Inorganica Chimica Acta, *105* (1985) 209–212

Methylation of Platinum Complexes by Methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide. Kinetics and Mechanism

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Received March 30, 1985

Abstract

The facile methyl-transfer reaction of methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide with platinum complexes requires platinum in both oxidation states (*i.e.* $PtCl_4^{2-}$ and $PtCl_6^{2-}$). The reaction occurs with ~1:1 stoichiometry between $PtCl_6^{2-}$ and methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide. The sole corrinoid product is $H_2O–B_{12}^+$ in its base-off form. Carbon-13 NMR is used to show that 85% of the methyl-transfer products are $(CH_3)PtCl_5^{2-}$, and 15% are ethane. Kinetic and spectrophotometric studies show that $PtCl_4^{2-}$ complexes with the methylcorrinoid prior to the rate-limiting methyl-transfer step, with $k = 0.88\ s^{-1}$ and $K = 330\ M^{-1}$ (23 °C, 1.0 M Cl^-). Detailed mechanism for the Co–C bond cleavage is discussed.

Introduction

Methylation of platinum complexes by methylcobalamin has been extensively investigated [1–4]. These studies revealed several interesting features, including complexation of Pt(II) with the corrinoid, followed by a reversible reaction of this complex with Pt(IV) to form a trinuclear intermediate, and a 'redox switch' between the Pt(II) and Pt(IV) complexes prior to the methyl-transfer step [3]. However, a crucial question, *i.e.* the nature of Co–C bond cleavage, remained largely unanswered although kinetic data suggested that a one-electron transfer from methylcobalamin to the bound platinum complexes may trigger the Co–C bond cleavage [3c].

In this report we present kinetic and mechanistic data for the demethylation of methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide by $PtCl_4^{2-}$ and $PtCl_6^{2-}$ complexes. Use of this corrinoid provides kinetic data that previously had been unobtainable. More importantly, examination of methyl-transfer products of this system provides nonkinetic evidence for the electron-transfer mechanism.

Experimental

Materials

The $Na_2PtCl_6 \cdot 6H_2O$ and K_2PtCl_4 complexes were purchased from D. F. Goldsmith Inc., and were recrystallized from aqueous solution. Methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide was a generous gift from H.P.C. Hogenkamp [5]. This corrinoid appears only in the base-off form, and hereafter will be referred to as 'CH_3-cba'. Concentrations of the corrinoid solutions were determined by their absorption spectra with the published molar absorptivities [6]. All other chemicals were reagent grade and were used as received.

Stoichiometry and Products

The consumption ratio of $PtCl_6^{2-}$:CH_3-cba was determined spectrophotometrically by titrating a solution containing 3.0×10^{-5} M CH_3-cba and 3.0×10^{-4} M $PtCl_4^{2-}$ with $PtCl_6^{2-}$ solutions. The corrinoid products were examined with a Cary Model 15 spectrophotometer. Pulse Fourier transformed ^{13}C (75.46 MHz) NMR spectra of the methyl-transfer products of $^{13}CH_3$-cba (90% enriched) were obtained at 4 °C with a Nicolet 300 spectrometer locked to the resonance of internal D_2O. The spectra were obtained under conditions of simultaneous proton noise decoupling. Chemical shifts were measured with respect to external TSP. In a typical experiment, 3.0 mM $PtCl_6^{2-}$ was added to a mixture of 3.0 mM $^{13}CH_3$-cba and 30 mM $PtCl_4^{2-}$ under a dim light. The spectra were recorded as soon as the reaction was complete.

Kinetic Measurements

The reaction rates were measured at 350 nm (γ-band of aquocorrinoid) with Cary 15 spectrophotometer at 23 ± 0.3 °C under a dim light. All the rate measurements were performed under pseudo-first

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0020-1693/85/$3.30

order conditions in methylcorrinoid. A typical CH_3-cba concentration was 3.0×10^{-5} M. Ionic strength was maintained at 1.0 M with NaCl. The HCl concentrations were in a range 0.10 to 0.50 M. Plots of log $(A_\infty - A_t)$ *vs.* time gave straight lines for 3 half-lives. Reproducibility was generally better than 8%.

