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Exocyclic oxygen atoms of platinated nucleobases as binding sites for alkali metal ions †

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Three complexes of model nucleobases with exocyclic oxygen atoms (1-methyluracilate, mura; 1-methylcytosine, mcyt; 9-methylguanine, Hmgua) which contain Pt^{II} bonded to a ring N atom and an alkali metal ion (Cs^+, K^+, Na^+) bonded to a keto oxygen of the bases, *trans*-$Cs[Pt(NH_3)(mura)I_2]\cdot 4H_2O$ **1**, *trans*-$K[Pt(NH_3)_2(mcyt)_2][PF_6]_3\cdot H_2O$ **2**, and *trans*-$[Pt(NH_3)(Hmgua)_2(mcyt)Na(H_2O)_2][ClO_4]_3\cdot 0.5H_2O$ **3**, have been prepared and their crystal structures determined. The compounds have been studied, among others, with regard to the role of alkali metal ions for the rotation of nucleobases when bound to Pt^{II}. While in the case of **1** the alkali metal ion is necessary for charge compensation and for this reason its binding to the platinated mura is not fully unexpected, it is surprising to see that alkali metal ions even bind to cationic complexes of Pt^{II} containing neutral nucleobases (**2**, **3**).

Introduction

Alkali metal ions represent the natural counter ions of the polyanionic nucleic acids. Apart from non-specific charge screening, also the possibility of distinct complexation of alkali metal ions by O and N donor atoms of the nucleobases exists.[1–4] Intrusion of alkali metal ions into electrostatic pockets of the minor groove of DNA and hence the spine of hydration is now generally accepted. Moreover, the role of these metal ions in stabilizing special nucleic acid structures (guanine quartets in telomeres,[5] uracil and thymine quartets,[6,7] quintet formation with isoguanine bases[8]) is well established. In the major groove of DNA the intrastrand GpG step is the most favorable electrostatic pocket for metal ion binding.[1] With RNA the complicated folding generates many more such sites.[9]

In previous reports others and ourselves have elaborated structural aspects of alkali metal ion containing quartets derived from the pyrimidine bases uracil (U)[10] and thymine (T).[11,12] Here we address the question if and how attachment of a heavy metal ion such as Pt^{II} to DNA bases possibly affects binding of the "natural" counter ions of DNA, specifically that of alkali metal ions. This aspect has not attracted much attention in the past.[13] Despite the availability of several crystal structure analyses of DNA fragments (intrastrand[14,15] and interstrand[16] GpG adducts of cisplatin, $[Pt(NH_3)_2Cl_2]$), nothing is known about the possible role and location of alkali counter ions within such DNA lesions. The difficulty to identify partially occupied positions of Na^+ ions and to differentiate them from water molecules in a distorted DNA fragment appears to be even more severe than in a regular DNA fragment. A major question is whether co-ordination of the positively charged platinum(II) ion to a nucleobase reduces the propensity of a nucleotide as a whole for interactions with other cations. As we have recently shown,[17] binding of Pt^{II} to N(7) of 5′-dGMP does influence binding of additional $Cu(phen)^{2+}$ and $Cu(bpy)^{2+}$ to the phosphate group of the nucleotide only marginally. More significant in the context of the present work is the question whether binding of Pt^{II} to the heterocyclic part of a nucleobase affects interactions with other metal ions, *e.g.* alkali metal ions. In numerous model studies it has been demonstrated that co-ordination of a heavy metal occurring with concomitant base deprotonation, hence anion formation, facilitates binding of additional metal ions. These may include also alkali metal ions, as demonstrated by X-ray crystallography for complexes of Hg,[18] Pd,[19,20] and Pt,[21] for a combination of two different metal ions, Pt/Ag[22] and Pt/Co.[23] We are not aware of any previous reports of alkali metal ion binding to metal complexes containing *neutral* nucleobases as discussed in this work. The use of neutral model nucleobases rather than anionic nucleotides (which require cations *per se* for charge neutralization) appeared to us advantageous in that they might reveal inherently favourable alkali metal ion binding sites irrespective of charge neutralization arguments.

† Electronic supplementary information (ESI) available: details of the structure of complex **1**, unit cell contents of **2**. See http://www.rsc.org/suppdata/dt/b0/b004906i/

Experimental

Complex preparations

***trans*-$Cs[Pt(NH_3)(mura)I_2]\cdot 4H_2O$ 1 (mura = 1-methyluracilate).** To a suspension of *cis*-$[Pt(NH_3)_2(mura\text{-}N^3)Cl]\cdot H_2O$ (941 mg, 1.5 mmol)[24] in 30 cm^3 of water were added $AgNO_3$ (255 mg, 1.5 mmol, dissolved in 5 cm^3 of water) and 3 cm^3 of Me_2SO. The reaction mixture (pH 4–5) was stirred at 40 °C for 4 d with daylight excluded. Subsequently the AgCl precipitate was filtered off, the filtrate evaporated to dryness and 15 cm^3 of water and KI (622 mg, 3.75 mmol) were added. After stirring the mixture at 50 °C for 2 d, yellow *cis*-$[Pt(NH_3)_2(mura\text{-}N^3)I]$ was filtered off (*ca.* 50% yield) and the filtrate allowed slowly to evaporate at 4 °C. After three weeks orange cubes of *trans*-$[Pt(NH_3)_4][Pt(NH_3)(mura\text{-}N^3)I_2]_2\cdot 5H_2O$ were filtered off (7% yield). Both products were characterized by elemental analysis and X-ray crystallography. Details will be reported elsewhere. To the filtrate an excess of CsI was added, which led to precipitation of complex **1** in 19% yield (Found: C, 7.5; H, 1.8; N, 5.3. Calc. for $C_5H_{16}CsI_2N_3O_6Pt$: C, 7.5; H, 2.0; N, 5.3%). IR: 3475m, 1643s, 1537s, 1476m, 1450s, 1369s, 1331m, 772w, 598w and 493w cm^{-1}.

