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# Complex Formation of Isocytosine Tautomers with $Pd^{II}$ and $Pt^{II}$

**Deepali Gupta, Markus Huelsekopf, Marta Morell Cerdà, Ralf Ludwig,* and Bernhard Lippert***

*Fachbereich Chemie, Universität Dortmund, 44221 Dortmund, Germany*

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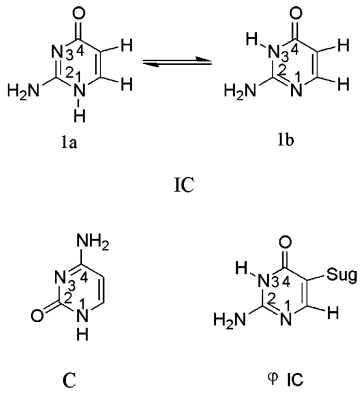
Isocytosine (ICH) exists in solution as two major tautomers, the keto form with N1 carrying a proton (**1a**) and the keto form with N3 being protonated (**1b**). In water, **1a** and **1b** exist in equilibrium with almost equal amounts of both forms present. Reactions with a series of $Pd^{II}$ and $Pt^{II}$ am(m)ine species such as (dien)$Pd^{II}$, (dien)$Pt^{II}$, and *trans*-$(NH_3)_2Pt^{II}$ reveal, however, a distinct preference of these metals for the N3 site, as determined by $^1$H NMR spectroscopy. Individual species have been identified by the pD dependence of the ICH resonances. p$K_a$ values (calculated for $H_2O$) for deprotonation of the individual tautomers complexes are 6.5 and 6.4 for the N3 linkage isomers of dien$Pd^{II}$ and dien$Pt^{II}$, respectively, as well as 6.2 and 6.0 for the N1 linkage isomers. The dimetalated species $[(dienM)_2(IC\text{-}N1,N3)]^{3+}$ (M = $Pd^{II}$ or $Pt^{II}$) are insensitive over a wide range of pD. The crystal structure analysis of $[(dien)Pd(ICH\text{-}N3)](NO_3)_2$ is reported. Ab initio calculations have been performed for tautomer compounds of composition $[(NH_3)_3Pt(ICH)]^{2+}$, *cis*- and *trans*-$[(NH_3)_2PtCl(ICH)]^{+}$, as well as *trans*-$[(NH_3)_2Pt(ICH)_2]^{2+}$. Without exception, N3 linkage isomers are more stable, in agreement with experimental findings. As to the reasons for this binding preference, an NBO (natural bond orbital) analysis for $[(NH_3)_3Pt(ICH\text{-}N3)]^{2+}$strongly suggests that intramolecular hydrogen bonding between trans-positioned $NH_3$ ligands and the two exocyclic groups of the ICH is of prime importance. The calculations furthermore show a marked pyramidalization of the $NH_2$ group of ICH in the complex once the heterocyclic ligand forms a dihedral angle <90° with the Pt coordination plane.

## Introduction

Isocytosine (ICH, 2-aminopyrimidin-4-(3*H*)-one, **1**) is a structural isomer of cytosine (C, **2**) and exists as two major tautomers, **1a** and **1b** (Chart 1). A third, minor tautomer (not shown) adopts an enol structure. Although not a natural nucleobase like cytosine, nucleosides of isocytosine have been reported to exhibit interesting pharmaceutical properties,[1,2] and the C5-glycosidic derivative pseudoisocytidine ($\psi$IC, **3**) has proven a valuable tool for molecular biology studies, e.g. for DNA triplex formation[3] and mechanistic studies of RNA catalysis.[4] Moreover, the potential usefulness of isocytosine as a H bonding partner of the likewise non-natural isoguanine in oligonucleotides has been a point of interest.[5−13] Our focus on isocytosine originates from its

**Chart 1**




potential metal binding properties and our general interest in metal complexes of ligands existing as different tau-

* Authors to whom correspondence should be addressed. E-mail: bernhard.lippert@uni-dortmund.de (B.L.).

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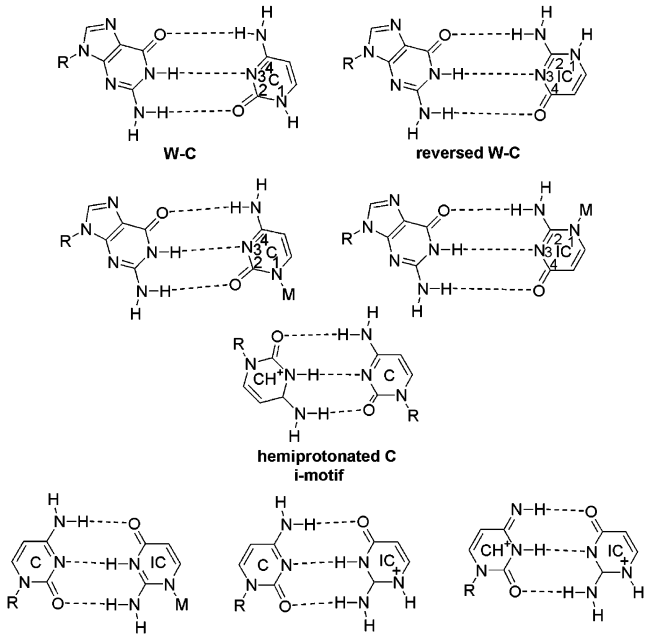
tomers.[14] Metal complex formation with isocytosine has previously been studied by other groups,[15] and there are examples of X-ray structurally characterized metal compounds with O4[15c] or N3[15d] coordination. Various substituted isocytosine derivatives (substituents at N1, N2, C5 positions) have likewise been studied.[16]

