated *cis*-A_2Pt was added at 5 °C (Fig. 4). After 15 minutes almost all 5′-GMP had reacted to form *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$. Signals originating from other reaction products were very small. Only after prolonged reaction times other resonances, due to formation of mixed *cis*-$A_2Pt(5'\text{-GMP})(5'\text{-AMP})$ compounds appeared, while the resonances of free 5′-AMP simultaneously decreased in intensity (Fig. 4).

Addition of one equivalent of aquated *cis*-A_2Pt to a mixture of equal amounts of 5′-GMP and 5′-dGMP showed that the platinum compound has no preference for one of these nucleotides.

Although reaction of aquated *cis*-A_2Pt with 3′-GMP results in a precipitate, the reaction of *cis*-A_2-Pt with a 1:1 mixture of 5′-GMP and 3′-GMP could still be studied in solution. Upon addition of one equivalent of the platinum compound some precipitation occurred in the NMR tube. The NMR spectrum showed that 5′-GMP had reacted to *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$, while a strong H8 signal due to unreacted 3′-GMP was still present (Fig. 5). The same competition experiment between 5-GMP and 3′-GMP was also performed with aquated dienPt. In this case both reaction products are soluble. 15 minutes after addition of one equivalent of dienPt at 5 °C most of the 5′-GMP had reacted to dienPt(5′-GMP), while only a small portion of the 3′-GMP had reacted (Fig. 5).

To determine whether *cis*-A_2Pt or dienPt reacts faster with 5′-GMP, a 1:1 mixture of these two

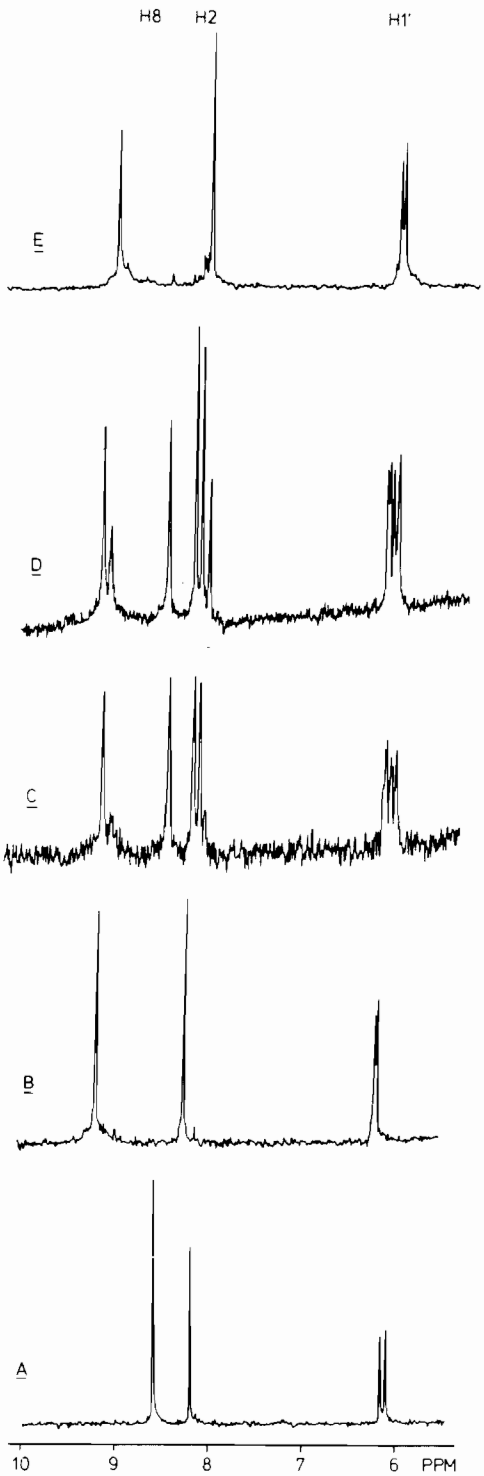

Fig. 3. Reaction of aquated *cis*-A_2Pt with 5'-IMP, followed by ^1H NMR (100 MHz), showing the H8, H2 and H1' resonances of 5'-IMP. A) 5'-IMP (20 mM, 5 °C). B) 15 minutes after addition of one equivalent of *cis*-A_2Pt at 5 °C. C) Immediately after addition of a second equivalent of 5'-IMP to the compound obtained in B. D) After one hour reaction at 5 °C. E) Product obtained after reaction for several hours at room temperature.