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Table 1 Crystallographic data for compounds **1**, **2** and **3**

	1	**2**	**3**
Chemical formula	$C_5H_{16}CsI_2N_3O_6Pt$	$C_5H_{11}F_9K_{0.5}N_4O_{1.5}P_{1.5}Pt_{0.5}$	$C_{17}H_{29}N_{14}O_{17.5}Cl_3NaPt$
M/g mol^{-1}	796.01	485.73	1033.97
Crystal system	Monoclinic	Monoclinic	Triclinic
Space group	$P2_1/n$	$C2/c$	$P\bar{1}$
a/Å	7.167(1)	8.229(2)	10.990(2)
b/Å	12.900(3)	22.036(4)	12.273(2)
c/Å	18.697(4)	15.385(3)	13.743(3)
α/°			89.84(3)
β/°	98.30(3)	90.48(3)	70.51(3)
γ/°			88.28(3)
V/Å^3	1710.5(6)	2784.8(10)	1746.6(6)
T/K	293(2)	163(2)	293(2)
Z	4	8	2
μ(Mo-Kα)/mm^{-1}	3.091	5.514	4.352
No. reflections collected	5913	4342	5977
No. independent reflections [$I > 2\sigma(I)$]	3040	2523	5977
R_{int}	0.059	0.040	0.050
$R1$ (obs. data)[a]	0.0422	0.0292	0.0547
$wR2$ (obs. data)[a]	0.0943	0.0642	0.1129

***trans*-K[Pt(NH$_3$)$_2$(mcyt-*N*^{3})$_2$][PF$_6$]$_3$·H$_2$O 2 (myct = 1-methylcytosine).** To an aqueous solution of *trans*-[Pt(NH$_3$)$_2$(mcyt-*N*^{3})$_2$][NO$_3$]$_2$[25] was added KPF_6 in excess (*ca*. 5 equivalents) and the mixture allowed to evaporate to dryness. The resulting mixture contained, among others, long colourless columns (estimated yield 20–25%) which were separated by hand under a microscope. According to EPXMA (electron probe X-ray microanalysis) these crystals contained both K and Pt in a 1 : 1 ratio. They were characterized by X-ray analysis.

***trans*-[Pt(NH$_3$)(Hmgua-*N*^{7})$_2$(mcyt-*N*^{3})Na(H$_2$O)$_2$][ClO$_4$]$_3$·0.5H$_2$O 3 (Hmgua = 9-methylguanine).** To a suspension of *trans*-[Pt(NH$_3$)(mcyt-*N*^{3})I$_2$] (296 mg, 0.5 mmol)[26] and Hmgua (165 mg, 1 mmol) in 40 cm^3 of water, $AgClO_4$ (205 mg, 0.99 mmol, dissolved in 10 cm^3 of water) was added dropwise over a period of 4 h. After stirring the mixture for 2 d at 40 °C and for 3 d at room temperature with daylight excluded, it was cooled to 4 °C and AgI filtered off. The filtrate was brought to a volume of 12 cm^3 by rotary evaporation and then allowed further to evaporate in air. The colourless precipitate that had formed (yield 62%) analysed as [Pt(NH$_3$)(Hmgua)$_2$(mcyt)][ClO$_4$]$_2$·4H$_2$O **3′** (Found: C, 21.5; H, 3.3; N, 20.9. Calc. for $C_{17}H_{32}Cl_2N_{14}O_{15}Pt$: C, 21.8; H, 3.4; N, 20.9%). A sample of this compound (188 mg, 0.2 mmol) was dissolved in the minimum amount of hot water, treated with 5 cm^3 of a saturated aqueous solution of $NaClO_4$ and then cooled to room temperature. Within 1–2 d a colourless precipitate of **3** formed, which was filtered off and washed with water. Cooling of the filtrate to 4 °C gave a second fraction of **3** as colourless crystals. The total yield was 75% (Found: C, 20.4; H, 2.7; N, 20.1. Calc. for $C_{17}H_{25}Cl_3N_{14}NaO_{15.5}Pt$: C, 20.5; H, 2.5; N, 19.6%). IR of **3**: 3584s, 3449vs, 3358vs, 3228vs, 3153vs, 1685vs, 1636vs, 1594vs, 1561s, 1198s, 1437s, 1358s, 1095vs, 775s, 657vs, 535w, 421m and 354m cm^{-1}.

Spectroscopic studies

IR spectra (KBr pellets) were recorded on a Bruker IFS 28 spectrometer, ^{1}H NMR spectra on Bruker AC200 and DRX 500 FT instruments. Chemical shifts are given in ppm and referenced to internal sodium 3-(trimethylsilyl)propane-1-sulfonate set to 0.0 ppm. 2-D NOESY spectra were recorded in 90% water–10% D_2O at 293 K with mixing times of 1.5 s. Water suppression was achieved with a modified WATERGATE 3-9-19 pulse sequence with gradients.[27] For the rotating-frame Overhauser enhancement spectroscopy (ROESY)[28] spectra in DMF-d_7 at the same temperature a mixing time of 800 ms and a recycling delay of 1.5 s were used. In both cases, a total of 256 t_1 increments, each with 2048 t_2 complex points, was collected with each free-induction decay (FID) as the average of 80 transients. Linear prediction to 512 data points was employed in the F1 dimension. A square cosine bell function was applied in both dimensions to improve the resolution. Integration of 2-D spectra was accomplished using Bruker XWINNMR Software, Version 2.5.

Crystallography

Intensity data for all crystal structures presented were collected on an Enraf-Nonius-KappaCCD (Mo-Kα, $\lambda = 0.71069$ Å, graphite monochromator). Data processing was performed using DENZO and SCALEPACK.[29] The structures were solved by standard Patterson methods[30] and refined by full-matrix least squares based on F^2 using the SHELXTL PLUS[31] and SHELXL-93 programs.[32] All non-hydrogen atoms have been refined anisotropically, except for one ring carbon atom, the disordered oxygens of the perchlorate anions and three half occupied water molecules in complex **3** in order to save parameters. The water proton in **2** was localized with Fourier syntheses and fully refined. Crystal data and data collection parameters are summarized in Table 1.

CCDC reference number 186/2133.

See http://www.rsc.org/suppdata/dt/b0/b004906i/ for crystallographic files in .cif format.