At the outset of our work, we were intrigued by the potential of isocytosine to form inert metal complexes of tautomer **1b**, hence metal compounds with N1 coordination. We had previously observed that with cytosine the minor tautomer could be complexed by metal ions to an extent that exceeded its occurrence in solution by far.[14a,b] Moreover, examples of both linkage isomers (metal at N1 or N3) were isolated on a preparative scale and characterized by X-ray crystal structure analysis. In its deprotonated form, the isocytosine tautomer **1a** provides, in principle, the ability of recognizing guanine nucleobases in a way different from that of cytosine, namely, in a reversed Watson−Crick manner (Chart 2, top). Similarly, a N1 metalated isocytosine anion can interact with guanine in this fashion, unlike N1 metalated cytosinate, which pairs according to Watson−Crick.[17] These considerations can be extended to pairing motifs between cytosine and neutral, protonated, or metalated isocytosine (Chart 2, bottom) and contrasted with the situation in hemiprotonated cytosine pairs ("i-motif").[18] Our interest in these systems relates to our continuing efforts to synthesize artificial oligonucleotide analogues consisting of a backbone that contains suitable metal ions.[17,19]

**Chart 2**




## Experimental Section

**Materials.** Isocytosine (ICH) was purchased from Sigma; $K_2PtCl_4$ and $K_2PdCl_4$ were purchased from Heraeus; 9-MeGH and 9-EtGH were obtained from Chemogen. [(dien)PtI]I,[20] [(dien)PdI]I,[21] *trans*-[$(NH_3)_2PtCl_2$],[22] *cis*-[$(NH_3)_2PtCl_2$],[22] 1-methylcytosine (1-MeC),[23] 1-methylthymine (1-MeT),[24] and the complexes *trans*-[$(NH_3)_2$Pt(9-MeGH-*N7*)Cl]Cl,[25] *trans*-[$(NH_3)_2$Pt(9-EtGH-*N7*)Cl]Cl,[25] *trans*-[$(NH_3)_2$Pt(1-MeC)Cl]Cl,[26] *cis*-[$(NH_3)_2$Pt(1-MeC)Cl]Cl,[27] and *trans*-[$(NH_3)_2$Pt(1-MeT)Cl]Cl[28] were prepared according to the methods given in the literature; [(dien)PdBr]Br was prepared analogously to [(dien)PdI]I.[21] Yellow crystals of [(dien)PdBr]Br were isolated and characterized by X-ray crystallography. Results will be published elsewhere.

**Synthesis.** [(dien)Pd(ICH-*N3*)]$(NO_3)_2$ was prepared as follows: A suspension of [(dien)PdBr]Br (79.8 mg, 0.216 mmol) in water (10 mL) was treated with $AgNO_3$ (73.4 mg, 0.432 mmol) for 5 h at 40 °C with the exclusion of daylight. The solution was then cooled to room temperature, AgI was filtered off, and isocytosine (314 mg, 0.216 mmol) was added. The mixture was again stirred for 5 h at 40 °C and kept for crystallization. Yellow crystals appeared after a few weeks and were later dried in air (yield 20%). Elemental anal. (%) for $C_8H_{18}N_8O_6Pd\cdot 0.5H_2O$ (437.64): C 21.9, H 4.2, N 25.6. Found: C 21.7, H 4.2, N 26.0. No water was detected in the X-ray crystal structure, however.

[(dienPt)$_2$(IC-*N1*,*N3*)]$(ClO_4)_3$ was prepared as follows: A suspension of [(dien)PtI]I (155.1 mg, 0.1405 mmol) in $D_2O$ (3 mL) was treated with $AgClO_4$ (95.5 mg, 0.281 mmol) for 5 h at 40 °C with the exclusion of daylight. The solution was then cooled to room temperature, AgI was filtered off, and the solution was divided into two parts. Isocytosine (7.65 mg, 0.216 mmol) was added to the first part of the solution. The mixture was again stirred for 5 h at 40 °C. The pD of the solution was later adjusted to 5.6, and 100 $\mu$L of [(dien)Pt$(D_2O)]^{2+}$ solution was added to the mixture solu-

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tion. The progress of the reaction was monitored by $^1$H NMR spectroscopy. This procedure was repeated till only a single set of signals due to [(dienPt)$_2$(IC-*N1*,*N3*)](ClO$_4$)$_3$ was present. Colorless crystals formed in the reaction mixture within 2 days at 3 °C. A single crystal was picked and characterized by X-ray crystallography. Unfortunately, the crystal was of poor quality, although X-ray data confirmed the composition. Elemental anal. (%) for $C_{12}H_{30}N_9O_{13}Pt_2Cl_3 \cdot 2H_2O$ (1040.97): C 13.8, H 3.3, N 12.1. Found: C 13.7, H 3.2, N 12.1.

All the other reactions mentioned in the text were carried out on the NMR scale.

**Computational Details.** The ab initio molecular orbital calculations were performed using the Gaussian 98 package.[29] A combination of Becke's three-parameter hybrid functional[30] with Lee−Yang−Parr's exchange functional[31] was applied for all structures. The structures of the two tautomers were optimized using two different basis sets, LANL2DZ and 6-31+G*. The metals in the Pd and Pt complexes have been described with the LANL2DZ basis set, including effective core potentials, while the 6-31G* basis set has been used for C, N, O, and H atoms. Additionally, the wave functions were analyzed by the natural bond orbital (NBO) method,[32] a standard option of Gaussian 98. The NBO analysis explains the strength of hydrogen bonds in terms of donor−acceptor interactions between doubly occupied lone pair orbitals and unoccupied antibond orbitals. The total energy was calculated as the sum of the electronic energy and the zero-point vibrational energy. Vibrational frequency calculations were carried out to ensure that the stationary points located on the potential energy surfaces by geometry optimization were minima.

**NMR Spectroscopy.** $^1$H NMR spectra were recorded on a Bruker AC200 (200.13 MHz) instrument in $D_2O$ using sodium 3-(trimethylsilyl)propanesulfonate (TSP) as internal reference.

**Vibrational Spectra.** IR spectra (KBr pellets) were recorded on an IFS 28 FT spectrometer and Raman spectra on a Coderg T800 with argon (514.5 nm) or krypton laser (647.1 nm) excitation.