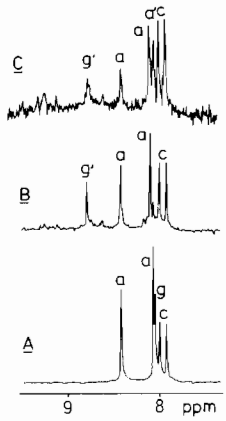

Fig. 4. Reaction of *cis*-A_2Pt with a 1:1:1 mixture of 5'-AMP, 5'-CMP and 5'-GMP, followed by ^1H NMR, showing the resonances of the base protons. A) Mixture of the nucleotides at 5 °C. B) 15 minutes after addition of one equivalent of *cis*-A_2Pt at 5 °C. C) After two hours reaction at room temperature. Resonances denoted a are from 5'-AMP, c from 5'-CMP and g from 5'-GMP. g' is from *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$ and a' is from H2 resonances of 5'-AMP coordinated to Pt. The other small peaks in spectrum C are due to reaction products containing both 5'-AMP and 5'-GMP.

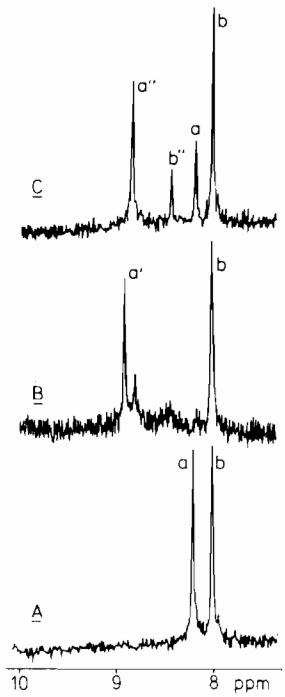

Fig. 5. Reaction of platinum compounds with mixtures of 3'-GMP and 5'-GMP at 5 °C. Only the H8 resonances are shown. The concentration of all reactants was 20 m*M*. A) Mixture of 5'-GMP (a) and 3'-GMP (b). B) 15 minutes after addition of one equivalent of *cis*-A_2Pt. The resonance denoted a' is from *cis*-$A_2Pt(5'\text{-GMP})(H_2O)$. The small peak at 8.80 ppm might be from *cis*-$A_2Pt(3'\text{-GMP})(H_2O)$. C) 15 minutes after addition of one equivalent of dienPt to the mixture of 5'-GMP and 3'-GMP. The resonance denoted a'' originates from dienPt(5'-GMP) and the one denoted b'' originates from dienPt(3'-GMP).

platinum compounds was allowed to compete for one equivalent of 5′-GMP. Both reaction products, *cis*-A_2Pt(5′-GMP)(H_2O) and dienPt(5′-GMP) appear at about an equal rate, indicating that there is hardly any preference for one of these platinum compounds.

Discussion

The reaction of aquated *trans*-A_2Pt with 5′-GMP is much slower than the reaction of aquated *cis*-A_2Pt or dienPt [23]. This difference in reaction rate can be ascribed to the *trans* effect [39]. According to the rules of the *trans* effect ligands that are *trans* with respect to an NH_3 or NR_3 ligand are more easily substituted than ligands *trans* with respect to a H_2O ligand. As is apparent from competition experiments, the reactions of *cis*-A_2Pt or dienPt with 5′-GMP are about equally fast. This is also in accordance with the rules of the *trans* effect [39].

Because the reaction of a second nucleotide with aquated *cis*-A_2Pt is much slower than the reaction of the first nucleotide, the 1:1 Pt–nucleotides can be obtained in a rather pure state in solution (see Fig. 3). These compounds may then be reacted further with for example the same, or another nucleotide, or with excess chloride. Reaction with another nucleotide may yield a platinum compound with two different nucleotides, as demonstrated for *cis*-A_2Pt(5′-GMP)(5′-IMP). Synthesis of this compound also shows that under the conditions used, no ligand exchange between the nitrogen-donors occurs in the platinum compounds.