Results and discussion

Alkali metal ion binding to platinum complexes containing nucleobase anions

Deprotonation of T and U bases at the N(3) position and subsequent binding of Pt^{II} to this site leads to an electron distribution that leaves substantial basicity at the exocyclic oxygen atoms. This situation, concluded qualitatively from IR spectroscopic changes of nucleobase vibrations[33,34] and quantitatively from UV spectroscopy[35] and potentiometric titration,[36] has been reflected by the ready formation of numerous dinuclear platinum complexes or heterometal Pt_xM_y complexes.[37] It therefore is not surprising to see alkali metal ion binding to exocyclic oxygen atoms of N(3) platinated U and T model base, even though the number of X-ray structurally characterized examples is rather limited.[21,22] We note, however, that the situation in the mixed Hg,Na[18] and mixed Pd, alkali metal[19,20] compounds of pyrimidine nucleobases as well as structurally related glutaramidate complexes of Pd^{II}[38] is to be considered analogously. Two cases can be differentiated. (i) The heavy metal complex is negatively charged. This situation applies,

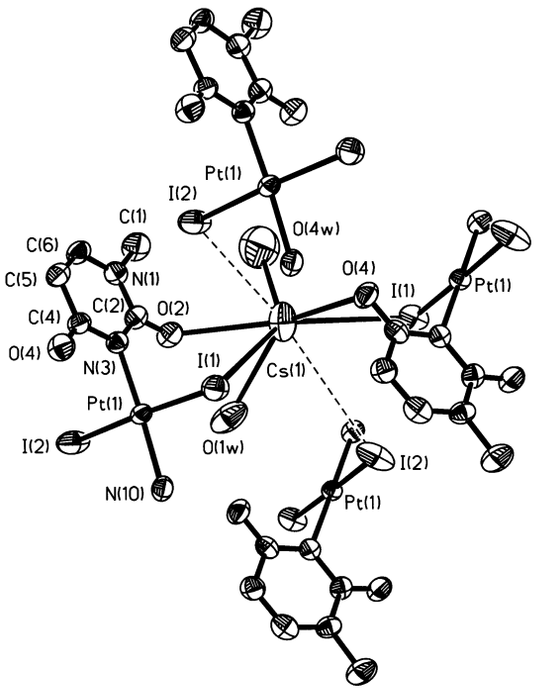


Fig. 1 Section of the polymeric structure of *trans*-Cs[Pt(NH_3)(mura-N^3)I_2]·4H_2O **1** with the atom numbering scheme. Ellipsoids are drawn at the 50% probability level.

Table 2 Selected bond length (Å), angles (°) and interatomic distances (Å) for *trans*-Cs[Pt(NH_3)(mura-N^3)I_2]·4H_2O **1** [a]

Pt(1)–N(10)	2.061(7)	Cs(1)–O(2)	3.163(7)
Pt(1)–N(3)	2.042(7)	Cs(1)–O(4)^2	3.139(7)
Pt(1)–I(1)	2.5778(10)	Cs(1)–O(1w)	3.32(1)
Pt(1)–I(2)	2.6059(9)	Cs(1)–O(4w)^3	3.11(2)
Cs(1)–I(1)	3.936(2)	Cs(1)···I(2)^4	4.206(2)
Cs(1)–I(1)^1	3.988(2)	Cs(1)···I(2)^5	4.457(2)
N(10)–Pt(1)–N(3)	177.4(3)	N(3)–Pt(1)–I(1)	93.5(2)
N(10)–Pt(1)–I(1)	88.6(2)	N(3)–Pt(1)–I(2)	89.0(2)
N(10)–Pt(1)–I(2)	88.8(2)	I(1)–Pt(1)–I(2)	177.46(2)

[a] Symmetry operations: 1 $-x-0.5$, $y-0.5$, $-z+0.5$; 2 $-x-0.5$, $y+0.5$, $-z+0.5$; 3 $x-1$, y, z; 4 $x+1$, y, z; 5 $-x+0.5$, $y-0.5$, $-z+0.5$.

for example, to *trans*-K_2[Pt(mura-N^3)$_2I_2$][19] and M_2[Pd(mura-N^3)$_4$].[20] Alkali metal binding involves all four and eight exocyclic oxygen atoms, respectively. Formation of these adducts is not really surprising considering that these cations are required for charge neutralization. (ii) The heavy metal species is neutral. This is the case with [HgMe(mthy-N^3)]·0.5$NaNO_3$[18] and [Pd(en)(mthy-N^3)$_2$]·2$NaNO_3$·H_2O[21] (mthy = 1-methylthymine anion). Clearly, there is no argument in favour of charge neutralization possible. Rather, this situation reflects the mentioned electronic structure in the metallated bases, which attracts additional cations. With more than one T or U base bonded per heavy metal ion,[19–22] the alkali metals are chelated by exocyclic oxygen atoms. In the other compound, [HgMe(mthy-N^3)]·0.5$NaNO_3$,[18] Na^+ is cross-linking oxygen atoms from different molecules.

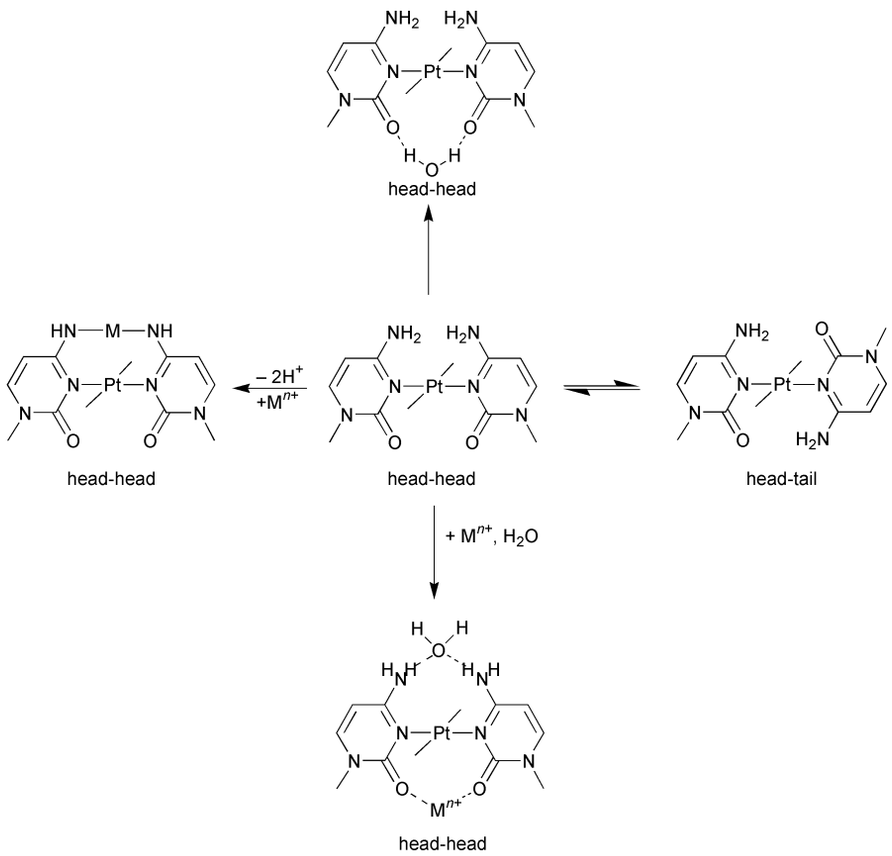
In *trans*-Cs[Pt(NH_3)(mura-N^3)I_2]·4H_2O **1** the alkali metal cation is again required for charge neutralization (case(i)). The structure of **1** is polymeric (Fig. 1, Table 2). The co-ordination geometry of Pt^{II} is square planar with no particularly unusual bond lengths and angles. The distance between Pt and the bridging iodo ligand (I(1)) is slightly larger than that between Pt and the terminal iodide (I(2)). The mura base is almost perpendicular to the platinum co-ordination plane (88.6(2)°). There are no significant differences in bond lengths and angles of the nucleobase as compared to other platinum(II) complexes with mura-N^3 ligands.[36] In particular, the CO bonds are not elongated as a consequence of Cs^+ binding. Cs^+ ions are bridging the complex ions *via* O(2) and O(4) (symmetry operation $-x-\frac{1}{2}$, $y+\frac{1}{2}$, $-z+\frac{1}{2}$) as well as through I(1) ligands of two anions. The octahedral co-ordination sphere of Cs^+ is completed by two water molecules (O(1w) and O(4w)). In addition there are two very long (4.206(2) and 4.457(2) Å) contacts of Cs^+ with I(2) ligands of two neighbouring complex anions. The polymeric network of **1** can be dissected into two interconnected chains running along the y and x axes, respectively. In the first one, Cs^+ ions are arranged in a zigzag fashion along the y axis, with Cs^+ bridged by I(1) ligands (3.936(2), 3.988(2) Å), by mura bases (O(2), O(4)), and in addition by the long $Cs^+\cdots$I(2) contacts mentioned above. When viewed along the x axis the Cs^+ ions are arranged in a collinear fashion, bridging pairs of Pt containing anions (Supplementary Material). Distances between neighbouring Cs^+ ions are 7.005(2) Å in the first chain and 7.167(1) Å in the second. There is partial stacking (3.5 Å) between mura bases of adjacent chains in the x direction.