Results

The Na_2PtCl_6 complex is inert to hydrolysis in 1.0 M chloride solution. Hydrolysis of $PtCl_4^{2-}$ in 1.0 M NaCl acidic solution is slow enough to be unimportant [7]. The $PtCl_4^{2-}$ alone did not demethylate CH_3-cba. The $PtCl_6^{2-}$ complex alone demethylated CH_3-cba very slowly. Addition of CH_3-cba to a solution containing both $PtCl_4^{2-}$ and $PtCl_6^{2-}$ complexes resulted in a rapid demethylation of CH_3-cba to H_2O-cba$^+$ (in its base-off form [6]), as shown by the spectral change of the corrinoid. Neither the electronic spectral change nor the demethylation rate is affected by the absence of oxygen. Spectrophotometric titration showed that 0.90 ± 0.05 moles of Na_2PtCl_6 were consumed per mole of CH_3-cba.

The ^{13}C NMR spectra of the methyl-transfer products of $^{13}CH_3$-cba in the absence of exogenous chloride ions are shown in Fig. 1*. The main resonance appears at 3.29 ppm, with a spin-spin coupling constant of 463 Hz obtained from the outer peaks. The relative intensities of the peaks are 1:4:1. This species has been assigned as $(CH_3)PtCl_5^{2-}$ [3b]. This compound is stable in the dark. However, it decomposes to CH_3Cl and $PtCl_4^{2-}$ with the addition of exogenous NaCl [3b]. In addition to (CH_3)-$PtCl_5^{2-}$, the $PtCl_4^{2-}/PtCl_6^{2-}$ demethylation of CH_3-cba produces a species with the ^{13}C resonance appearing at 4.18 ppm (*ca.* 15%). It is important to note that the NMR examination was carried out at 4 °C with the sample tube well sealed.

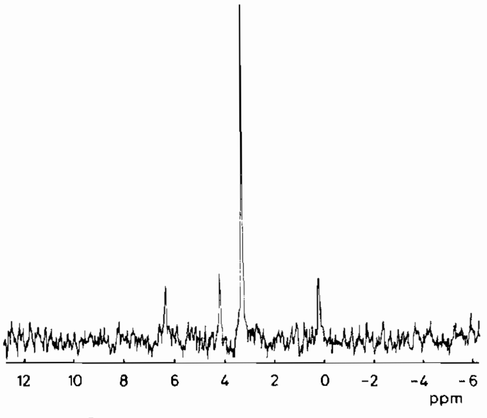

Fig. 1. ^{13}C NMR spectrum of reaction of $^{13}CH_3$-cba (90% enriched) with $PtCl_4^{2-}/PtCl_6^{2-}$ couple (4 °C).

We have previously demonstrated that the demethylation of methylcobalamin by $PtCl_4^{2-}/PtCl_6^{2-}$ couple regenerate $PtCl_4^{2-}$ as one of the platinum products. This was done by using ^{195}Pt NMR [3b]. Adopting this result, the main reaction (*ca.* 85%) between CH_3-cba and $PtCl_4^{2-}/PtCl_6^{2-}$ couple can be described by eqn. 1:

$$CH_3\text{-cba} + \overset{*}{\mathrm{Pt}}Cl_4^{2-} + PtCl_6^{2-} + H_2O \longrightarrow$$
$$H_2O\text{-cba}^+ + (CH_3)\overset{*}{\mathrm{Pt}}Cl_5^{2-} + PtCl_4^{2-} + Cl^- \qquad (1)$$

