Bond Energies and Phosphate-Coordination of Magnesium Hydrate to Pyrimidine Nucleotide 5'-Monophosphates (CMP, UMP, dTMP) and NMR Shielding Tensors

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ABSTRACT:

The structure of model magnesium hydrate pyrimidine nucleotides (CMP, UMP, dTMP) adducts has been studied at the Hartree-Fock level and by using LANL2DZ basis set for magnesium and 6-31g* basis set for other atoms. The basis set superposition error (BSSE) begins to converge for used Method/basis set. A natural bond orbital calculation was carried out at the Hartree-Fock level and 6-31g* basis set of theory to determined donor-acceptor (bonding-antibonding) interactions. The gauge-invariant atomic orbital (GIAO) method and the continuous-set-of-gauge-transformation (CSGT) procedure were employed to calculate isotropic atomic shielding of the nucleotides and magnesium hydrate-pyrimidine nucleotides by using density functional theory at the B3LYP/6-31g** and HF/6-31g** level.

Keywords: *Ab initio Calculation, NMR shielding Tensor, Nucleotide 5'- Monophosphates, Magnesium Complexes.*

INTRODUCTION

Magnesium, a divalent metal ion usually found in relatively high concentrations in vivo, stabilizes both DNA and RNA against thermal denaturation and in many cases is required for these molecules to function properly /1, 2, 3, 4/.

A nucleotide consists of three main subunits, the nucleobase residue (purine or pyrimidine), the sugar part and the phosphate group(s). The structures of the three common pyrimidine-nucleoside 5-monophosphates are shown in Fig 1 /5, 6, 7, 8/.

Nucleotide ligands offer a variety of potential binding sites for metal ions. In addition, these ligand molecules can adopt various conformations and thus exhibit complex steric properties. The whole area of

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metal ion-nucleotide interactions is therefore of interest also from the point of general coordination chemistry. Main-group metal ions apparently bind only to the phosphates groups of nucleotides, whereas transition metal ions bind to the organic base as well /9, 10/, forming chelate complexes. The structurally characterized metal complexes of mononucleotides involve phosphate monoesters that carry two negative charges per phosphate group, in the normal protonation state consequently the role of the phosphate groups is either through direct metal binding or through electrostatic interaction with the metal. In solution, structures, of metal-nucleotide complexes have been studied mainly by NMR methods /1, 2/.

Fig. 1: Chemical structures of the pyrimidine-nucleoside 5'-monophosphates (UMP, dTMP, CMP).

Nuclear magnetic resonance techniques applied to poly- or mononucleotides can be directly used to detect the effects caused by the binding of a metal ion on these molecules in aqueous solutions. The magnetic resonance of protons has been most widely used. Depending on the nature of the metal in the complex, it will be convenient to measure one or another NMR parameter /15/. Moreover, the main NMR spectroscopic parameters are dependent on the second-order molecular property tensors /16/. The NMR shielding tensor σ can be all written as second derivatives of the energy $\sigma = (\partial^2 E / \partial B \partial m)_{B=0, m=0}$, where E is the energy of the molecule, B external magnetic field, E electric intensity, and n, m nuclear magnetic moments. Scalar parameter and σ observable for an isotropic medium are defined as 1/3 of the traces of the tensors J and σ , respectively /13/.

All of the optimized structure calculations were carried out at the HF/6-31g* level. Electronic structures

were analyzed with the natural bond order method (NBO). In the NBO method, for optimally transforming a given wave function into localized form, corresponding to the one-center (lone pair) and two-center (bond) elements of the chemist's Lewis structure picture. This is carried out by examining all possible interactions between filled (donor) Lewis type NBOs and empty (acceptor) non Lewis NBOs and estimating their energetic importance by two second-order perturbation theory. Since these interactions lead to loss of occupancy from the localized NBOs of the idealized Lewis structure into the empty non-Lewis orbitals (and thus, to departures from the idealized Lewis structure description), they are referred to as delocalization corrections to the zeroth-order natural Lewis structure.