**Elemental Analysis.** Elemental analysis was performed with a Carlo Erba model 1106 Strumentazione element-analyzer.

**X-ray Crystallography.** Crystal data for compound [(dien)Pd(ICH-*N3*)](NO$_3$)$_2$ were collected at 150 K on an Enraf-Nonius-KappaCCD diffractometer[33] using graphite-monochromated Mo Kα radiation ($\lambda$= 0.7107 Å). For data reduction and cell refinement, the programs DENZO and SCALEPACK (Nonius, 2000)[34] were used. The structure was solved by conventional Patterson methods and subsequent Fourier syntheses and refined by full-matrix least squares on $F^2$ using the SHELXTL 5.1 program.[35] All atoms were refined anisotropically. Hydrogen atoms were located in a difference Fourier map and refined isotropically. Crystallographic data and details of refinement are reported in Table 1.

**Table 1.** Crystallographic Data for [(dien)Pd(ICH-*N3*)](NO$_3$)$_2$

| | |
|---|---|
| empirical formula | $C_8H_{18}N_8O_7Pd$ |
| habit, color | block, yellow |
| crystal size (mm$^3$) | 0.32 × 0.10 × 0.06 |
| fw | 444.72 |
| cryst syst | triclinic |
| space group | $P\bar{1}$ |
| $a$ (Å) | 6.6310(13) |
| $b$ (Å) | 10.651(2) |
| $c$ (Å) | 12.214(3) |
| α (deg) | 65.80(3) |
| $\beta$ (deg) | 87.08(3) |
| $\gamma$ (deg) | 74.07(3) |
| $V$ (Å$^3$) | 754.7(3) |
| $Z$ | 2 |
| $\rho_{calcd}$, (g cm$^{-3}$) | 1.957 |
| $T$ (K) | 150(2) |
| radiaton (Å) | Mo Kα (=0.71073) |
| 2$\theta$ range (deg) | 6.4−55.06 |
| $h,k,l$ collected | ±8, -±13, ±15 |
| $\mu$ (mm$^{-1}$) | 1.285 |
| reflns collected | 3463 |
| unique reflns with $\|F_o\| \geq 4\sigma\|F_o\|$ | 2824 |
| refinement | $F_o^2$ |
| solution method | direct methods |
| no. of params refined | 289 |
| $R_1$[a] | 0.0345 |
| $wR_2$[b] | 0.0863 |
| $R_{int}$ | 0.0386 |
| GOF | 1.081 |

[a] $R_1 = \Sigma||F_o| - |F_c||/\Sigma|F_o|$. [b] $wR_2 = [\Sigma[w(F_o^2 - F_c^2)^2]/\Sigma[w(F_o^2)^2]]^{1/2}$.

**p$K_a$ values.** pH* values refer to uncorrected pH meter readings (Metrohm 6321; combination glass electrode) in $D_2O$ solutions. pH* values were adjusted by addition of $DNO_3$ or NaOD solutions; pD values were obtained by adding 0.4 to the pH meter reading. The p$K_a$ values of isocytosine and complexes of isocytosine were determined using pH dependent $^1$H NMR spectroscopic measurements in $D_2O$. p$K_a$ values were evaluated with a Newton−Gauss[36] nonlinear least-squares fit method that deals with the changes in chemical shifts of all nonexchangeable protons with the change in pD of the solution. The obtained acidity constants were then transformed to the values valid for $H_2O$ according to the literature.[37]

## Results and Discussion

**Tautomerism of Isocytosine.** Isocytosine crystallizes in two tautomeric forms: 1,4-dihydro-2-amino-4-oxo-pyrimidine (keto-N1H, **1a**) and 3,4-dihydro-2-amino-4-oxo-pyrimidine (keto-N3H, **1b**), in an exact 1:1 ratio.[38,39] The tautomers are hydrogen bonded to each other in a manner similar to that of the guanine and cytosine pair in DNA.[40]


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A number of electronic spectroscopic studies[41−46] have been carried out to study the tautomerism of isocytosine. These studies reveal that the ratio of the two tautomers present in solution is solvent and temperature dependent. In ethanol and diethyl ether, keto-N3H form **1b** predominates over keto-N1H form **1a**, while in aqueous solution the two forms are present in almost 1:1 ratio.[42,43,45] Temperature dependent solution studies[43] indicate that in aqueous solution, with the rise in temperature, keto-N1H tautomer **1a** becomes more stable as compared to keto-N3H tautomer **1b**. Morita and Nagakura[42] have also determined the energy differences and entropy changes between the two (keto-N1H vs keto-N3H) forms to be 1.3 kcal/mol and 4.7 cal/mol K, respectively, on the basis of the temperature dependence of the absorption spectra of the molecules.

Several quantum-mechanical computational studies[47−49] have been carried out on different tautomers of isocytosine. In the gas phase, the keto-N3H tautomer **1b** is more stable than the keto-N1H tautomer **1a**. From the calculated and experimental data, it was found that in solid argon matrix[47] isocytosine is present as a mixture of both enol and keto-N3H tautomeric forms with the enol form being dominant. Our calculations on two tautomers (keto-N1H and keto-N3H) in the gas phase are in agreement with the calculations carried out earlier.[47,48] The energy difference between keto-N3H form **1b** and the keto-N1H form **1a** using Lanl2dz and 6-31+G* basis sets is −8.4 and −11.0 kcal/mol, respectively.

**Solution Studies of Isocytosine.** Brown and Teitei[45] reported p$K_a$ values of 4.0 and 9.59, which were obtained by means of ultraviolet spectroscopy. In the course of the present work, p$K_a$ values of isocytosine were determined using pH dependent $^1$H NMR spectroscopic measurements. p$K_a$ for the protonated $ICH_2^+$ was found to be 4.07 ± 0.01, and for the neutral ICH 9.47 ± 0.03 (Figure 1). Chart 3 depicts the two acid−base equilibria.