Kinetic Measurements

Demethylation of CH_3-cba by $PtCl_4^{2-}/PtCl_6^{2-}$ couple is first-order in CH_3-cba in accord with eqn. 2:

$$\frac{-d[CH_3\text{-cba}]}{dt} = \frac{d[H_2O\text{-cba}^+]}{dt} = k_{obs}[CH_3\text{-cba}] \qquad (2)$$

The reaction is first-order in $PtCl_6^{2-}$, as demonstrated in Fig. 2. The reaction is first-order in $PtCl_4^{2-}$ at relatively low concentration, but decreases to zero-order as $[PtCl_4^{2-}]$ is increased. Plots of $[PtCl_6^{2-}]/k_{obs}$ *vs.* $[PtCl_4^{2-}]^{-1}$ are shown in Fig. 3. The overall reaction can be described by eqn. 3:

$$k_{obs} = \frac{k \times K[PtCl_4^{2-}][PtCl_6^{2-}]}{1 + K[PtCl_4^{2-}]} \qquad (3)$$

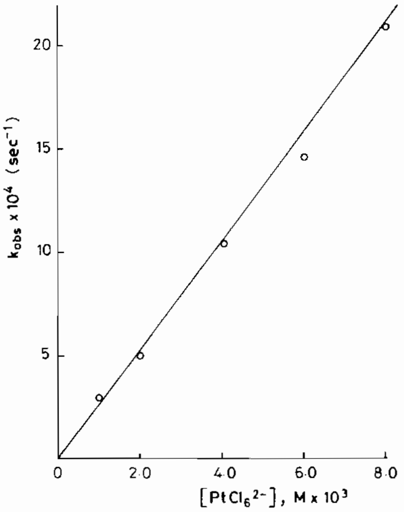

Fig. 2. Plots of k_{obs} *vs.* $[PtCl_6^{2-}]$ at a constant $[PtCl_4^{2-}]$ (0.0010 M).

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*Carbon-13 resonance of $^{13}CH_3$-cba appears at 0.039 ppm (ext. TSP).

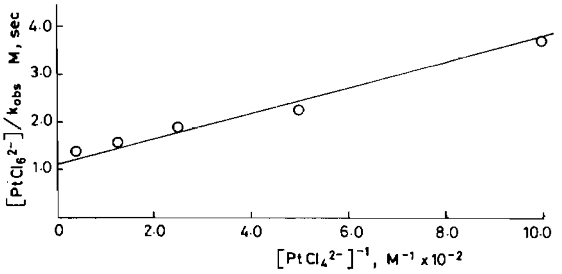

Fig. 3. Plots of $[\mathrm{PtCl_6}^{2-}]/k_{\mathrm{obs}}$ *vs.* $[\mathrm{PtCl_4}^{2-}]^{-1}$.

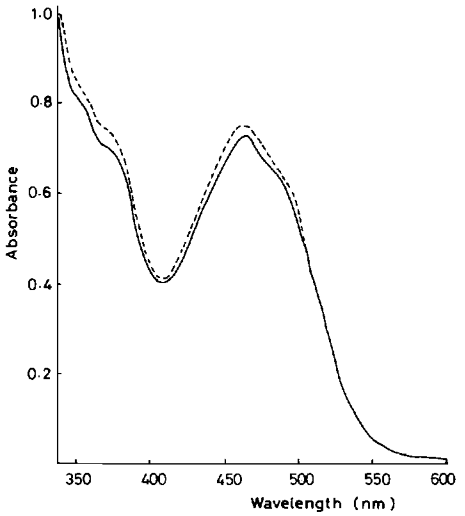

Fig. 4. Effect of $\mathrm{PtCl_4}^{2-}$ on the visible spectrum of $\mathrm{CH_3}$-cba; (———); $\mathrm{CH_3}$-cba alone (6.0×10^{-5} M). (------): $\mathrm{CH_3}$-cba (6.0×10^{-5} M) and $\mathrm{PtCl_4}^{2-}$ (2.0×10^{-2} M).