Reaction of *cis*-A_2Pt(5′-GMP)(H_2O) or *cis*-A_2Pt-(5′-IMP)(H_2O) with an excess of chloride leads to formation of almost pure *cis*-A_2Pt(5′-GMP)Cl (and *cis*-A_2Pt(5′-IMP)Cl) with a single sharp H8 resonance. In a mixture of compounds, prepared upon reaction of 5′-GMP with a tenfold excess of *cis*-A_2PtCl_2 in a very large excess of chloride, Clore and Gronenborn [40] assigned two of the observed H8 resonances to rotamers of *cis*-A_2Pt(5′-GMP)Cl. However, under our conditions only one sharp H8 resonance is observed for *cis*-A_2Pt(5′-GMP)Cl. Furthermore, because we recently showed that rotation of purines in Pt–purine compounds is fast on the NMR time scale when unsubstituted amines are coordinated to platinum [41], we feel confident that only one H8 resonance should be observed for *cis*-A_2Pt(5′-GMP)Cl. This reasoning is confirmed by preliminary experiments, using the conditions described by Clore and Gronenborn [40], that suggest the other product to be *cis*-A_2Pt(5′-GMP)$_2$ instead of a rotamer of *cis*-A_2Pt(5′-GMP)Cl.

Mansy *et al.* have shown that *cis*-A_2Pt has a preference for binding to 5′-GMP when it is allowed to react with a mixture of 5′-nucleoside monophosphates [4]. Binding to 5′-AMP was also observed, but to a smaller extent. Our results show that, as a first reaction product, *cis*-A_2Pt(5′-GMP)(H_2O) is formed almost exclusively. The second binding reaction with 5′-AMP is much slower (Fig. 4). The preference for 5′-GMP over 5′-AMP may be due to the fact that hydrogen bonding of coordinated NH_3 or H_2O to the O6 of 5′-GMP is possible, whereas no hydrogen bonding is possible from the N7 bound platinum adduct to the C6-NH_2 group of 5′-AMP. Such hydrogen bonding interactions can be important for binding at N7, as was shown for a variety of Co-complexes by Marzilli *et al.* [42].

As shown in the competition experiments of aquated *cis*-A_2Pt or dienPt with mixtures of 5′-GMP and 3′-GMP (Fig. 5) the platinum compounds have a distinct kinetic preference for the nucleotides containing a 5′-phosphate group. Several observations in the literature indicate that the assistance of the 5′-phosphate group is important in the reaction rate of the binding of platinum compounds to the N7 site of purines [11, 43]. An enhancement of the N7/N1 binding ratio in the equilibrium binding of palladium compounds with adenosine or inosine nucleotides containing a 5′-phosphate group was observed by Vestues *et al.* [44]. A first interaction with a 5′-phosphate group (probably of ionic or dipolar nature) could bring the platinum compound near the N7 position of a purine. On the other hand, when the platinum compound is near the 3′-phosphate group by such ionic or dipolar interactions, a simultaneous interaction with the purine N7 is unlikely.

The H1′ sugar resonances of all platinum–nucleotide compounds, except the *cis*-bis(nucleotide)platinum compounds are shifted to lower field as compared with the same resonances in the free nucleotides. For the *cis*-bis(nucleotide)platinum compounds small upfield shifts are found. These upfield shifts may be due to a mutual influence of the two nucleotides on each other in the complex, *e.g.* a shielding effect [22, 23, 31, 45]. Such an influence is highly likely, since it has been shown in several crystal structures of *cis*-bis(nucleotide)platinum compounds that the angles between the planes through the nucleotide ligands can be significantly smaller than 90 °C [21, 31, 35, 36]. This shielding effect could also be the origin of the smaller chemical shifts of the H8 resonances in *cis*-bis(nucleotide) compounds than in for example the corresponding *trans*-bis(nucleotide) compounds [23].

Compared with the free nucleotides the $J_{H1'-H2'}$ coupling constants in the N7 platinated compounds are relatively small (see Table I). This decrease of the coupling constants is ascribed to a change of the conformational equilibrium in the sugar moiety in the direction of the N-type conformer [46]. This conformational equilibrium may be effected by the temperature, as is seen in the compound *cis*-A_2Pt(5′-GMP)$_2$ where the $J_{H1'-H2'}$ coupling constant decreases from 5.0 Hz at 70 °C to 2.5 Hz at 5 °C [23].

Dipolar interaction between the platinum adduct and the 5′-phosphate group, or hydrogen bond interactions, as proposed by Vestues *et al.* [44], and as found in a *trans*-A_2Pt(transfer-RNA) crystal structure [47] may in part account for the observed conformational changes. Such interactions seem unlikely in compounds containing 3′-GMP. Yet, the coupling constants for the compounds containing 3′-GMP are also relatively small (see Table I). It is of interest to note that in the crystal structure of the compound *cis*-$A_2Pt(3'\text{-CMP})_2$ the riboses are also found to adopt an N-type conformation [48].

