

Vibrational Analysis of Heavy Metal-Nucleotide Interactions

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The interaction of Co^{2+} and Pd^{2+} with deoxyguanosine 5'-monophosphate (dGMP) in an aqueous solution at neutral pH with cation/dGMP molar ratios (r) 1/5 and 1 was characterized by FT-Raman spectroscopy. Spectroscopic results show the formation of several types of cation-dGMP complexes. Heavy metal perturbation of the phosphodiester band at 1091 cm^{-1} is higher for Pd^{2+} than for Co^{2+} . The spectral changes observed for the phosphodiester vibrations show that more than 50 % of the backbone geometry is eliminated in Co^{2+} and 100 % in case of Pd^{2+} . In Co-dGMP complex, the glycosyl bond shows more flexibility, whereas in presence of Pd^{2+} ion, the guanine nucleoside exhibits both C2'-endo/anti and C3'-endo/syn conformation. Both the ions have strong affinity for the guanine base and the effect is more pronounced in the presence of Pd^{2+} ions.

Key Words: Vibrational analysis, Heavy metal, Nucleotide.

INTRODUCTION

Studies on interaction of metal ions with nucleosides and nucleotides are very essential because metal ions stabilize the structure of nucleic acids in water. They also control the equilibrium between different forms of secondary and tertiary structures, take part in the processes of DNA transcription and replication. Recent investigations have suggested that some metal ions act as carcinogens in human and animals. Nucleic acids (DNA, RNA) are the main target of recently developed antibacterial and antiviral medicines. Most of these antibiotics bind to DNA by cationic mechanism. Therefore the complexes of nucleosides and nucleotides with metal cations can be applied as model systems in studies on the action of such substances on the DNA structure. The aim of the present work is to obtain information on the binding of heavy metals, particularly Co(II) and Pd(II) to deoxyguanosine 5'-monophosphate by using FT-Raman spectroscopy.

EXPERIMENTAL

Deoxyguanosine monophosphate (dGMP) was purchased from the Sigma Chemicals Co. and used as supplied. Chloride salts of the metal cations were obtained from Aldrich Chemical and used without further purification.

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Sample preparation: A solution of dGMP (0.1M) was prepared and kept at 4 °C for 24 h to ensure the formation of a homogeneous solution. This solution was then mixed with a solution of the metal salt (*e.g.* palladium and cobalt) to give mixture with the desired metal:dGMP ratio(1:5 and 1:1). An aliquot of 50 μ L was sealed in glass capillary tubes for Raman analysis.

Experimental technique: FT-Raman spectra of the dGMP samples presented in the present work were measured with an IFS FT-IR spectrometer coupled to a FRA 106 FT-Raman accessory (Bruker). Raman scattering was excited by a 1.06 μ m continuous-wave ND: YAG laser using approximately 180 mW of radiant power at the sample. The FT-Raman accessory, used with a near-IR beam splitter, collected stokes data over a range of 1750-600 cm^{-1} . Data collection and processing were carried out with data system CS-42 (Bruker).

RESULTS AND DISCUSSION

The FT-Raman spectrum of aqueous dGMP and that in the presence of Co(II) and Pd(II) at 1:5 and 1:1 metal:dGMP ratios are given in Figs. 1-5, respectively. Conformationally sensitive Raman bands are discussed and the results are summarized in Table-1 along with the assignments.

TABLE-I

dGMP	Frequency(Intensity) cm^{-1}				Assignments
	Co ²⁺ /dGMP (r = 1/5)	Co ²⁺ /dGMP (r = 1)	Pd ²⁺ /dGMP (r = 1/5)	Pd ²⁺ /dGMP (r = 1)	
629.4(6.7)	–	–	644.1(8.8)	626.47(7.8)	dG
676.5(5.55)	682.2(9.39)	685.3(9.15)	682.4(8.8)	688.40(8.8)	$\gamma(\text{C1}'\text{-N9})$
803.7(5.35)	812.3(6.3)	–	–	–	$\gamma_s(\text{-C-O-P-O-C-})$
–	–	819.8(8.87)	–	813.70(8.31)	B-backbone
844.1(3.63)	841.6(4.2)	–	829.41(5.43)	–	Sugar phosphate (Shoulder)
886.3(10.0)	886.2(10.0)	886.9(10.0)	887.9(10.0)	888.00(10.0)	$\gamma_s(\text{PO}_2^-)$
1091.5(6.06)	1093.4(8.33)	1096.7(8.3)	1092.7(7.06)	1102.2(7.53)	dG(ring stretching)
1333.7(2.42)	1348.3(2.5)	1350.0(4.22)	1330.2(2.82)	1330.3(3.37)	dG imidazole ring mode vibration
1373.6(2.22)	1388.2(3.02)	1388.2(4.50)	1374.5(3.04)	1373.8(4.28)	dG $\gamma(\text{N7-C8})$
1495.0(4.94)	1488.2(6.81)	1499.9(4.92)	1489.4(4.15)	1493.6(5.97)	dG $\gamma_s(\text{N7-C8})$
1578(4.04)	1571.6(6.66)	1564.7(3.5)	1569(2.7)	1575(4.15)	dG(N1-H, N2H ₂)
1605.8(3.23)	1597.8(5.45)	1591.8(5.35)	1593(3.9)	1593.8(5.71)	$\delta(\text{N1-H})$
1633.8(4.34)	1630.0(6.21)	1626.5(6.6)	1628.1(4.34)	1617.5(5.32)	dG[$\delta(\text{N1-H})\delta(\text{N2H}_2)\gamma_s(\text{C6=O})$]
1647(4.04)	1647.3(6.66)	1641.4(6.76)	1647.3(4.02)	1644.0(5.71)	$\delta(\text{N2H}_2)$
1680.2(3.43)	–	1688.2(4.91)	1658.8(4.02)	1658.8(5.97)	dG[$\delta(\text{N1-H}, \text{N2H}_2)\gamma_s(\text{C6=O})$]

