Bioorganometallic Chemistry. 4. The Bonding Role of Carbonyl and Amino Functionalities in the Reactions of Guanine Nucleobase Derivatives with (η^5-Pentamethylcyclopentadienyl)rhodium Complexes in Methanol and Aqueous Solutions

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Surprisingly, there have been relatively few reported studies on the reactions of organometallic complexes with DNA/RNA nucleobases, nucleosides, nucleotides, and oligonucleotides.[1] These bonding studies, in aqueous solution, with these biologically significant nitrogen ligands, are of interest to our group in view of the future utility of the (η^5-pentamethylcyclopentadienyl)-rhodium aqua complex,[1a,2] $[Cp^*Rh(H_2O)_2(OTf)_2]_x$, as an anchor for single DNA molecules, in conjunction with surface microscopy techniques, for application to the goals of the human genome program.[3]

Recently, we determined the structure of the product from reaction of the Cp*Rh aqua complex with the nucleoside guanosine (Guo), in water (isolated at pH 5.4), principally by ^{1}H NMR, FAB-MS, and elemental analysis, and found that bonding occurred at N7 of the Guo nucleus to provide [Cp*Rh(η^1(N7)-Guo)(H_2O)(OH)](OTf) (**1**).[1a] Several questions were posed about the monomeric nature of the structure of **1**, a hydroxy–aqua derivative, and included the following: What was the bonding role, if any, of the carbonyl group at C6? What was the role of the NH1 group? What is the steric role of the NH_2 group at C2 on the guanine nucleus? As well, did the pH of the aqueous media or the use of an organic solvent affect the product structure? In order to answer these questions, we studied the reactions of Cp*Rh complexes with two guanine derivatives, 9-ethylguanine (9-EG) and 9-methylhypoxanthine (9-MH), a guanine nucleobase analogue without the NH_2 group at C2, in methanol and aqueous solutions (pH profile) and present these novel results in this communication.

The reaction of 1 equiv of 9-EG with *in situ* generated [Cp*Rh-$(MeOH)_3$](OTf)$_2$ in methanol produces [Cp*Rh(η^1(N7)-9-EG)$(CH_3OH)_2$](OTf)$_2$, **2**, whose ^{1}H NMR in MeOH-d_4 provided evidence of exclusive Cp*Rh-N7 bonding with an H8 downfield shift in comparison to free 9-EG of $\Delta\delta$ = 0.7 ppm.[4] Similarly, [Cp*Rh(η^1(N7)-9-MH)$(CH_3OH)_2$](OTf)$_2$ (**3**) was also prepared and showed a ^{1}H NMR (DMSO-d_6) downfield shift for H8 in comparison to free 9-MH of $\Delta\delta$ = 0.53 ppm and H2 of $\Delta\delta$ = 0.15 ppm, which is also consistent with Cp*Rh-N7 binding.[1a,5]

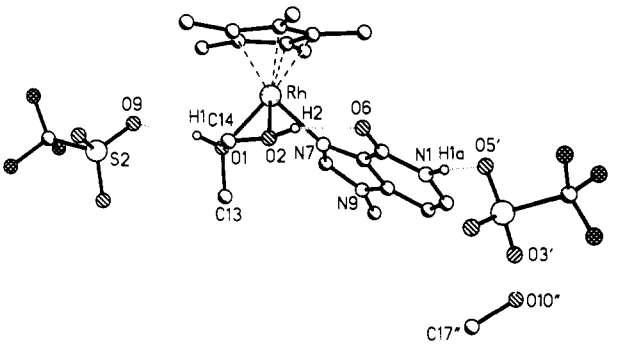

Figure 1. Molecular structure of **3**, [Cp*Rh(η^1(N7)-9-MH)$(CH_3OH)_2$]-(OTf)$_2$·CH_3OH. Selected bond lengths (Å) and angles (deg): Rh–C (average), 2.13; Rh–N7, 2.127(4); Rh–O1, 2.172(5); Rh–O2, 2.172(5); H2--O6, 1.83(1); H1--O9, 1.86(1); H1a--O5′, 1.89(1); O3′--O10″, 2.734-(1); N7–Rh–O2, 82.9(2); N7–Rh–1, 86.3(2); O1–Rh–O2, 83.0(2).