Raman spectra of isocytosine were recorded in the solid state as well as in different solvent mixtures of DMF and $H_2O$. For the solid isocytosine, in the spectral range 400−1700 cm$^{-1}$, there was a series of characteristic twin bands[50] due to complex ring bending (797.2 cm$^{-1}$, 785.4 cm$^{-1}$) and stretching modes of the ring (1231.9 cm$^{-1}$, 1209.6 cm$^{-1}$) for the two tautomers. The band at 797.2 cm$^{-1}$ for the deformation mode can be assigned to the keto-N1H tautomer **1a** of isocytosine.[51] The band at 785.4 cm$^{-1}$ is consequently assigned to the keto-N3H tautomer **1b** of isocytosine. Similarly, the band at 1231.9 cm$^{-1}$ is assigned to the stretching mode of the keto-N1H tautomer **1a** of isocytosine and, correspondingly, the other band at 1209.6 cm$^{-1}$ to the keto-N3H tautomer **1b**. There are also twin bands at 791 and 787 cm$^{-1}$ for isocytosine in pure DMF, but there is only one band observed at 1214 cm$^{-1}$, which is strongly asymmetric, however. In water, there are a broad band at 791 cm$^{-1}$ and another one at 1232 cm$^{-1}$, which possibly is due to two species present, although there is no clear separation of individual peaks. As DMF is successively diluted with water, there is a shift to higher wavenumbers, i.e., from 789 to 791 cm$^{-1}$ and from 1214 to 1232 cm$^{-1}$, yet there is no separation of individual peaks as is the case for isocytosine in the solid state and in pure DMF.

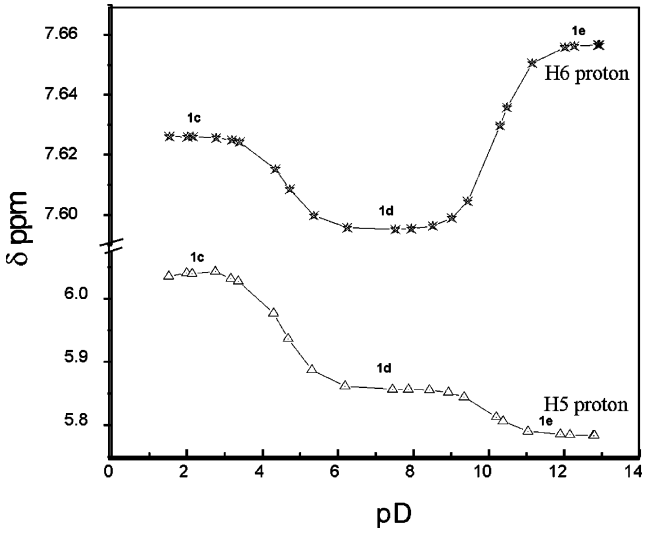
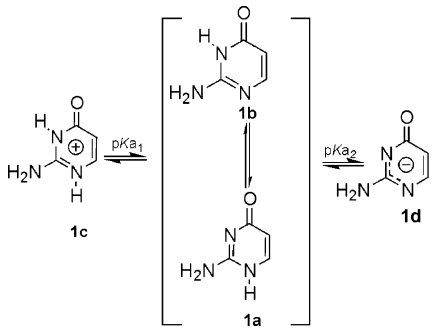



**Figure 1.** pD dependence ($\delta$, ppm) of H5 and H6 resonances of isocytosine in $D_2O$.

**Chart 3**




**Reactions of (dien)Pd$^{II}$ with Isocytosine.** In order to study the metal binding to isocytosine and to identify the various binding sites, the reactions were carried out on the NMR scale. 1:1, 1:3, and 3:1 mixtures of isocytosine and $[(dien)Pd(D_2O)](NO_3)_2$ were prepared, and reactions were carried out without pH* adjustment at the start of the reaction. The samples were kept at 40 °C for a few hours until no further changes were detected in the $^1$H NMR spectra (Figure 2). A typical spectrum consisted of four sets of aromatic H5 and H6 resonances (doublets each, $^3J$ ∼6.7−7.3 Hz). The four sets of resonances A−D were assigned on the basis of pD dependent $^1$H NMR spectroscopic

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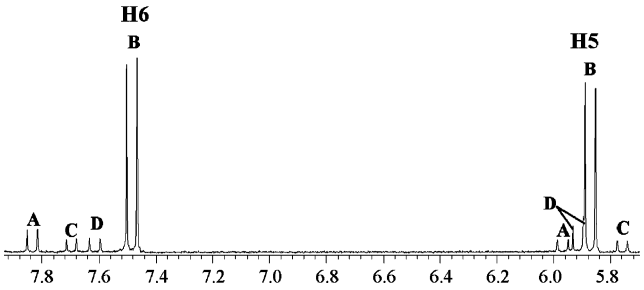


**Figure 2.** $^1$H NMR spectrum ($D_2O$, pH* 3.96, isocytosine resonances only) of reaction mixture (1:1) of $[(dien)Pd(D_2O)]^{2+}$and isocytosine after a few hours at 40 °C. Assignment of resonances A−D was made on the basis of their pD dependence.

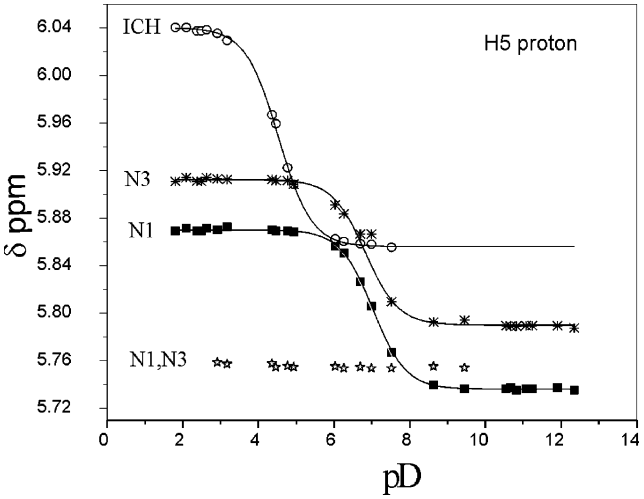


**Figure 3.** pD dependence ($\delta$, ppm) of H5 proton for $[(dien)Pd(ICH\text{-}N1)]^{2+}$, $[(dien)Pd(ICH\text{-}N3)]^{2+}$, $[(dien)Pd(IC\text{-}N1,N3)]^{2+}$, and free isocytosine in $D_2O$.

measurement (Figure 3). Typically, 1:1 reaction mixtures were treated with acid and/or base over a wide pH* range and $^1$H NMR spectra were recorded.