DG = deoxyguanine, gas (O-P-O) = phosphodiester antisymmetrical stretching mode, $\gamma_s(\text{PO}_2^-)$ = phosphodioxo symmetrical stretching mode, $\delta(\text{N1-H})$ = imino in-plane bending mode, $\delta(\text{N2H}_2)$ = amino scissoring mode, $\gamma_s(\text{C6=O})$ = stretching vibrations of carbonyl groups, $\gamma(\text{N7-C8})$ = stretching vibration of (N7-C8)

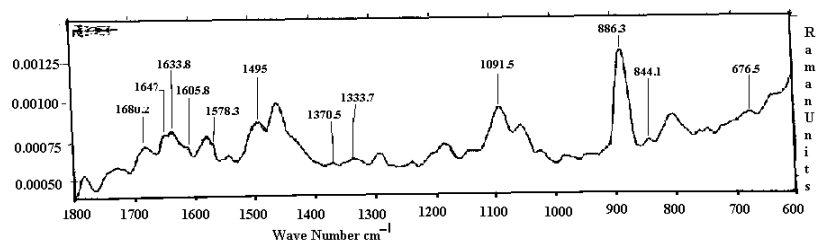
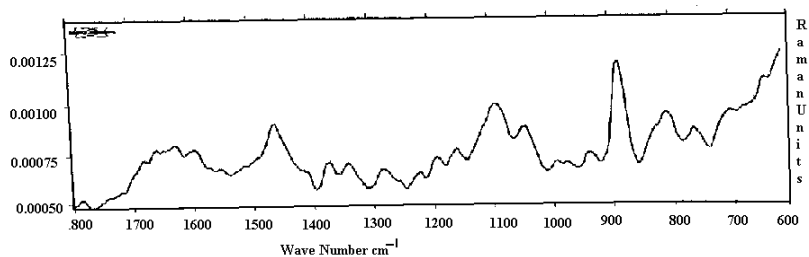
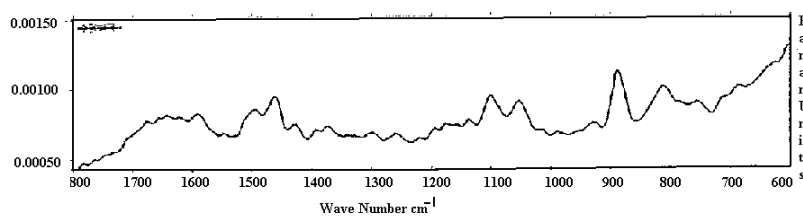
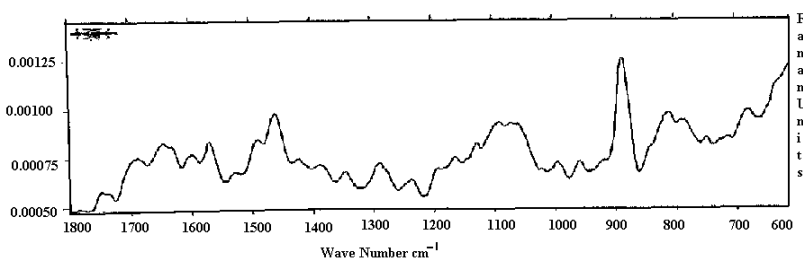
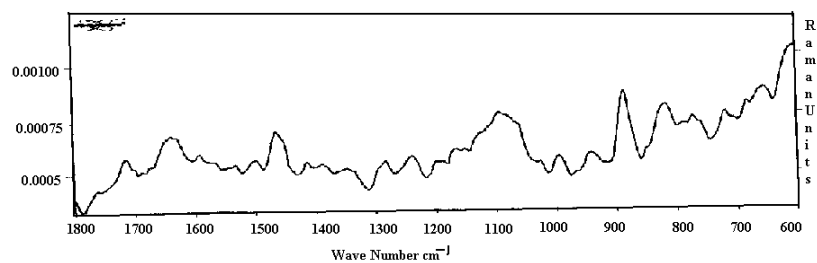


Fig. 1. FT-Raman spectrum of free dGMP

Fig. 2. FT-Raman spectrum of Pd/dGMP ($r = 1/5$)Fig. 3. FT-Raman spectrum of Pd/dGMP ($r = 1$)Fig. 2. FT-Raman spectrum of Co/dGMP ($r = 1/5$)Fig. 2. FT-Raman spectrum of Co/dGMP ($r = 1$)

Raman bands in the region $800\text{-}600\text{ cm}^{-1}$ arise due to C-C and C-N bonds of the guanine base residues such ring breathing vibrations invariably give often the most intense Raman bands in the FT-Raman spectrum of dGMP in aqueous solution¹.