Alkali ion binding to cationic platinum nucleobase complexes

If there is a rationale for alkali metal ion binding to a deprotonated nucleobase bound to a heavy metal in anionic or neutral complexes, there is certainly not an obvious one in the case of neutral nucleobases bound to a dipositive heavy metal cation, unless the latter exerts a profound "back bonding" effect (*e.g.* with Ru^{II}). Usually heavy metal binding to a neutral nucleobase acidifies the ligand and reduces the donor strength of other sites. While this effect can be quantified in cases where still available endocyclic N donors permit protonation of the metallated base (*e.g.* with adenine and guanine nucleobases), it is more difficult to assess the influence of the heavy metal on the donor properties of exocyclic oxygen atoms. In the following we describe two cases of unexpected alkali metal ion binding to O(2) of myct and O(6) of Hmgua in cationic platinum(II) complexes.

trans-[Pta$_2$(mcyt-N^3)$_2$]$^{2+}$ (a = NH_3[25] or CH_3NH_2[39,40]) has previously been shown preferentially to crystallize in a *head-tail* arrangement of the two bases (Scheme 1), although in solution it coexists with the *head-head* rotamer. Only under special conditions the isolation of a *head-head* form has been possible.[39] There a water molecule, interacting with the O(2) oxygen atoms through hydrogen bonds, appears to be responsible for allowing this orientation (Scheme 1, top). Another way of stabilizing a head-head arrangement involves metal cross-linking of the deprotonated exocyclic amino groups[41] and shall not be discussed here (Scheme 1, left).

We have now isolated a K^+ adduct of the bis(1-methylcytosine) complex, *trans*-K[Pt(NH_3)$_2$(mcyt-N^3)$_2$][PF_6]$_3$·H_2O **2** which, at least in the solid state, likewise displays a head-head orientation of the two bases (Fig. 2) due to simultaneous chelation of the alkali metal ion by the two O(2) oxygen atoms and hydrogen bonding of a water molecule *via* the N(4)H_2 positions (Scheme 1, bottom). The two mcyt rings form a relatively large dihedral angle of 41.0(2)° which leads to separations between the two carbonyl oxygens and the two amino groups of 3.966(7) and 4.827(9) Å, respectively. The angle N(3)–Pt(1)–N(3) (174.6(2)°) (in direction of the K^+ ion) deviates significantly from the ideal one in a square planar environment. The potassium ion is surrounded by six fluorine atoms of the hexafluorophosphate anions (2.70(4)–3.170(5) Å) and the two carbonyl oxygens (2.689(4) Å). The two O(2) oxygen atoms and the six fluorine atoms form a distorted antiprism. Another stabilization of the head-head conformation in the solid state is achieved through hydrogen bonding of a water molecule to both exocyclic amino groups (O(1w)···N(4), 2.923(6) Å). The protons of the water molecule whose oxygen atom lies on a C_2 axis were located by Fourier difference synthesis. The packing



Scheme 1

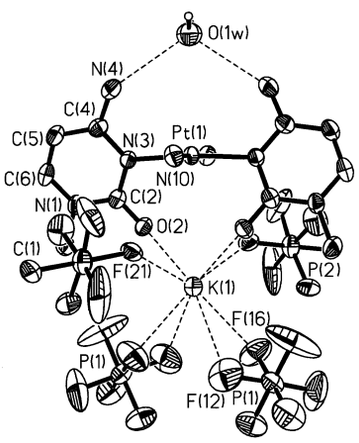


Fig. 2 Section of *trans*-K[Pt(NH_3)$_2$(mcyt-N^3)$_2$][PF_6]$_3$·H_2O **2** and the atom numbering scheme. Ellipsoids are drawn at the 50% probability level.

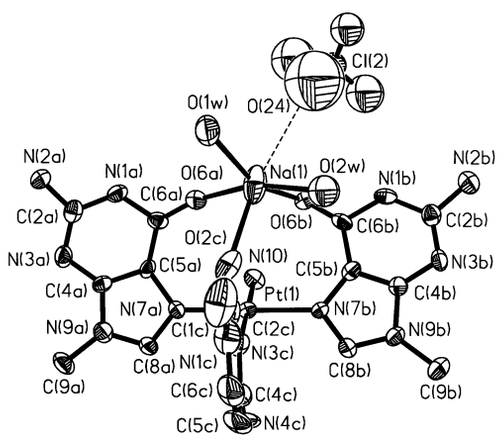


Fig. 3 Section of *trans*-[Pt(NH_3)(Hmgua-N^7)$_2$(myct-N^3)Na(H_2O)$_2$]-[ClO_4]$_3$·0.5H_2O **3** with the atom numbering scheme. Ellipsoids are drawn at the 30% probability level.