Conclusions

As the physiologically-active form of the antitumor drug *cis*-A_2PtCl_2 is probably a hydrolyzed form of this compound [49], the reactions of the aquated forms of the platinum compound are of great biological interest.

The first interaction of aquated *cis*-A_2Pt occurs very fast and very specifically with 5′-GMP and other 5′-(6-oxopurine nucleoside) monophosphates, whereas reaction with the aquated *trans*-A_2Pt compound is much slower [23]. Important factors for binding to nucleotides, and possibly also to DNA, appear to be:

1) the substitution of a coordinated water molecule from platinum, as mainly governed by the *trans* effect,

2) the presence of a 6-oxo group on the purine, to which hydrogen bonding from coordinated NH_3 or H_2O ligands of platinum can occur, and

3) the presence of a 5′-phosphate group, which may help to bring the platinum compound and the binding site together, probably through dipolar interaction and/or hydrogen bonds. In DNA, such interactions do not lead to a preference for a particular base, but it could facilitate reaction with the bases of DNA in general.

In the so-formed 1:1 Pt–nucleotide compounds hydrogen bonds between the platinum adduct and the O6 and/or 5′-phosphate may still be present and be responsible for the relative stability of *cis*-$A_2Pt(H_2O)$(5′-GMP). A further reaction with a nucleic acid base is now possible, which after some reorganization and/or rotation could lead to inter- or intrastrand crosslinks in DNA.

Acknowledgements

This research was supported by the Netherlands Foundation for Chemical Research (SON) with financial aid from the Netherlands Organization for the Advancement of Pure Research (ZWO).

References

1 J. J. Roberts, in G. L. Eichhorn and L. G. Marzilli (Eds.) 'Metals ions in genetic information transfer'. Elsevier/North-Holland, New York (1981) p. 273.
2 A. T. M. Marcelis and J. Reedijk, *Recl. Trav. Chim. Pays Bas, 102*, 121 (1983).
3 L. L. Munchausen and R. O. Rahn, *Biochim. Biophys. Acta, 414*, 242 (1975).
4 S. Mansy, G. Y. H. Chu, R. E. Duncan and R. S. Tobias, *J. Am. Chem. Soc., 100*, 607 (1978).
5 J. P. Macquet and J. L. Butour, *Biochimie, 60*, 901 (1978).
6 L. A. Zwelling, T. Anderson and K. W. Kohn, *Cancer Res., 39*, 365 (1979).
7 P. Lecointe, J. P. Macquet and J. L. Butour, *Biochem. Biophys. Res. Commun., 90*, 209 (1979).
8 P. J. Stone, A. D. Kelman and F. M. Sinex, *Nature, 251*, 737 (1974).
9 G. L. Cohen, J. A. Ledner, W. R. Bauer, H. M. Ushay, C. Caravana and S. J. Lippard, *J. Am. Chem. Soc., 102*, 2487 (1980).
10 J. C. Chottard, J. P. Girault, G. Chottard, J. Y. Lallemand and D. Mansuy, *J. Am. Chem. Soc., 102*, 5565 (1980).
11 J. P. Girault, G. Chottard, J. Y. Lallemand and J. C. Chottard, *Biochemistry, 21*, 1352 (1982).
12 A. T. M. Marcelis, G. W. Canters and J. Reedijk, *Recl. Trav. Chim. Pays-Bas, 100*, 391 (1981).
13 A. T. M. Marcelis, J. H. J. den Hartog and J. Reedijk, *J. Am. Chem. Soc., 104*, 2664 (1982).
14 A. M. J. Fichtinger-Schepman, P. H. M. Lohman and J. Reedijk, *Nucl. Acids Res., 10*, 5345 (1982).
15 J. P. Girault, J. C. Chottard, E. R. Guittet, J. Y. Lallemand, T. Huynh-Dinh and J. Igolen, *Biochem. Biophys. Res. Commun., 109*, 1157 (1982).
16 J. P. Caradonna, S. J. Lippard, M. J. Gait and M. Singh, *J. Am. Chem. Soc., 104*, 5793 (1982).
17 T. D. Tullius and S. J. Lippard, *J. Am. Chem. Soc., 103*, 4620 (1981).
18 B. Royer-Pokora, L. K. Gordon and W. A. Haseltine, *Nucleic Acids. Res., 9*, 4595 (1981).
19 B. Rosenberg, *Biochimie, 60*, 859 (1978).
20 L. G. Marzilli, in G. L. Eichhorn and L. G. Marzilli (Eds.) 'Metal ions in genetic information transfer'. Elsevier/North-Holland, New York (1981) p. 47 and references cited therein.
21 D. M. L. Goddgame, I. Jeeves, F. L. Philips and A. C. Skapski, *Biochim. Biophys. Acta, 378*, 153 (1975).
22 G. Y. H. Chu, S. Mansy, R. E. Duncan and R. S. Tobias, *J. Am. Chem. Soc., 100*, 593 (1978).
23 A. T. M. Marcelis, C. G. van Kralingen and J. Reedijk, *J. Inorg. Biochem., 13*, 213 (1980).
24 S. C. Dhara, *Indian. J. Chem., 8*, 193 (1970).
25 G. W. Watt and W. A. Cude, *Inorg. Chem., 7*, 335 (1968).
26 C. G. van Kralingen, *Ph.D. Thesis*, Delft University of Technology (1979).
27 R. Faggiani, B. Lippert, C. J. L. Lock and B. Rosenberg, *J. Am. Chem. Soc., 99*, 777 (1977).
28 N. P. Johnson, J. D. Hoeschele and R. O. Rahn, *Chem. Biol. Interactions, 30*, 151 (1980).
29 R. W. Gellert and R. Bau, *J. Am. Chem. Soc., 97*, 7379 (1975).
30 R. E. Cramer, P. L. Dahlstrom, M. J. T. Seu, T. Norton and M. Kashiwagi, *Inorg. Chem., 19*, 148 (1980).
31 L. G. Marzilli, P. Chalilpoyil, C. C. Chiang and T. J. Kistenmacher, *J. Am. Chem. Soc., 102*, 2480 (1980).
32 P. C. Kong and T. Theophanides, *Bioinorg. Chem., 5*, 51 (1975).
33 K. H. Scheller, V. Scheller-Krattinger and R. B. Martin,