On the basis of the reaction stoichiometry and kinetic behavior, and because of the similar reactions between base-on methylcobalamin and $\mathrm{PtL_4}^{2-}$/$\mathrm{XPtL'_4y}^{2-}$ couples [3c], eqn. 3 can be described within the framework of reactions 4 and 5:

$$\mathrm{CH_3\text{-}cba} + \mathrm{PtCl_4}^{2-} \overset{K}{\rightleftharpoons} \mathrm{CH_3\text{-}cba, PtCl_4}^{2-} \tag{4}$$

$$\mathrm{CH_3\text{-}cba, PtCl_4}^{2-} + \mathrm{PtCl_6}^{2-} \xrightarrow{k} \text{products} \tag{5}$$

Analysis of Figs. 2 and 3 yields $k = 0.88 \pm 0.18\ \mathrm{s^{-1}}$ and $K = (3.3 \pm 0.37) \times 10^2\ \mathrm{M^{-1}}$. The solid lines in Figs. 2 and 3 were generated by a non-linear least squares fit in accord with eqn. 3. The values of k and K are independent of $[\mathrm{H^+}]$.

Spectral Study of the Interaction between $\mathrm{CH_3}$-cba and $\mathrm{PtCl_4}^{2-}$

The visible spectra of $\mathrm{CH_3}$-cba (6.0×10^{-5} M) without and with $\mathrm{PtCl_4}^{2-}$ are shown in Fig. 4. A prominent shoulder appears in the region from 360 to 380 nm. The spectral change is reversible, and occurs rapidly. However, we did not have an accurate measurement for the formation constant from the spectrophotometric titration.

Discussion

In our previous papers [3] the reactions of methylcobalamin with mixtures of Pt(II) ($\mathrm{Pt(CN)_4}^{2-}$ or $\mathrm{PtCl_4}^{2-}$) and Pt(IV) ($\mathrm{PtCl_6}^{2-}$, $\mathrm{Pt(CN)_4Cl_2}^{2-}$, or $\mathrm{Pt(CN)_5Cl}^{2-}$) are described by the Scheme:

$$\mathrm{CH_3\text{-}B_{12}} + \mathrm{PtL_4}^{2-} \overset{K}{\rightleftharpoons} \underset{\mathbf{A}}{\mathrm{CH_3\text{-}B_{12}, PtL_4}^{2-}} \tag{6}$$

$$\mathbf{A} + \mathrm{XPtL'_4y}^{2-} \overset{K'}{\rightleftharpoons} \underset{\mathbf{B}}{\mathrm{CH_3\text{-}B_{12}, PtL_4\text{–}X\text{–}PtL'_4}^{3-}} + \mathrm{y}^- \tag{7}$$

$$\mathbf{B} + \mathrm{H_2O} \xrightarrow{k'} \mathrm{H_2O\text{-}B_{12}^+} + \mathrm{(CH_3)PtL_4X}^{2-} + \mathrm{PtL'_4}^{2-} \tag{8}$$

This scheme also applies to the methyl-transfer reaction between methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide and $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ couple, with $k = K'k'$. We had previously been unable to obtain the values of k and K of the reaction between base-off methylcobalamin and $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ [3c]. This is because of the very great difference in activities between base-on and the protonated base-off forms of methylcobalamin. Thus, the reaction was dominated by the base-on form of methylcobalamin even at high level of HCl [3a]. Since methylaquo-(3,5,6-trimethylbenzimidazolyl)cobamide appears only in the base-off form, it provides us the opportunity to examine the activity of base-off $\mathrm{CH_3\text{-}B_{12}}$ with $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ couple. The present report allows us to obtain $k = 0.88\ \mathrm{s^{-1}}$ and $K = 330\ \mathrm{M^{-1}}$ (1.0 M $\mathrm{Cl^-}$, 23 °C) for the demethylation of base-off $\mathrm{CH_3\text{-}B_{12}}$ by $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ couple. The K value is close to the formation constant between the base-off $\mathrm{CH_3\text{-}B_{12}}$ and $\mathrm{Pt(CN)_4}^{2-}$ (230 $\mathrm{M^{-1}}$ at 1.0 M $\mathrm{Cl^-}$, 23 °C) [3c]. A striking difference between reactions of methylcobalamins with $\mathrm{Pt(CN)_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$, and with $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ is that the k values for the base-on and base-off $\mathrm{CH_3\text{-}B_{12}}$ are virtually identical in the $\mathrm{Pt(CN)_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ system, while base-on $\mathrm{CH_3\text{-}B_{12}}$ has a k value of a factor of 125 larger than that of base-off $\mathrm{CH_3\text{-}B_{12}}$ in the $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ system.