pattern of the structure reveals no contacts between cations. Worth mentioning is that one of the PF_6^- anions bridges two K^+ ions to give infinite chains along the *x* axis. Each water molecule makes additional contacts to four fluorine atoms of the PF_6^- anion, 2.957(5) and 3.068(6) Å (Supplementary Material). Selected bond length, interatomic distances (Å) and angles (°) are summarized in Table 3.

trans-[Pt(NH_3)(Hmgua-N^7)$_2$(mcyt-N^3)]$^{2+}$ **3′** is a tris(nucleobase) complex which upon cocrystallization with $NaClO_4$ yields *trans*-[Pt(NH_3)(Hmgua-N^7)$_2$(mcyt-N^3)Na(H_2O)$_2$] [ClO_4]$_3$·0.5H_2O (**3**). In the cation all three nucleobases are in a *head, head,head* conformation as far as the positions of the carbonyl groups are concerned (Fig. 3). The sodium ion is located nearly perpendicular above the platinum (Na(1)–Pt(1)–N, 84.2(2)–95.7(2)°). It is surrounded by six oxygen atoms, namely the three carbonyl oxygen atoms of the nucleobases, two water oxygen atoms, and an oxygen atom of the perchlorate ion. The latter

Table 3 Selected bond length (Å), angles (°) and interatomic distances (Å) for *trans*-K[Pt(NH_3)$_2$(mcyt-N^3)$_2$][PF_6]$_3$·H_2O **2**[a]

Pt(1)–N(1)	2.035(4)	K(1)···F(12)	3.170(5)
Pt(1)–N(3)	2.051(4)	N(4)···O(1w)[3]	2.923(6)
K(1)–O(2)	2.689(4)	O(1w)···F(14)[4]	3.068(6)
K(1)–F(21)[2]	2.84(4)	O(1w)···F(16)[4]	2.957(5)
K(1)–F(16)	2.829(4)		
N(10)–Pt(1)–N(3)	90.5(2)	N(3)–Pt(1)–N(3)[1]	174.6(2)
N(10)–Pt(1)–N(10)[1]	179.6(2)		

[a] Symmetry operations: 1 $-x+1$, y, $-z+1.5$; 2 $-x+2$, $-y+1$, $-z+2$; 3 $-x+1.5$, $-y+1.5$, $-z+2$; 4 $-x+1$, $-y+1$, $-z+2$.

distance is significantly longer (2.88(2) Å) than the other five distances, which range between 2.261(9) and 2.58(1) Å. Moreover, one of the water molecules (O(2w)) and the perchlorate oxygen atoms, *viz.* also O(24), show disorder (occupancy fac-

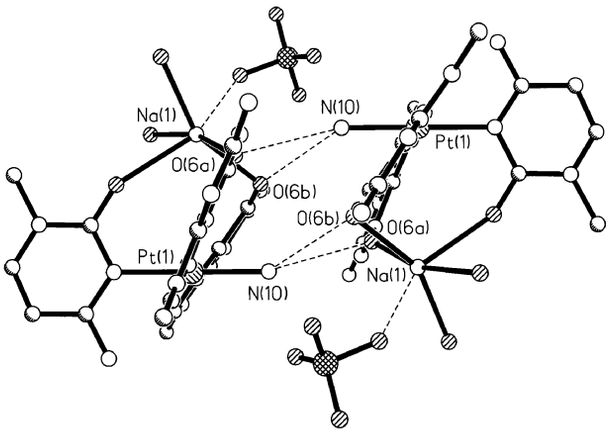


Fig. 4 Pair of cations of complex **3** with hydrogen bonding interactions indicated. There is also stacking between guanine bases.

Table 4 Selected bond length (Å) and interatomic distances (Å) for *trans*-[Pt(NH$_3$)(Hmgua-N^7)$_2$(mcyt-N^3)Na(H$_2$O)$_2$][ClO$_4$]$_3$·0.5H$_2$O **3**[a]

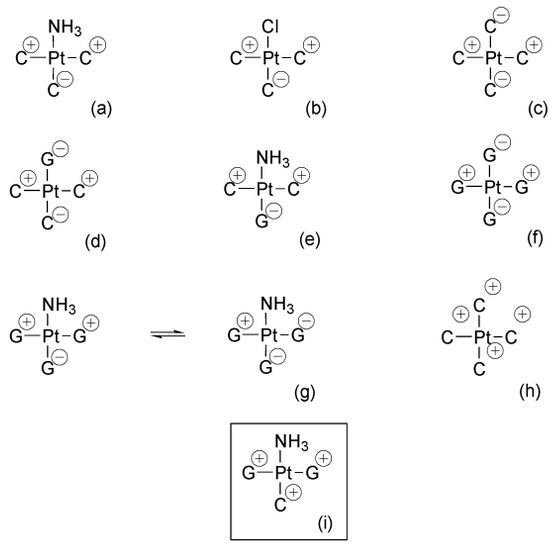
Pt(1)–N(10)	2.020(7)	Pt(1)–N(7b)	2.014(7)
Pt(1)–N(7a)	2.015(7)	Pt(1)–N(3c)	2.039(8)
N(10)···O(6a)[1]	3.054(9)	Na(1)–O(2c)	2.58(1)
N(10)···O(6b)[1]	3.230(10)	Na(1)–O(1w)	2.308(9)
Na(1)–O(6a)	2.261(9)	Na(1)–O(2w)	2.40(2)
Na(1)–O(6b)	2.379(8)	Na(1)···O(24)	2.88(2)

[a] Symmetry operation: 1 $-x$, $-y+1$, $-z+1$.

tors of 0.5 each). The geometry of the Na^+ ion is, with the perchlorate oxygen included and considering the disorder of O(24) and O(2w), distorted octahedral. If the perchlorate oxygen atom is not considered a ligand of Na^+, the environment of Na^+, as analysed by its structure index τ,[42] is not unambiguous. Depending on the position of the disordered water molecule O(2w), O(2wa), either a distorted square pyramid ($\tau = 0.18$, O(2wa)) or a distorted trigonal bipyramid ($\tau = 0.69$, O(2w)) is the result of such an analysis. Calculating the structure index τ considering just the mean position of the two half occupied water molecules gives the more reasonable value of 0.44, pointing to an arrangement which is approximately in between the two possible extremes. The planes of the two Hmgua bases form dihedral angles with the PtN_4 plane of 67.3(2) and 55.3(2)° thus deviating significantly from a perpendicular orientation. Through this the formation of hydrogen bonded dimers with bonds between the carbonyl oxygens of the Hmgua bases of one cation and the ammine ligand of the second one is made possible (3.054(9) and 3.230(10) Å, respectively; Fig. 4). In these dimers also a stacking interaction between crystallographically independent guanine bases is observed (3.4 Å, dihedral angle 12.0(4)°). The packing pattern reveals also stacking interactions between equivalent Hmgua nucleobases (3.5 Å each) but in the case of base B only the exocyclic amino group overlaps with the other ring system. For selected bond length and interatomic distances (Å) see Table 4.