The single-crystal X-ray structure of **3**, determined on crystals grown from methanol/Et_2O, verifies this N7 coordination mode (Figure 1).[6] In addition, the X-ray structure of **3** also reveals an interesting pseudochelation of the rhodium center that involves a covalent Rh–N(7) bond and a hydrogen bond between the C=O6 of 9-MH and a coordinated MeOH ligand (1.83 Å). Hydrogen bonding also occurs between coordinated MeOH and one of the triflate oxygens (CF_3SO_2O--$HOCH_3$, 1.86 Å), as well as the other triflate anion to NH1 (CF_3SO_2O--HN, 1.89 Å). It is important to note that the carbonyl functionalities of oxopurines are known to coordinate to metals in weakly coordinating

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(4) [Cp*Rh(9-EG)$(MeOH)_2$](OTf)$_2$ (**2**): ^{1}H NMR (DMSO-d_6, δ (ppm)): 11.19 (–NH, 1H), 7.99 (H_8, 1H), 6.85 (NH_2, 2H), 4.11 (CH_2, q, 2H), 3.15 (CH_3OH, s, 6H), 1.47 (Me_5C_5, 15H), 1.33 (CH_3, t, 3H). ^{1}H NMR (MeOH-d_4, δ(ppm)): 8.43 (H_8, 1H), 4.25 (CH_2, q, 2H), 1.63 (Me_5C_5, 15H), 1.51 (CH_3, t, 3H). Anal. Calcd for $Rh_1C_{19}H_{24}N_5O_7S_2F_6$: C, 31.90; H, 3.38; N, 9.79. Found: C, 31.58; H, 3.37; N, 9.54. It should be noted that the elemental analysis was consistent with [Cp*Rh(9-EG)](OTf)$_2$, indicating that the weakly bound methanol ligands were removed under vacuum.

(5) [Cp*Rh(9-MH)$(MeOH)_2$](OTf)$_2$(**3**): ^{1}H NMR (DMSO-d_6, δ(ppm): 12.65 (–NH, 1H), 8.47 (H_8, 1H), 8.45 (H_2, 1H), 4.11 (Me, 3H), 3.15 (CH_3OH, s, 6H), 1.71 (Me_5C_5, 15H). (MeOH-d_4, δ(ppm): 8.72 (H_8, 1H), 8.31 (H_2, 1H), 4.01 (Me, 3H), 1.64 (Me_5C_5, 15H). Anal. Calcd for $Rh_1C_{20}H_{29}N_4O_9S_2F_6$: C, 32,01; H, 3.89; N, 7.47. Found: C, 32.31; H, 3.53; N, 7.78.

(6) The [Cp*Rh(9-MH)$(MeOH)_2$](OTf)$_2$·MeOH (**3**) crystals were obtained from methanol/Et_2O solution at –30°C under an inert atmosphere. Crystal data: Mo Kα (λ = 0.71073 Å); T = 130 K; space group $P2_1/n$; Z = 4; a = 16.962(6) Å, b = 9.059(2) Å, c = 21.346(7) Å, β = 105.53-(2)°; 0° > 2θ > 50°; 4004 observed reflections ($F > 4.0\sigma(F)$); R = 0.0523 and R_w = 0.0522. The structure was solved by the Patterson method using SHELXTL PLUS. The hydrogens bonded to the oxygens were located in a difference map and refined with a fixed isotropic U value of 0.040 Å^2, a fixed 0.90-Å O–H distance, and idealized geometry. No hydrogen was located for the oxygen of the methanol of solvation.