In the methyl-transfer reactions between methylcobalamin and $\mathrm{Pt(CN)_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ couple, we have used the lack of axial ligand effect, *i.e.* 5,6-dimethylbenzimidazole *vs.* $\mathrm{H_2O}$, on the k values as a support for the electron-transfer methyl-transfer mechanism. The present study indicates that the k values are not similar for the base-on and base-off $\mathrm{CH_3\text{-}B_{12}}$ in the $\mathrm{PtCl_4}^{2-}$/$\mathrm{PtCl_6}^{2-}$ system. Can this observation be reconciled with an electron-transfer mechanism?

One explanation is that the factor of 125 for the k values between the base-on and base-off CH_3-B_{12} in the $PtCl_4^{2-}/PtCl_6^{2-}$ system simply reflects the very great difference in K' values (reaction 7). Thus, the k' values which represent the rate-limiting steps, are similar for the base-on and base-off forms of CH_3-B_{12}. Although this interpretation seems to be reasonable, the present study nevertheless casts doubt on the application of kinetic data to assert electron-transfer mechanism for the CH_3-B_{12} methylation of platinum complexes.

The NMR examination of the methyl-transfer products, however, has revealed valuable information that had not been shown before. By using methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide, 15% of the ^{13}C resonance appears at 4.18 ppm. This species apparently is ethane. The product ethane could have arisen only via radical reaction:

$$\mathrm{CH_3\text{-}cba} \overset{-e}{\rightleftharpoons} \mathrm{CH_3\text{-}cba^+} \longrightarrow \mathrm{H_2O\text{-}cba^+} + \mathrm{CH_3\cdot} \qquad (9)$$

Thus, the present study provides nonkinetic evidence for the one-electron-transfer mechanism of the methyl-transfer step (reaction 8). The generation of ethane from the oxidative cleavage of CH_3–Co bonds has precedences in the reaction of CH_3-B_{12} with deficient amounts of $IrCl_6^{2-}$ (in the absence of Cl^-) [8], and in the oxidative demethylation of $(CH_3)_2$-Co(DpnH) [9]. Curiously, formaldehyde was not formed. Since the NMR examination of the methyl-transfer products was carried out in the presence of air, one would expect that the methyl radical would react with oxygen to generate formaldehyde, just as the photolysis of methylcobalamin under an aerobic condition [10]. We can offer no explanation for the failure to detect formaldehyde in the present system.

In conclusion, the production of ethane from the reaction of methylaquo(3,5,6-trimethylbenzimidazolyl)cobamide with $PtCl_4^{2-}/PtCl_6^{2-}$ strongly suggests one-electron-transfer from the methylcorrinoid to platinum complexes. However, the present study also indicates that kinetic data are not conclusive in regard to the mechanism of the CH_3-B_{12} methylation of platinum complexes.

Acknowledgements

I thank Professor H. P. C. Hogenkamp for his generosity and kindness. This research was carried out during my postdoctoral period in his laboratory. I also thank Dr. J. J. Pignatello for helpful discussions and D. S. Philson for assistance in NMR experiments.

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