Nucleobase orientation in complex 3

A comparison of structurally characterized tris- and tetrakis-(nucleobase) complexes of Pt^{II} (Scheme 2)[43–48] reveals that the orientation of the bases usually is such as to produce an orientation of exocyclic oxygen atoms which is *up*,*down*,*up* and *up*,*down*,*up*,*down*, respectively, relative to the platinum coordination plane, meaning that the two *trans*-arranged bases are head,head. Such an orientation appears frequently to lead to a maximum of intramolecular hydrogen bonds between exocyclic nucleobase groups (*e.g.* NH_2 and O with cytosine bases) and may be favoured for this reason. Alternatively the mutual



⊕ exocyclic oxygen up, relative to Pt co-ordination plane

⊖ exocyclic oxygen down, relative to Pt co-ordination plane

C = mcyt-N^3

G = H(mgua)-N^7 or H(egua)-N^7

(a) ref.43; (b) ref. 44; (c) ref. 44; (d) ref. 45; (e) ref. 46; (f) ref. 48; (g) ref. 47; (h) ref. 23 (Co/Na adduct); (i) this work, compound **3** (Na adduct)

Scheme 2

orientation of the dipole moments of the individual bases may be particularly favourable for this arrangement. Only in a single case, in one of two crystallographically different cations of the tris(nucleobase) complex *trans*-[Pt(NH$_3$)(Hmgua-N^7)$_3$]$^{2+}$, an exception has been observed (*up*,*down*,*down*).[47] A second exception refers to a heteronuclear derivative of [Pt(mcyt-N^3)$_4$]$^{2+}$, which contains a Co^{III} bound to four deprotonated exocyclic amino groups of the nucleobase and a Na^+ bound to the four exocyclic O(2) oxygen atoms of the same bases.[23] Clearly, in the latter case binding of the two different metal ions with their different affinities for N (Co^{III}) and O (Na^+) donors is of importance.

Solution behaviour of complex 2

As has previously been shown by us, the rotamer equlibrium of bis(nucleobase) complexes of type *trans*-[Pta$_2$(nb)(nb′)]X$_n$ (with a = NH_3 or $MeNH_2$; nb, nb′ = nucleobase; X = anion) depends on several factors such as solvent, concentration and counter anion X.[39,49] Results of the crystal structure analysis of **2** suggest that the presence of cations capable of cross-linking exocyclic oxygen atoms of nucleobases likewise might influence the rotamer distribution.

The ^{1}H NMR spectrum of *trans*-[Pt(NH$_3$)$_2$(mcyt-N^3)$_2$][NO$_3$]$_2$ recorded in D_2O consists of two sets of cytosine resonances in a 1.7:1 ratio due to two rotamer forms, head-tail and head-head. Addition of a large excess of an alkali metal salt (KPF_6, 10 equivalents) does not influence the rotamer distribution measurably, unlike metal ion binding that involves deprotonation of the exocyclic amino groups and cross-linking of the two cytosine bases. The latter leads to a complete shift toward the head-head form.[39,41] Consequently K^+ binding to the two cytosine bases as seen in **2** is to be considered relatively weak. Compound **2** does not readily dissolve in water. Once dissolved (*ca.* 2 min at 25 °C), however, the ^{1}H NMR spectrum reveals a rotamer distribution that is virtually identical with that of the starting compound in the absence of K^+. Thus, unlike in the methylamine analogue, for which rotamer equilibration is slow

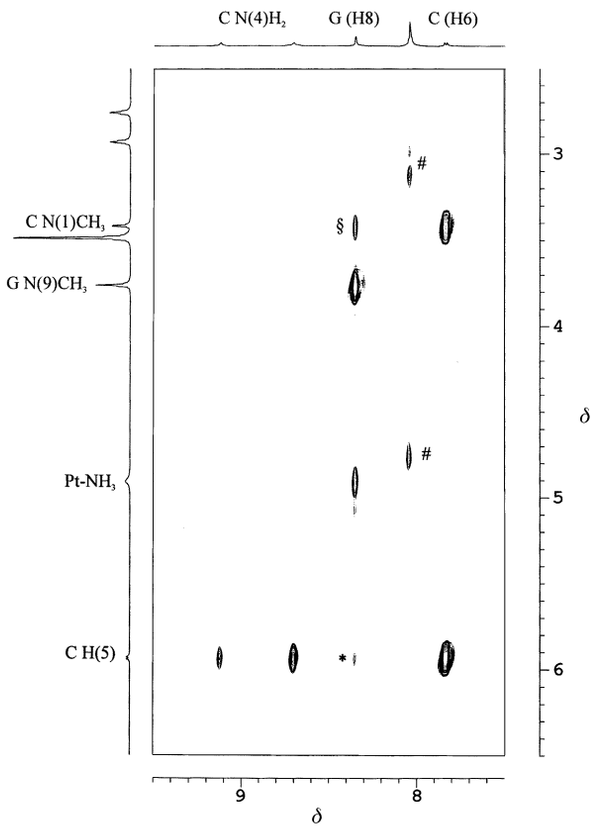


Fig. 5 2-D ROESY spectrum (DMF-d_7, room temperature, 500 MHz, t_m 800 ms) of a solution of *trans*-[Pt(NH_3)(Hmgua)$_2$(mcyt)][ClO_4]$_2$ **3′**. The cross-peaks between guanine H(8) and methylcytosine N(1)CH_3 (§) and methylcytosine H(5) (*), respectively, are marked. The signals indexed with # are artefacts due to t_1 noise from the solvent peak.

(*ca.* 8 h at 22 °C, pD 7.2),[39] the NH_3 ligands in **2** represent no major blockage for rotation of the cytosine nucleobases.