J. Am. Chem. Soc., 103, 6833 (1981).
34 J. Y. Lallemand, J. Soullié and J. C. Chottard, *J. Chem. Soc. Chem. Commun.*, 436 (1980).
35 T. J. Kistenmacher, C. C. Chiang, P. Chalilpoyil and L. G. Marzilli, *J. Am. Chem. Soc., 101*, 1143 (1979).
36 T. J. Kistenmacher, C. C. Chiang, P. Chalilpoyil and L. G. Marzilli, *Biochem. Biophys. Res. Commun., 84*, 70 (1978).
37 R. E. Cramer and P. L. Dahlstrom, *J. Am. Chem. Soc., 101*, 3679 (1979).
38 A. T. M. Marcelis, J. L. van der Veer, J. C. M. Zwetsloot and J. Reedijk, *Inorg. Chim. Acta, 78*, 195 (1983).
39 F. Basolo and R. G. Pearson, *Progr. Inorg. Chem., 4*, 381 (1962).
40 G. M. Clore and A. M. Gronenborn, *J. Am. Chem. Soc., 104*, 1369 (1982).
41 A. T. M. Marcelis, H. J. Korte, B. Krebs and J. Reedijk, *Inorg. Chem., 21*, 4059 (1982).
42 L. G. Marzilli and T. J. Kistenmacher, *Acc. Chem. Res., 10*, 146 (1977).
43 A. B. Robins, *Chem.-Biol. Interactions, 6*, 35 (1973).
44 P. I. Vestues and R. B. Martin, *J. Am. Chem. Soc., 103*, 806 (1981).
45 G. Y. H. Chu and R. S. Tobias, *J. Am. Chem. Soc., 98*, 2641 (1976).
46 C. Altona and M. Sundaralingam, *J. Am. Chem. Soc., 95*, 2333 (1973).
47 A. Jack, J. E. Ladner, D. Rhodes, R. S. Brown and A. Klug, *J. Mol. Biol., 111*, 315 (1977).
48 S. M. Wu and R. Bau, *Biochem. Biophys. Res. Commun., 88*, 1435 (1979).
49 A. F. Leroy, *Cancer Treatment Rep., 63*, 231 (1979).