Resonances C are due to the dinuclear species, [(dien)-Pd(IC-*N1*,*N3*)Pd(dien)]$^{3+}$, as this set of resonances is insensitive to pD. An intense set of resonances, B, is assigned to the N3 linkage isomer, $[(dien)Pd(ICH\text{-}N3)]^{2+}$/$[(dien)Pd(IC\text{-}N3)]^{+}$, p$K_a$ 6.5 ± 0.01, by comparison with the spectrum of the isolated compound, the structure of which has been established through X-ray crystallography (see below). The set of resonances furthest downfield, A, is assigned to the N1 linkage isomer, $[(dien)Pd(ICH\text{-}N1)]^{2+}$/$[(dien)Pd(IC\text{-}N1)]^{+}$, p$K_a$ 6.2 ± 0.01. Finally, the set of resonances D belongs to free isocytosine, $ICH_2^+$/ICH, p$K_a$ 4.0 ± 0.01.

**Characterization of [(dien)Pd(ICH-*N3*)](NO$_3$)$_2$.** The reaction of (dien)Pd$^{II}$ with isocytosine was done on a preparative scale, and the solution was kept at room temperature for crystallization. The yellow crystals were isolated, and the characterization of the compound was achieved by X-ray analysis as well as IR, Raman, and NMR spectroscopy.$^1$H NMR spectra were recorded in $D_2O$, and it was found that there was an immediate equilibration of the compound in solution to give four sets of resonances A−D, with set B dominating.

A view of the molecular cation of $[(dien)Pd(ICH\text{-}N3)](NO_3)_2$ is given in Figure 4. Pd is bonded through the N3 position of the isocytosine (Pd−N3, 2.029(3) Å). Distances

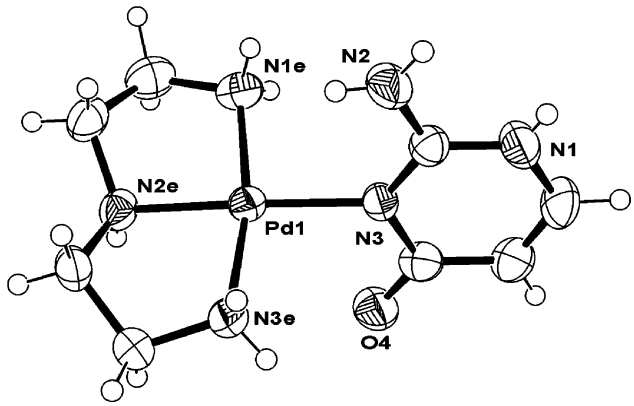


**Figure 4.** View of cation of $[(dien)Pd(ICH\text{-}N3)](NO_3)_2$ with atom numbering scheme.

and angles within the ICH ligand are not unusual. The N1 position is carrying a proton, consistent with the internal ring angle of 121.3(4)° at N1. The Pd−N distances range from 2.002(4) Å (Pd−N2E) to 2.039(4) Å (Pd−N3E) and are likewise normal. Angles about the Pd deviate markedly from ideal square-planar (e.g., N1E−Pd−N2E, 84.83 (14)°; N1E−Pd−N3E, 167.77(16)°), as expected. The dien chelate adopts the characteristic sting-ray geometry[52] with the methylene groups C2E and C3E above the $PdN_4$ plane. The ICH plane forms an angle of 73.2(1)° with the Pd coordination plane. A comparison of the external angles at N3 (Pd−N3−C4, 114.5(3)°; Pd−N3−C2, 125.2(3)°) shows these to be unequal. Consequently, O4 is closer to Pd as compared to N2. N(1)H, N(2)$H_2$, and O4 of the ICH ligand are involved in numerous hydrogen bonding interactions (Supporting Information). For example, the proton at N1 is "chelated" by two oxygen atoms of a nitrate anion, whereas the two protons of N2 form H bonds to oxygens of two different nitrate anions. O4, on the other hand, is bonded to a proton of the amino group N3E of the dien ligand. None of these contacts (2.9−3.1 Å) between heavy atoms is particularly short.

In the IR spectra for isocytosine, tentative band assignments (cm$^{-1}$) according to the literature[53] are $\nu_{sym}$ ($NH_2$) 3275 s, b; $\nu_{asym}$($NH_2$) 3150 s; $\nu$(C=O) 1655 sh; $\nu$(ring) 1388 s; and $\nu$(N−H out of plane) 810 s. Upon Pd$^{II}$ complexation, the bands are shifted: $\nu_{sym}$($NH_2$) 3134 s, b; $\nu_{asym}$($NH_2$) 3406.6 s; $\nu$(C=O) 1679 sh; $\nu$(ring) 1382.8; and $\nu$(N−H out of plane) 823.9 s. The Raman spectrum (solid state) of $[(dien)Pd(ICH\text{-}N3)](NO_3)_2$ has characteristic bands at 797.5 and 1229.6 cm$^{-1}$.