NMR solution spectra of complexes 3 and 3′

The ^{1}H NMR spectrum of *trans*-[Pt(NH_3)(Hmgua)$_2$(mcyt)]-[ClO_4]$_2$ **3′** in D_2O (pD 5.7, ambient temperature) displays single sets of resonances for Hmgua (δ H8, 8.16; CH_3, 3.74) and mcyt (δ H6, 7.53, d, ^3J 7.1 Hz; H5, 5.91, d; CH_3, 3.42) of the expected relative intensities of 2:1. The aromatic protons of the mcyt ligand are slightly upfield from those of the free, neutral nucleobase (δ H6, 7.54; H5, 5.95, pD 9) rather than downfield, which is generally the rule for binding of Pt^{II} to N(3), unless stacking effects come into play. It appears that the π systems of the two guanine ligands are responsible for this feature, similar to the situation in [Pt(mcyt-N^3)$_3$Cl]$^+$.[46] There is no indication from the ^{1}H NMR spectrum for the presence of a second rotamer in the temperature range +20 to +70 °C.

If DMF-d_7 is used as the solvent, there is likewise no sign of the existence of individual rotamers in the temperature range from −55 to +72 °C, *viz.* no second set of carbon bonded protons of the nucleobases is observed. The only dynamic processes seen refer to coalescence of the two cytosine NH_2 resonances above *ca.* 55 °C and of the two guanine NH_2 resonances above −10 °C. While it is not surprising to see the NH_3, NH_2 and NH resonances being temperature-sensitive (*e.g.* downfield shifts with decreasing temperature), it is striking that also the aromatic protons of nucleobases undergo slight downfield shifts with decreasing temperature, which are in the order of 0.3 ppm for both guanine H(8) and cytosine H(6) and 0.1 ppm for cytosine H(5) protons, respectively over the entire temperature range. In contrast, the CH_3 protons of the three bases are virtually unaffected.

NOESY and ROESY spectra (water–D_2O 90:10; DMF-d_7) reveal cross-peaks of the guanine H(8) with both cytosine-N(1)CH_3 (strong) and cytosine-H(5) (weak) (Fig. 5). Inspection of a model of complex **3′** reveals that a cross-peak between HG–H(8) and C–N(1)CH_3 is to be expected only if guanine and cytosine bases are mutually *up* and *down*, hence in an orientation not seen in **3**. On the other hand, the second (weak) cross-peak is to be expected only when these two bases are mutually *up*,*up*. This finding strongly suggests that there is rapid interconversion (on the ^{1}H NMR timescale) of the bases over the entire temperature range. In a structurally related tris(nucleobase) complex, [Pt(NH_3)(Hmgua-N^7)$_3$]$^{2+}$ [47] we have obtained crystallographic evidence for the existence of two different rotamers (Scheme 2, (g)) in the solid state, while in solution an averaged structure occurs, according to ^{1}H NMR spectroscopy. As in this case, we propose that the two guanine ligands in **3′** have a preference for an *up*,*down* orientation with respect to cytosine (which gives rise to the intense cross-peak), but that it is easy for the guanine to swing past the NH_3 ligand into an *up*,*up* arrangement with cytosine. Once in this orientation the Na^+ ion is capable of stabilizing such an orientation (*cf.* crystal structure of **3**).

Addition of $NaClO_4$ to a solution of complex **3** in D_2O causes slight upfield shifts of all proton resonances. For example, at c(**3′**) = 10^{-2} M and c(Na^+) = 2×10^{-1} M the shifts are in the order of 0.03–0.06 ppm. It is difficult to say whether this effect is due to a non-specific solvent effect, due to Na^+ co-ordination, a change in the rotamer equilibrium, or a combination of all. In any case averaged resonances are observed only. Our attempt to correlate the intensity of the ROESY cross-peak between HG–H(8) and C–N(1)CH_3 in the absence and presence (20 fold excess) of Na^+ was not fully satisfactory, although it followed the expected trend. Thus a *ca.* 25% reduction of the cross-peak intensity (relative to cross-peak between cytosine H(5) and H(6) protons) is consistent with an increase in the *up*,*up*,*up* rotamer concentration due to Na^+ binding.

Conclusion

Here we have shown that alkali metal ions are capable of co-ordinating to the three O-containing nucleobases U, C and G even when these are already platinated and regardless if the base is deprotonated or neutral and thus irrespective of the charge of the Pt–nucleobase moiety (anionic, neutral, cationic). Our findings strongly suggest that inner-sphere binding of alkali metal ions not only is of importance in unperturbed nucleic acids or in the stabilization of particular multistranded nucleic acid structures, but possibly in nucleic acids modified or distorted by heavy metal ions as well. Considering the negative charge of polynucleotides it is in fact likely that this feature is of even greater significance in reality than in model systems such as the one studied in this work. The fact that this aspect has not gained much attention so far is a consequence of the relatively few high-resolution crystal structures presently available on heavy metal–DNA fragments and the problem of differentiating Na^+ and H_2O. After all it took almost 20 years and numerous crystal structures of DNA fragments to establish alkali metal ion binding sites in undisturbed DNA! We are aware that a major problem for proving a role of alkali metal ion binding to metallated nucleobases or nucleic acids is the relative weakness of such interactions in solution. Moreover, we recognize a general shortcoming of such studies, namely the problem of solubility which is crucial for crystallization of a particular adduct. However, our findings with compounds **2** and **3** imply that alkali metal chelation, hence co-ordination by several O donor atoms, may be sufficiently strong to effect nucleobase orientation. It therefore must not be overlooked.