A band observed at 676 cm^{-1} in the free dGMP is assigned to a breathing motion of the guanine coupled through C1'-N9 glycoside bond to deoxyribose vibration. The band also characterizes the C2'-*endo/anti*-conformation of B-DNA in aqueous solution².

In the presence of Pd²⁺ ions at $r = 1/5$ and $r = 1$, the above band shifts, respectively to 682 cm^{-1} with enhanced intensity. Similar effect is observed in the presence of Co²⁺ ions. This result indicates that in the presence of Pd(II) and Co(II) ions the glycosyl bond becomes more flexible. In addition to the above two band, the Pd:dGMP complex shows band at 644 and 626 cm^{-1} , respectively at $r = 1/5$ and $r = 1$. This result indicates that guanine nucleoside exhibits both C2'-*endo/anti* and C3'-*endo/syn* conformation in the presence of Pd²⁺ ions.

The band observed at 844 cm^{-1} in free dGMP is due to antisymmetrical phosphodiester vibration³⁻⁵. This band is considered as diagnostic of B-conformation^{3,6,7} and is perturbed in the presence of both Co²⁺ and Pd²⁺ ions. Nearly, 50 % of the B-backbone is eliminated in Co-dGMP whereas it is 100 % in the case of Pd-dGMP complexes⁸⁻¹⁰.

The 1091 cm^{-1} band of metal free dGMP is assigned to symmetrical vibrations of PO₂⁻ groups^{3,4,6,11}. In the presence of Pd²⁺ ions at $r = 1/5$ and $r = 1$, the above band is shifted to 1092 and 1102 cm^{-1} and show increased intensity. Similar result is observed in Co-dGMP complex where the effect is less pronounced. These results indicate non-specific electrostatic interaction of metal ions with the PO₂⁻ groups.

In the region $1600\text{-}1180\text{ cm}^{-1}$ many bands exhibits shift in frequency with intensity change in the presence of both Pd²⁺ and Co²⁺ ions at the metal ion concentration increases. Most of these bands are determined by vibrations due to in-plane ring stretching vibration of guanine^{1,12-16} and include bands near 1333 cm^{-1} , 1375 cm^{-1} in free dGMP which is due to a vibration involving a large displacement of N7 and C8 atoms of guanine base. In the presence of Pd²⁺ ions the above band exhibits lower frequency shift and intensity increase. The above result indicates metal binding at N7 atom of guanine *via* water molecule which increases the strength of the hydrogen bond between the N7 of guanine and hydrogen of water causing a decrease in frequency of the 1495 cm^{-1} band in Pd-dGMP complex. Similar effect is observed for these bands at 1333 , 1373 and 1495 cm^{-1} . On the other hand in the presence of Co²⁺ ions all the above bands except the band at 1578 cm^{-1} exhibit higher frequency shifts as the metal ion concentration increases. The higher frequency shift observed for the bands at 1333 , 1373 and 1495 cm^{-1} are constant with direct metal binding at N7 atom of guanine.

The region $1750\text{-}1600\text{ cm}^{-1}$ in Raman spectrum is often called the "double-bond region" contains bands due to stretching vibrations of carbonyl double bonds. The bands in this region also include contribution from in-plane deformation of

NH groups (amino and imino). In the present study all the vibrations (Table-1) of free dGMP exhibit lower frequency shift with enhanced intensity in the presence of Pd²⁺ ions for both the concentrations. These results are consistent with strengthening of the hydrogen bonds associated with carbonyl groups and weakening of the hydrogen bonds associated with N1-H and N2-H₂ groups of guanine base. Similar results are observed for in-plane deformation vibration of the N-H groups (both amino and imino) in the presence of Co²⁺ ions. On the other hand, the higher frequency shift observed for the 1680 cm⁻¹ of metal free dGMP in the presence of Co²⁺ ions indicates metal binding at C6=O in Co-dGMP complex.

Conclusion

The following conclusions are derived from the present study of the FT-Raman spectra of the solutions of dGMP in the presence of Pd(II) and Co(II) ions.

(1) The B-conformation of the backbone of dGMP remains essentially unchanged in both Pd-dGMP and Co-dGMP complexes.

(2) In Pd-dGMP complex, the guanine nucleosides exhibit both C2'-*endo/anti* and C3'-*endo/syn* conformation.

(3) Both the ions interact with the N7 of guanine and the effect is more pronounced in Co-dGMP complex.

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