**Reactions of (dien) Pt$^{II}$ with Isocytosine.** Analogous NMR scale reactions were also performed with $[(dien)Pt(D_2O)](NO_3)_2$ and isocytosine. The reaction conditions were similar except that the completion of the reactions took 2 days in the case of Pt. Later, pD dependent $^1$H NMR spectroscopic measurements (Figure 5) were carried out. Depending on the pD of the solution, there were three or four sets of resonances observed. As explained above, resonances A′−D′ were assigned on the basis of their pD

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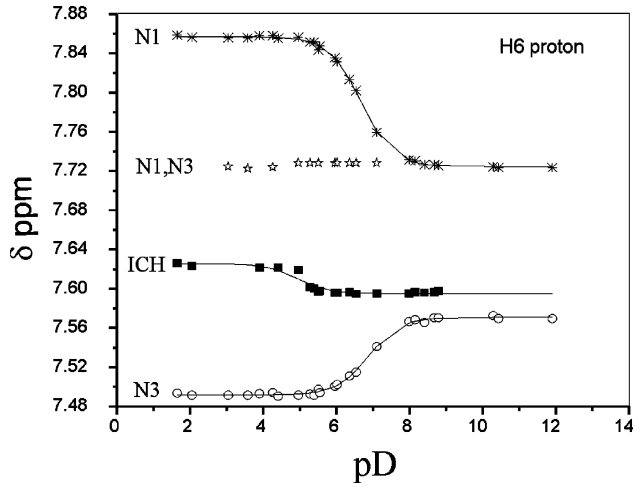


**Figure 5.** pD dependence ($\delta$, ppm) of H6 proton for $[(dien)Pt(ICH\text{-}N1)]^{2+}$, $[(dien)Pt(ICH\text{-}N3)]^{2+}$, $[(dien)Pt(IC\text{-}N1,N3)]^{2+}$, and free isocytosine in $D_2O$.

dependence. The species distribution was similar to that in the $(dien)Pd^{II}$/ICH system. Relevant $pK_a$ values are 6.39 ± 0.03 for $[(dien)Pt(ICH\text{-}N3)]^{2+}$ and 5.97 ± 0.03 for $[(dien)Pt(ICH\text{-}N1)]^{2+}$.

**Reactions of *trans*-$(NH_3)_2Pt^{II}$ with Isocytosine.** In an attempt to obtain metal complexes containing the N1 linkage isomers of isocytosine, several reactions in different ratios Pt:ICH (1:1, 1:3, 3:1, etc.) were performed with *trans*-$[(NH_3)Pt(D_2O)_2]^{2+}$ and isocytosine and followed by $^1H$ NMR spectroscopy. Unfortunately, due to the large number of resonances (ca. 12 sets of doublets) it was not possible to interpret the complicated spectra obtained after the reaction. However, it was clearly evident that there was always a predominance of N3 linkage isomers (H5 resonances in the range 5.6−6.2 ppm) over N1 linkage isomers (H6 resonances, 7.4−8.2 ppm), if chemical shifts of the $(dien)Pt^{II}$ linkage isomers were taken as a reference.

**Other Attempts To Obtain N1 Linkage Isomers.** Attempts to prepare and isolate N1 linkage isomers of isocytosine using different metal species, such as $K_2PdCl_4$, $K_2PtCl_4$, *cis*-$[(NH_3)_2Pt(1\text{-}MeC)Cl]Cl$, *trans*-$[(NH_3)_2Pt(1\text{-}MeC)Cl]Cl$, *trans*-$[(NH_3)_2Pt(9\text{-}RGH\text{-}N7)Cl]Cl$, and *trans*-$[(NH_3)_2Pt(1\text{-}MeT)Cl]Cl$ (with 1-MeC = 1-methylcytosine, 9-RGH = 9-methyl- or 9-ethylguanine, 1-MeT = 1-methylthymine), or different solvents (ethanol, DMF, acetone) were likewise unsuccessful. It was found that there was always a predominance of N3 linkage isomers over N1 isomers. N1 linkage isomers were formed only in small amounts.

**N1, N3 Bridged Species.** A dinuclear complex, $[(dienPt)_2(IC\text{-}N1,N3)]^{3+}$ was prepared and isolated in crystalline form as described in the Experimental Section. In the $^1H$ NMR spectrum ($D_2O$, pD 6.7) H5 and H6 doublets of the ICH ligand ($^3J$ ~7 Hz) are observed at 5.78 and 7.77 ppm. The latter displays unresolved $^{195}Pt$ satellites ($^3J$ ~25−30 Hz). The dien resonances occur between 2.83 and 3.27 ppm as a multiplet. Relative signal intensities of these resonances and the aromatic IC protons are consistent with the dinuclear structure. In the Raman spectrum (solid state) IC bands are observed at 801.8 $cm^{-1}$ and 1235.5 $cm^{-1}$.

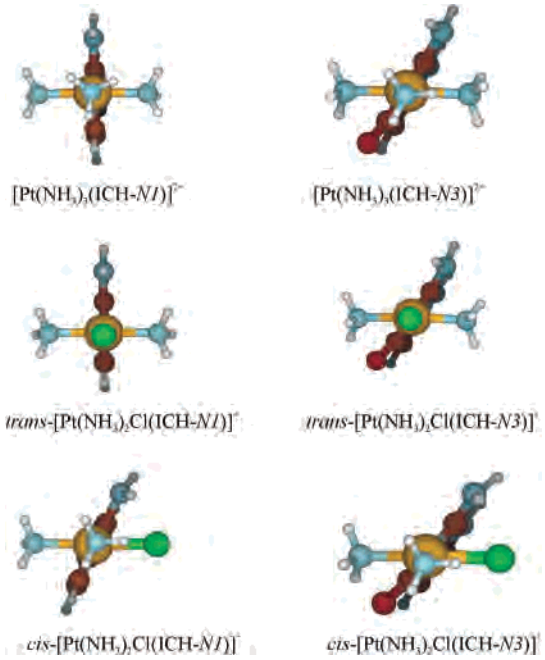


**Figure 6.** Optimized computed structures of N1 and N3 linkage isomers of $[Pt(NH_3)_3(ICH)]^{2+}$, *trans*-$[Pt(NH_3)_2Cl(ICH)]^+$, and *cis*-$[Pt(NH_3)_2Cl(ICH)]^+$.