In this study, structures of magnesium hydrate complexes of pyrimidine nucleotide 5'-monophosphate have been investigated by using *ab initio* quantum-chemical calculation at the Hartree-Fock (HF) theoretical level with 6-31g* basis set for predicted polarization effects of the magnesium hydrate at the phosphate oxygen atom coordination. LANL2DZ for Mg and pyrimidine nucleotide 5'-monophosphates have been carried out. Hydrogen bonding will be discussed in term of observable properties such as geometry. The structures was supported by comparing the measured ¹H-NMR spectra to the results of *ab initio* gauge-invariant atomic orbital (GIAO) /15/ and continuous-set-of-gauge-transformation (CSGT) /16/ computations of chemical shifts. *Ab initio* calculations were compared using the experimental geometry, based on the structures derived from X-ray diffraction data for nucleotide 5'-monophosphates. The basis set superposition error (BSSE) was computed, through the counterpoise method /17/ implemented in the Gaussian code 98, for the most stable complexes.

NBO analysis is based on a method for optimally transforming a given wave function into localized form, corresponding to the one-center (lone pair) and two center (bond) elements of the chemist's Lewis structure picture /18/.

METHOD

The structures of all the systems were optimized using the framework of the Hartree- Fock and LANL2DZ basis set for magnesium and 6-31g* basis set /19, 20/ for other atoms. A natural bond orbital (NBO /18/ analysis was performed to the second-order perturbative estimates of donor-acceptor (bonding-antibonding) interactions.

NMR analysis was performed using 6-31g** basis set and the B3LYP /20-25/ level. The GIAO /15/ and CSGT /16/ methods were used to calculate the isotropic NMR shielding at the B3LYP/6-31g** and HF/9-31g** of theory.

The interaction energies of the counterparts were estimated as the energy difference between the complex and the isolated components and were corrected for the basis set superposition error (BSSE). The Boys-Bernardi counterpoise method /26/, applied at the magnesium hydrate nucleotide complexes geometry, is used to account for BSSE. According to this method:

$$E_{\text{Corr.}} = E_{\text{Interaction}} + \Delta E_{\text{BSSE}}$$

where E_{Corr} is corrected-interaction energies, and:

$$\Delta E_{\text{BSSE}} = [E^*_{\text{Magnesium Hydrate}} - E_{\text{Magnesium Hydrate(Complex)}}] + [E^*_{\text{Pyrimidine nucleotide}} - E_{\text{Pyrimidine nucleotide(Complex)}}]$$

where E^* indicates that the energy of components at complex geometry is calculated from Methods/Basis set of complex geometry.

RESULTS

The fully optimized molecular structures for conformations of magnesium hydrate-pyrimidine nucleotides at the HF/6-31g*:LANL2DZ is shown in Figure 2 and calculated optimized geometries are listed in Table 1:

Table 1

Optimized bond length (Å) of pyrimidine nucleotide complexes in the 6-31g*:LANL2DZ basis set

Bond Length	Mg(H ₂ O) ₄ CMP	Mg(H ₂ O) ₅ UMP	Mg(H ₂ O) ₅ dTMP
Mg-O(H ₂ O)	2.105, 2.112, 2.068, 2.099	2.138, 2.170, 2.141, 2.149, 2.096	2.161, 2.122, 2.126,
Mg-O(PO ₄)	1.923	1.942	2.120, 2.155
O(PO ₄)---H(H ₂ O)	1.643	1.677, 1.722	1.952
O1'---H(H ₂ O)	1.952	1.981, 2.592	1.632
O7---H(H ₂ O)	1.926		
N3----H(H ₂ O)	2.129		
P-O1	1.530	1.526	1.476
P-O3	1.462	1.462	1.526
P-O2	1.550	1.557	1.565

Hard Lewis acids, including alkali, alkaline earth, and rare earth metal ions, display almost equal affinities to all the nucleoside monophosphates studied, suggesting that coordination takes place at the phosphate dianion without involvement of the base moiety.