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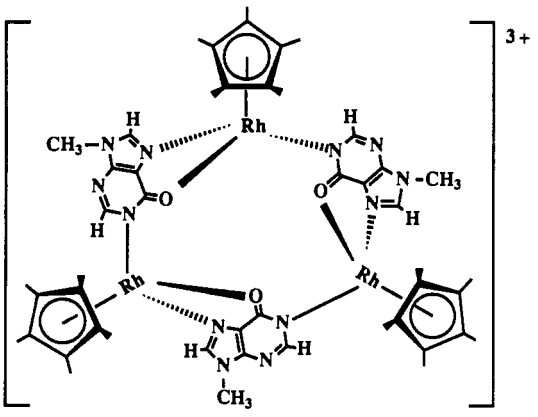
solvents,[1i,l,7] while hydrogen bonding of the carbonyl functionality of guanine sites in DNA to the amino ligands in *cis*-platin constitutes a directional influence and may assist in the binding specificity of *cis*-platin to guanine sites in DNA.[8]

In a recently published paper, we reported that the reaction of the Cp*Rh aqua complex with 1-methylcytosine (1-MC) in water provided the trans-$[Cp^*Rh(\eta^1(N3)\text{-}1\text{-}MC)(\mu\text{-}OH)]_2^{2+}$ complex, while in acetone the mononuclear complex, $[Cp^*Rh(\eta^1(N3)\text{-}MC)(\eta^2(O2,N3)\text{-}1\text{-}MC)](OTf)_2$, was isolated.[1b] These results were a clear demonstration that solvent dictated structure via the presence of either a monomeric $[Cp^*Rh(S)_3]^{2+}$ (S = acetone or methanol) or dimeric $[Cp^*Rh(H_2O)_2(OTf)_2]_x$ aqua-starting complex.

Therefore, to further investigate the effect of an aqueous reaction media, as a function of pH, an ^1H NMR study was performed with **2** and **3**, since it would also allow us to determine the steric role, if any, of the NH_2 group at C2 (**2** only, H8) and the bonding mode of NH1 (**3** only, H8, H2). The pH profile of **3** dissolved in D_2O is recorded in Figure 2 (supplementary material).[8] At pD 2.45–5.13, the downfield chemical shifts for both H8 (8.48 ppm, $\Delta\delta$ = 0.44 ppm) and H2 (8.35 ppm, $\Delta\delta$ = 0.17 ppm; selective H8 exchange in refluxing D_2O verified these assignments) compared to free 9-MH (H8, 8.04 ppm and H2, 8.18 ppm)[8] are consistent with exclusive N7 binding. However, at pD 6.45, the Cp*Rh-9-MH complex provides the dramatic chemical shifts we have found to be diagnostic for cyclic trimer formation,especially for H8 (downfield shift) and H2 (upfield shift) at 8.60 (H8, $\Delta\delta$ = 0.56 ppm) and 7.78 ppm (H2, $\Delta\delta$ = 0.40 ppm) as well at 3.73 ppm (9-CH_3, $\Delta\delta$ = 0.09 ppm), and 1.84 ppm (Cp*) and strongly suggests the presence of the unusual and unprecedented Cp*Rh-guanine nucleobase derivative structure $[Cp^*Rh\text{-}\mu\text{-}\eta^1(N1)\text{:}(\eta^2(O6,\ N7)\text{-}9\text{-methylhypoxanthyl})]_3^{3+}$ (**4**), analogous to similar structures found for adenine derivatives, 9-methyladenine, adenosine, and methyl-5′- and 3′-adenosine monophosphates.[1a,c] Unfortunately, all attempts to isolate **4** at pD 6.45 for further characterization were not successful.