**Theoretical Calculations.** Molecular geometries of a variety of compounds have been computed in an attempt to better understand the apparent preference of $Pt^{II}$ and $Pd^{II}$ electrophiles for the N3 site of ICH. Figure 6 provides views of optimized structures of the N1 and N3 linkage isomers of $[Pt(NH_3)_3(ICH)]^{2+}$, *trans*-$[Pt(NH_3)_2Cl(ICH)]^+$, and *cis*-$[Pt(NH_3)_2Cl(ICH)]^+$. Calculations of relative energies of N1 and N3 linkage isomers in the case of $(NH_3)_3Pt^{II}$ reveal a preference of the $Pt^{II}$ entity for N3 ($\Delta E$= 12.2 kcal/mol). The comparison between the two geometries shows that in the N1 linkage isomer the heterocycle is perpendicular to the $Pt(NH_3)_3$ plane. The lack of any H bonding between the two trans-positioned $NH_3$ groups at the Pt and the exocyclic amino group of ICH and the inability of O4 to interact with any of the other ligands of Pt is consistent with this finding. On the other hand, in the N3 linkage isomer the isocytosine ring forms an angle of ca. 55° with the $Pt(NH_3)_3$ plane due to the possibility of intramolecular H bonding between O4 of ICH and one of the $NH_3$ ligands of Pt (O···N, 2.79 Å). The computations furthermore suggest weak H bonding between a second $NH_3$ and the exocyclic amino group $N(2)H_2$ (N···N, 3.27 Å), which has undergone some pyramidalization. This finding is consistent with results of recent ab initio calculations which suggest that, contrary to long-time beliefs that nucleobase-$NH_2$ groups are perfectly planar, exocyclic amino groups are indeed partially $sp^3$ hybridized[54] with the ability to function as a H bonding acceptor in nucleic acid structures.[55]

The situation with the two linkage isomers of *trans*-$[Pt(NH_3)_2Cl(ICH)]^+$ closely parallels that of $[(NH_3)_3Pt(ICH)]^{2+}$. In the N1 linkage isomer, the ICH ring is

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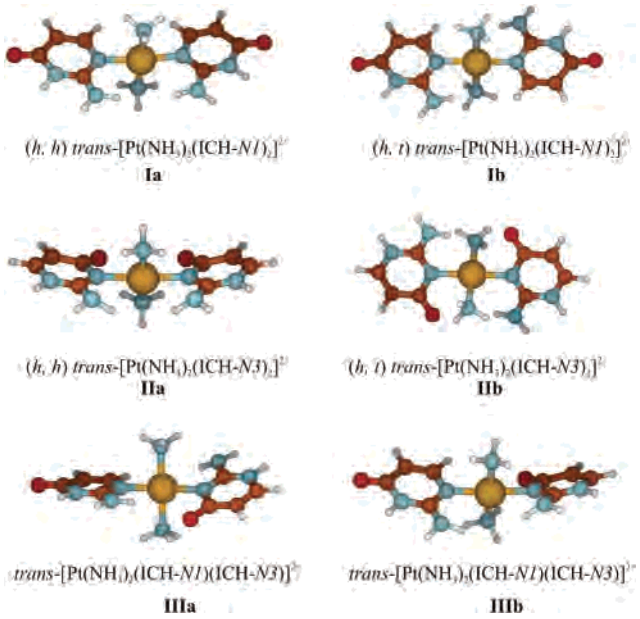


**Figure 7.** Optimized computed structures of the three types of bis(isocytosine) complexes of *trans*-$(NH_3)_2Pt^{II}$ with different rotamers considered.

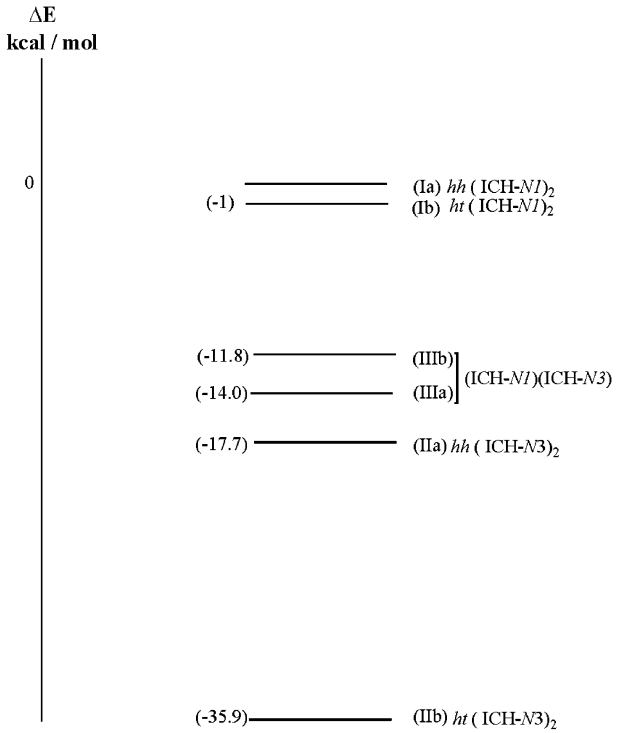


**Figure 8.** Relative energies of the six bis(isocytosine) complexes of *trans*-$(NH_3)_2Pt^{II}$ shown in Figure 7.

perpendicular to the $PtN_2Cl$ plane, while it is tilted (56.9°) in the case of the N3 isomer. The energy difference is 4.2 kcal/mol, again with the N3 linkage isomer being the more stable one.

For the two isomers of *cis*-$[Pt(NH_3)_2Cl(ICH)]^+$, the energy difference is 6.9 kcal/mol, with the N3 linkage isomer being again the more stable one. In both isomers, the ICH plane is tilted with respect to the $PtN_2Cl$ plane by 63.8° (N1 isomer) and 55.7° (N3 isomer). It appears that, at least with the N1 isomer, weak H bonding between $N(2)H_2$ of ICH and the Cl ligand is responsible for this feature. With the N3 linkage isomer, in addition to such a possibility, also H bonding between O4 of ICH and one $NH_3$ ligand (O···N, 2.75 Å) as well as the Cl and one $NH_3$ ligand (Cl···N, 3.01 Å) is possible. It is to be noted that none of the intramolecular hydrogen bonds is to be considered particularly strong, simply because the angles at the acceptor (e.g., Cl) are not very favorable, e.g., Pt−Cl···H, 62.8°, in *cis*-$[Pt(NH_3)_2Cl(ICH\text{-}N3)]^+$ (see, however, below).