Sigel /17/ has shown, by comparing the stabilities of metal ion complexes of nucleoside monophosphates and simple phosphate monoesters, that the base moiety does not play any role in the complexing of CMP, UMP, dTMP. In other words, the complex stabilities are solely determined by the basicity of the phosphate group and the metal ion affinity of the phosphate group.

Interaction with the phosphate group in the magnesium purine nucleotides was suggested to be either a direct one or hydrogen bonding to the 5'-phosphate through a molecule of water /28, 29/.

In the magnesium hydrate complexes of pyrimidine nucleotides the four and five H₂O ligands are coordinated to the magnesium cation, which is then bonded to the oxygen atom of the phosphate group at

distances ranging between 1.912 and 2.150 Å, in agreement with the available experimental results (2.02 Å and 2.11 Å) [30].

Fig. 2: Optimized structures of Mg-nucleotide complexes in the HF/6-31g*:LANL2DZ Indirect (outer-sphere or hydrogen bonding) phosphate coordination probably occur between atoms oxygen phosphate atom and water molecules of Mg coordination.

The conformation of the sugar ring in nucleotides and nucleosides can be examined by using a concept of pseudorotation, which utilizes a quantitative description of puckering and conformation in terms of the maximum torsion angle (τ_m) and the "Phase angle" of pseudorotation (P), which is a function of the interrelationship between the five torsion angles (τ_0 - τ_4) in the nonplanar five-membered ring [31]. The phase

angle, P , and the maximum pucker, τ_m , are calculated with eq 1 and 2 /31/.

$$\tan P = (\tau_4 + \tau_1) - (\tau_3 + \tau_0) / 2\tau_2(\sin 36 + \sin 72) \quad (1)$$

$$\tau_2 = \tau_m \cos P \quad (2)$$

All the possible conformations are grouped into two categories, type N($P=0\pm 90^\circ$) or 3' endo and type S($P=180\pm 90^\circ$) or 2' endo/C1' exo /32-34/. We have theoretically computed the P values for all nucleotides using HF/6-31g* level on the optimized structures. (Table 2) Pictorial representation of the anti and syn conformational ranges, defined by the torsion angle χ_{CN} about the glycosidic bond C1'-N1, is defined in terms of the four nucleoside atoms C2-N1-C1'-O1' /35, 38, 39/

Table 2
Dihedral Angles Complexes at 6-31g* Basis Set

Dihedral Angles	Mg(H ₂ O)4CMP	Mg(H ₂ O)5UMP	Mg(H ₂ O)dTMP
MgOPO5'	83.0	76.7	110.1
PO5'C5'C4	-155.7	-162.6	60.5
O5'C5'C4'C3'(ψ)	164.7	179.1	-88.4
O1'C1'N1C6	-117.0	-110.0	-109.8
O1'C1'N1C2	56.2	66.3	61.4
O5'C5'C4'O1'	48.5	62.6	152.7
C3'C4'O1'C1'(τ_4)	32.2	38.7	70.0
C3'C21'C1'O1'(τ_1)	8.6	2.4	28.1
O1'C4'C3'C2'(τ_3)	24.7	-35.0	11.0
C4'O1'C1'C2'(τ_0)	-26.2	-25.9	-22.4
C4'C3'C2'C1'(τ_2)	10.0	19.8	-23.0

Table 3
P Angles Complexes in HF/6-31g* Level

Complexes	P Angle
Mg(H ₂ O)4CMP	3.0
Mg(H ₂ O)5UMP	1.7
Mg(H ₂ O)5dTMP	-1.5

The results of these calculations showed that metal-nucleotide sugar conformations fall into categories C3' endo, syn.

The computed stabilization energies of the complexes for no-metalated and metalated pyrimidine

nucleotides are compared by HF/6-31g* method (Table 4). Their relative stabilities are markedly influenced by the metalation. Metalation of pyrimidine nucleotides has been found to give more stability than in the nonmetalated molecule.