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References

1 M. A. Young, B. Jayaram and D. L. Beveridge, *J. Am. Chem. Soc.*, 1997, **119**, 59 and refs. therein.
2 X. Shui, L. Mc Fail-Isom, G. G. Hu and L. D. Williams, *Biochemistry*, 1998, **37**, 8341.
3 V. Tereshko, G. Minasov and M. Egli, *J. Am. Chem. Soc.*, 1999, **121**, 470.
4 V. Tereshko, G. Minasov and M. Egli, *J. Am. Chem. Soc.*, 1999, **121**, 3590.
5 G. Laughlan, A. I. H. Murchie, D. G. Norman, M. H. Moore, P. C. E. Moody, D. M. Lilley and B. Luisi, *Science*, 1994, **265**, 520; C. Kang, X. Zhang, R. Ratliff, R. Moyzis and A. Rich, *Nature* (*London*), 1992, **356**, 126.
6 G. Gupta, A. E. Garcia, Q. Guo, M. Li and N. R. Kallenbach, *Biochemistry*, 1993, **32**, 7098.
7 C. Cheong and P. B. Moore, *Biochemistry*, 1992, **31**, 8406.
8 J. C. Chaput and C. Switzer, *Proc. Natl. Acad. Sci. U.S.A.*, 1999, **96**, 10614.
9 T. Hermann and E. Westhof, *Structure*, 1998, **6**, 1303.
10 B. Fischer, H. Preut, B. Lippert, H. Schöllhorn and U. Thewalt, *Polyhedron*, 1990, **9**, 2199.
11 N. H. Sarma, J. Luo, K. Umemoto, R.-D. Yuan and R. H. Sarma, *J. Biomol. Struct. Dyn.*, 1992, **9**, 1131.
12 E. Freisinger, A. Schimanski and B. Lippert, submitted.
13 G. Mallet, S. Ansiss and D. Vasilescu, *J. Biomol. Struct. Dyn.*, 1998, **16**, 21.
14 P. M. Takanara, A. C. Rosenzweig, C. A. Frederick and S. J. Lippard, *Nature* (*London*), 1995, **377**, 649.
15 U.-M. Ohndorf, M. A. Rould, Q. He, C. O. Pabo and S. J. Lippard, *Nature* (*London*), 1999, **399**, 708.
16 F. Coste, J.-M. Malinge, L. Serre, W. Shepard, M. Roth, M. Leng and C. Zelwer, *Nucl. Acids Res.*, 1999, **27**, 1837.
17 M. S. Lüth, B. Song, B. Lippert and H. Sigel, *Inorg. Chem.*, 2000, **39**, 1305.
18 F. Guay and A. Beauchamp, *Inorg. Chim. Acta*, 1982, **66**, 57.
19 W. Micklitz, B. Lippert, F. Lianza and A. Albinati, *Inorg. Chim. Acta*, 1994, **227**, 5.
20 M. Mizutani, K. Jitsukawa, H. Masuda, Y. Aoyana and H. Einaga, *Chem. Lett.*, 1998, 663; M. Mizutani, K. Jitsukawa, H. Masuda and H. Einaga, *Chem. Commun.*, 1996, 1389.
21 O. Renn, B. Lippert and I. Mutikainen, *Inorg. Chim. Acta*, 1994, **218**, 117.
22 F. Zamora, H. Witkowski, E. Freisinger, J. Müller, B. Thormann, A. Albinati and B. Lippert, *J. Chem. Soc.*, *Dalton Trans.*, 1999, 175.
23 A. Hegmans, E. Zangrando, E. Freisinger, F. Pichierri, L. Randaccio, E. Mealli, M. Gerdan, A. X. Trautwein and B. Lippert, *Chem. Eur. J.*, 1999, **5**, 3010.
24 B. Lippert, D. Neugebauer and G. Raudaschl, *Inorg. Chim. Acta*, 1983, **78**, 161.
25 B. Lippert, C. J. L. Lock and R. A. Speranzini, *Inorg. Chem.*, 1981, **20**, 808.
26 M. Wienkötter, M. Sabat, G. Fusch and B. Lippert, *Inorg. Chem.*, 1995, **34**, 1022.
27 V. Sklenar, M. Piotto, R. Leppik and S. V. Saudek, *J. Magn. Reson. Ser. A*, 1993, **102**, 241.
28 A. Bax and D. G. Davis, *J. Magn. Reson.*, 1985, **63**, 207.
29 Z. Otwinowsky and W. Minor, DENZO and SCALEPACK, *Methods Enzymol.*, 1997, **276**, 307.
30 G. M. Sheldrick, *Acta Crystallogr.*, *Sect. A*, 1990, **46**, 467.
31 G. M. Sheldrick, SHELXTL PLUS (VMS), Siemens Analytical X-Ray Instruments, Inc., Madison, WI, 1990.
32 G. M. Sheldrick, SHELXL-93, Program for Crystal Structure Refinement, University of Göttingen, 1993.
33 B. Lippert and D. Neugebauer, *Inorg. Chim. Acta*, 1980, **46**, 171.
34 F. Guay, A. L. Beauchamp, C. Gilbert and R. Savoie, *Can. J. Spectrosc.*, 1983, **28**, 13.
35 O. Krizanovic and B. Lippert, *Platinum and Other Metal Coordination Compounds in Cancer Chemotherapy*, ed. M. Nicolini, Martinus Nijhoff, Boston, 1988, p. 700.
36 H. Schöllhorn, U. Thewalt and B. Lippert, *J. Am. Chem. Soc.*, 1989, **111**, 721 and refs. therein.
37 B. Lippert, *Prog. Inorg. Chem.*, 1989, **37**, 1 and refs. therein; B. Lippert, *Coord. Chem. Rev.*, 2000, in press.
38 D. Michalska, B. Morzyk, W. Wojciechowski and T. Glowiak, *Inorg. Chim. Acta*, 1996, **248**, 159.
39 D. Holthenrich, I. Sóvágó, G. Fusch, A. Erxleben, E. C. Fusch, I. Rombeck and B. Lippert, *Z. Naturforsch.*, *Teil. B*, 1995, **50**, 1767; D. Holthenrich, I. Sóvágó, G. Fusch, A. Erxleben, E. C. Fusch, I. Rombeck and B. Lippert, *Z. Naturforsch.*, *Teil. B*, 1996, **51**, 1368.
40 E. Zangrando, L. Randaccio, F. Pichierri, B. Lippert and D. Holthenrich, *Acta Crystallogr.*, *Sect. C*, 1996, **52**, 2468.
41 C. Mealli, F. Pichierri, L. Randaccio, E. Zangrando, M. Krumm, D. Holtherich and B. Lippert, *Inorg. Chem.*, 1995, **34**, 3418 and refs. therein.
42 A. W. Addison, T. N. Rao, J. Reedijk, J. van Rijn and G. C. Verschoor, *J. Chem. Soc.*, *Dalton Trans.*, 1984, 1349.
43 R. Faggiani, C. J. L. Lock and B. Lippert, *Inorg. Chim. Acta*, 1985, **106**, 75.
44 A. Hegmans, E. Freisinger, E. Zangrando, A. Ashfar, E. Hübener, T. G. Appleton and B. Lippert, *Inorg. Chim. Acta*, 1998, **279**, 152.
45 M. B. L. Janik, A. Hegmans, E. Freisinger and B. Lippert, *J. Biol. Inorg. Chem.*, 1999, **4**, 1130.
46 A. Hegmans, M. Sabat, I. Baxter, E. Freisinger and B. Lippert, *Inorg. Chem.*, 1998, **37**, 4921.
47 E. Freisinger, S. Meier and B. Lippert, *J. Chem. Soc.*, *Dalton Trans.*, 2000, 10.1039/b004993j.
48 H.-J. Korte and R. Bau, *Inorg. Chim. Acta*, 1983, **79**, 251.
49 S. Metzger, A. Erxleben and B. Lippert, *J. Biol. Inorg. Chem.*, 1997, **2**, 256.