An interesting detail of both isomers of *cis*-$[Pt(NH_3)_2Cl(ICH\text{-}N3)]^+$ is the pyramidalization of the amino group of ICH, in that it is opposite to that seen in the *trans*-$[Pt(NH_3)_2Cl(ICH\text{-}N3)]^+$ and $[Pt(NH_3)_3(ICH\text{-}N3)]^{2+}$. It indeed strongly suggests that there is hydrogen bonding between $N(2)H_2$ and Cl.

Finally calculations were performed on a series of bis(isocytosine) complexes of *trans*-$(NH_3)_2Pt^{II}$, which differed in their combinations of binding sites (linkage isomers) and their rotamer forms (Figure 7). As far as relative energies are concerned, the following picture emerged (Figure 8): (i) The *head*−*head* form of *trans*-$[Pt(NH_3)_2(ICH\text{-}N1)_2]^{2+}$ (Ia) is the least stable form, and the *head*−*tail* rotamer of *trans*-$[Pt(NH_3)_2(ICH\text{-}N3)_2]^{2+}$ (IIb) is the most stable one ($\Delta E$= −25.9 kcal/mol). (ii) The *head*−*tail* rotamer (Ib) of *trans*-$[Pt(NH_3)_2(ICH\text{-}N1)_2]^{2+}$ is only very slightly more stable than its *head*−*head* rotamer (−1 kcal/mol), while the second rotamer IIa (*head*−*head*) of *trans*-$[Pt(NH_3)_2(ICH\text{-}N3)_2]^{2+}$ is considerably less stable than its *head*−*tail* rotamer, albeit still the second most stable species (−17.7 kcal/mol). There are two rotamer forms of the mixed complex *trans*-$[Pt(NH_3)_2(ICH\text{-}N3)(ICH\text{-}N1)]^{2+}$ either with O4 of (ICH-*N3*) opposite to $N(2)H_2$ of (ICH-*N1*) (IIIa) or with O4 of (ICH-*N3*) opposite to H6 of (ICH-*N1*) (IIIb). The former is stabilized by 14 kcal/mol relative to the least stable form (Ia), and (IIIb) is more stable by 11.8 kcal/mol. Taken together, these results again confirm the preference of *trans*-$(NH_3)_2Pt^{II}$ for N3 binding to ICH. With regard to the most stable compound, the *head*−*tail* rotamer IIb, it is reminiscent of the *head*−*tail* rotamer of *trans*-$[(NH_3)_2Pt(cytosine\text{-}N3)]^{2+}$, which is likewise more stable than its *head*−*head* form.[56] Although internucleobase H bonding is not possible because of the large separation between opposite exocyclic O4 and $N(2)H_2$ groups, there is a possibility for H bonding between $NH_3$ ligands and the O4 groups (2.84 Å), leading to a tilting of the bases with respect to the Pt coordination plane (56.9°), very much as in the minimum energy structure of the bis(cytosine) compound (60°).[56] With N1 Pt coordination, no such H bonding is possible.

As to the reasons for the preference of the N3 site of ICH for Pt and Pd am(m)ine complexes, we considered a favorable orientation of the dipole moment of one of the two tautomers (**1a**, Chart 1) as a possible explanation, very much as previously considered for the preferential binding of metal ions to N7 of guanine nucleobases.[57] However, the results of these calculations (not shown) were inconsistent

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with the experimental findings and the energy calculations. NBO[32] calculations were subsequently carried out which showed that in the N3 linkage isomer of $[Pt(NH_3)_3(ICH\text{-}N3)]^{2+}$ not only were hydrogen bonds between the $NH_3$ ligand and O4 of ICH formed but, in addition, hydrogen bonding took place between a second $NH_3$ ligand and the substantially pyramidalized exocyclic amino group of ICH. In fact, both hydrogen bonds stabilize the minimum structure in the same order of magnitude. NBO delocalization energies $\Delta E^{(2)}\ n_O \rightarrow o^*_{NH}$ and $\Delta E^{(2)}\ n_N \rightarrow o^*_{NH}$ were obtained by starting out from the minimum structure and by varying the dihedral angle between the ICH plane and the $Pt(NH_3)_3$ plane between 64° and 89°. The decreasing delocalization energies can be readily correlated to decreasing hydrogen bond strengths and decreasing total energies of the structures. This suggests that the observed preference of the $(NH_3)_3Pt^{II}$ entity for N3 of ICH is primarily caused by favorable intramolecular hydrogen bonding. We assume that similar reasons apply to the other complexes. Interestingly, the degree of pyramidalization of the exocyclic amino group is largest when the ICH plane is substantially tilted with respect to the Pt plane, and lost once the heterocycle approaches a perpendicular orientation. We are aware that the inclusion of solvent molecules and the consideration of anions may modify the picture, yet the experimental findings in aqueous solution are consistent with the picture drawn on the basis of the NBO calculations.

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## Summary

Our initial goal of synthesizing, on a preparative scale, isocytosine complexes with N1 bound $Pt^{II}$ was not reached. Rather N3 metal binding is preferred with the metal am(m)ine compounds studied thus far. The results of the calculations tentatively suggest that a fine-tuning of the coligands at the heavy metal may eventually permit the synthesis of the desired compounds. Work toward this goal is in progress.

**Acknowledgment.** This work was supported by the Deutsche Forschungsgemeinschaft (DFG), the Fonds der Chemischen Industrie (FCI), and the International Max Planck Research School Dortmund/Bochum (fellowship for D.G.).

**Supporting Information Available:** An X-ray crystallographic file, in CIF format, for $[(dien)Pd(ICH\text{-}N3)](NO_3)_2$, pD dependencies (H6 of ICH/dien$Pd^{II}$; H5 of ICH/dien$Pt^{II}$), $^1$H NMR spectra (ICH/dien$Pt^{II}$; ICH/*trans*-$(NH_3)_2Pt^{II}$), and computed structures. This information is available free of charge via the Internet at http://pubs.acs.org.

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