Table 4

Relative Energies (Hartree) for Mg-Pyrimidine Nucleotide Complexes in Gas Phase with HF/6-31g* Level

Complexes HF/6	31g*
CMP	1450. 764
UMP	1470. 623
dTMP	1434.751
Mg(H ₂ O) ₄ CMP	1954. 752
Mg(H ₂ O) ₅ UMP	2050. 633
Mg(H ₂ O) ₅ dTMP	2014. 823

Table 5 shows the interaction energies of metalated pyrimidine nucleotides. The increase in interaction energies of the metalated pyrimidine nucleotides is due to hydrogen bonding in metalated pyrimidine nucleotides (Table 1). Table 5 shows the value of BSSE and E_{I+BSSE} for the structures. Clearly for the all complexes, values of BSSE are rather small. Therefore, for these cases BSSE is negligible.

Table 5

E_I (Interaction Energies), BSSE and E_{I+BSSE} (Hartree) for Complexes in HF/6-31g* Level

Complexes	E_I	BSSE	E_{I+BSSE}
Mg(H ₂ O) ₄ CMP	-0. 36075	-0. 0188594	-0. 3796106
Mg(H ₂ O) ₅ UMP	-0. 36299	0. 0282317	-0. 3347583
Mg(H ₂ O) ₅ dTMP	-0. 37151	-0. 0111472	-0. 3826603

NBO calculation shows the π -bonding contribution in the base moiety of nucleotides (Table 6). Strong electron delocalization in a best Lewis structure will also show up as donor-acceptor interactions. Table 7 shows the interactions that give the strongest stabilization.

The Lewis NBOs in Table 8 describe percentage of the total density, with the remaining non-Lewis density found primarily in the valence-shell antibonding. Also, analysis of the atomic charges is done by the natural bonding orbital (NBO) method. It was found that the charge on O2 in metalated pyrimidine nucleotides is higher than in nonmetalated pyrimidine nucleotides (O2 coordinated to Mg) (Table 9). As shown above, metalation of the pyrimidine nucleotides strongly influences their electronic structure, and this leads to energetic stabilization of the structures. There for, the increasing O2 basicity of the metalated nucleotides can be attributed to the relative stabilization and increased stability of Mg-nucleotide complexes.

Table 6

Hybridization Coefficient Of Bonds Calculated By NBO Method in HF/6-31g* Level

Bond	Mg(H ₂ O)4CMP	Mg(H ₂ O)5UMP
P-O2	σ 0.4426(SP ^{3.02})P + 0.8967(SP ^{1.77})O2	0.4360(SP ^{3.03})P + 0.8999(SP ^{1.55})O2
C2-O7	σ 0.5865(SP ^{2.03})C2 + 0.8100(SP ^{1.29})O7 π 0.5289(SP ^{1.00})C2 + 0.8487(SP ^{1.00})O7	0.58821(SP ^{1.87})C2+0.8087(SP ^{1.26})O7 0.4907(P ^{1.00})C2 + 0.8713 (P ^{1.00})O7
C4-N3	σ 0.7655(SP ^{1.61})N3+0.6434(SP ^{2.06})C4 π 0.8314(SP ^{1.00})N3+0.55561(SP ^{1.00})C4	
C5-C6	σ 0.7059(SP ^{1.79})C5+0.7084 (SP ^{1.51})C6 π 0.7599(SP ^{1.00})C5+0.6500 (SP ^{1.00})C6	0.7025(SP ^{1.73})C5+0.7117(SP ^{1.45})C6 0.7613(SP ^{1.00})C5+ 0.6484(SP ^{1.00})C6
C4-O8	σ π	0.5865(SP ^{1.93})C4+0.8099(SP ^{1.20})O8 0.5259(SP ^{1.00})C4+ 0.856(SP ^{1.00})O8
Bond	Mg(H ₂ O)5dTMP	
P-O2	σ 0.4452(SP ^{3.12})P+0.8954(SP ^{1.75})O2	
C2-O7	σ 0.5912(SP ^{1.89})C2+0.8065(SP ^{1.30})O7 π 0.5381(SP ^{1.00})C2+0.8429(SP ^{1.00})O7	
C4-N3	σ π	
C5-O6	σ 0.7079(SP ^{1.73})C5+0.7063(SP ^{1.41})O6 π 0.7338(SP ^{1.00})C5+0.6793(SP ^{1.00})O6	
C4-O8	σ 0.5895(SP ^{2.05})C4+0.8078(SP ^{1.25})O8 π 0.5575(SP ^{1.00})C4+0.8302(SP ^{1.00})O8	