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Complex **4** would result from η^2(N7, O6) 5-membered ring formation followed by the facile deprotonation of NH1 to form the third coordination site to an adjacent $[Cp^*Rh\text{-}\eta^2(N7,\ O6)$ 9-methylhypoxanthyl)$]^+$ moiety, since the pK_a of this site would be substantially reduced upon N7 coordination to the electrophilic Cp*Rh metal center in comparison to the pK_a of the free ligand (9.5).[9] As well, the Cp*Rh–N1 bond formation would further be driven by a condensation reaction between this NH1 site and a reactive Cp*Rh hydroxy species from the adjacent $[Cp^*Rh\text{-}\eta^2(N7, O6)$ 9-methylhypoxanthyl)$]^+$ moiety; it is also conceivable that **4** has an amide enolate tautomeric, cyclic trimer structure with N7, N=C6—O⁻, and OC=N1 bonding.

In addition, at pD 7.96 (Figure 2, supplementary material) we observe some free 9-MH, which suggests a possible equilibrium between free and bound 9-MH. This equilibrium is further supported by a titration experiment, where increasing concentrations of the Cp*Rh aqua complex are added to 9-MH at pD 7.0 to show line broadening of the 9-MH resonances. The presence of a possible $[Cp^*Rh(\mu\text{-}OH)]_2$ dimer species, analogous to that recently found for 1-methylcytosine,[1b] above pD 8 is also suggested by the upfield shifted resonances at 7.96 (H2), 7.89 (H8), 3.76 (9-CH_3), and 1.65 ppm (Cp*), but at this time we are still trying to isolate this complex for possible X-ray structural elucidation

A similar pH profile with 2 shows no discernable cyclic trimer formation. This conclusion is based on no prominent H8 signals in the 8.5–8.8 region and no upfield chemical shifts for the methylene protons of the ethyl group, as well as no downfield shift for the Cp*Rh signal; these ^1H NMR shifts were diagnostic for other cyclic trimer structures such as **3**, as well as those for 9-methyladenine,[1a] adenosine[1a], and methyl-5′- and 3′-adenosine monophosphates.[1c] This result suggests that the NH_2 group at C2 on the guanine nucleus, whose steric and electronic effects have not been previously well defined during the metal coordination process,[8] *plays a significant steric role*, in this instance, in preventing cyclic trimer formation; steric rather than electronic due to NH1 pKa similarities for 9-EG and 9-MH.

It is evident from this study and our recently published 1-methylcytosine results[1b] that a profound difference in Cp*Rh–nucleobase structure occurs depending on the reaction media (including the important role of the pH) and, therefore, the starting Cp*Rh complex. As well, the important find that the carbonyl at C6 and the NH1 position interaction with the Cp*Rh metal center are linked to cyclic trimer formation only for the guanine nucleobase derivative, 9-MH, in aqueous solution at intermediate pH, and this suggests a stabilizing effect and new roles for these ligand sites compared to other metals.[7,8] These novel results also imply a newly found dominant steric role for the exocyclic NH_2 group that inhibits cyclic trimer formation. Future studies will be concerned with the selective binding of the Cp*Rh aqua complex to sequence-specific oligonucleotides and its role in tethering a single λ-DNA molecule to glass and electrode surfaces.

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Supplementary Material Available: Text detailing the synthesis of **2** and **3**, a figure showing the structure of **3**, tables of crystal data, atomic coordinates and isotropic displacement coefficients, bond lengths, bond angles, anisotropic displacement coefficients, and H-atom coordinates for **3**, and Figure 2, the ^1H NMR pH profile of **3** (12 pages). Ordering information is given on any current masthead page.

(7) A recent report by Dunbar et al. [Dunbar, K. R.; Matonic, J. H.; Saharan, V. P.; Crawford, C; Chirstou, G. 205th National Meeting of the American Chemical Society; Denver, CO, March 28–April 2, 1993; Abstract INOR 484] reveals a new binding mode of 9-ethylguanine and guanosine to $Rh_2(OAc)_4$ that shows a bridging N7–O6 dimer complex.

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