Table 7

The Stabilization Energy E(2) Associated with Delocalization for Interactions Giving the Strongest
Stabilization in HF/6-31g* Level

Complex	Donor NBO	Acceptor NBO	E(2)KCal mol ⁻¹
CMP	LP(3)O7	BD*(2)C2-N3	92.97
		BD*(1)C2-O	2.96
		BD*(1)C2'-H	0.41
UMP	LP(3)O8	BD*(1)C4-C5	1.37
		BD*(2)C4-C5	44.85
		BD*(1)C4-O	1.69
dTMP	LP(3)O8	BD*(1)C4-C5	0.26
		BD*(2)C4-C5	84.20
		BD*(1)C4-O	4.51
		BD*(1)N8-C4	0.55

Mg(H ₂ O) ₄ CMP	LP(1)N1	BD*(2) C5-C6	46.87
		BD*(2)C2-O7	53.45
		BD*(1)C2'-C1'	4.50
		BD*(1) C1'-O1'	7.87
Mg(H ₂ O) ₅ UMP	LP(1)N1	BD*(2) C5-C6	52.40
		BD*(2)C2-O7	77.90
		BD*(1)C2'-C1'	5.29
		BD*(1) C1'-O1'	13.8
Mg(H ₂ O) ₅ dTMP	LP(1)N1	BD*(2)C4-C6	37.11
		BD*(2)C2-O7	60.86
		BD*(2)C2'-C1'	3.85
		BD*(1) C1'-O1'	8.83

Table 8

%Total Lewis, Highest Energy Lewis NBO and Lowest Occupancy of Complexes in HF/6-31g* Level

Complexes	% Total Lewis	Lowest occupancy (N1)	Highest energy Lewis NBO (a.u) (N1)
Mg(H ₂ O) ₄ CMP	98.1650	1.58491	-0.30486
Mg(H ₂ O) ₅ UMP	98.2263	1.62492	-0.25863
Mg(H ₂ O) ₅ dTMP	98.5947	1.71447	-0.42866

Table 9

Natural Population of Phosphate Oxygen Atoms and P, Mg in Metalated Pyrimidine Nucleotides
in HF/6-31g* Level

Complexes	O1	O2	O3	P	Mg
Mg(H ₂ O) ₄ CMP	-1.20815	-1.35704	-1.12146	2.58446	1.73320
Mg(H ₂ O) ₅ UMP	-1.31380	-1.43884	-1.23475	2.83431	1.81722
Mg(H ₂ O) ₅ dTMP	-1.16998	-1.35613	-1.19831	2.61300	1.73812

NMR Chemical Shift Assignments

The NMR shielding tensors were calculated using the gauge-including atomic orbital (GIAO) method and CSGT procedure by using B3LYP/6-31g** and HF/6-31g** level /39/. The isotropic part σ_{iso} of σ is measured by taking the average of σ with respect to the orientation to the magnetic field, i. e., $\sigma_{iso} = (\sigma_{11} + \sigma_{22} + \sigma_{33})/3$. The results calculated are summarized in Tables 11-14).

The anisotropy is $\zeta = |\sigma_{33} - \sigma_{iso}|$, and the asymmetry is $\eta = (\sigma_{22} - \sigma_{11})/\zeta$ /40/.

Ab initio calculations yield the data in Tables 11, 13 at B3LYP/6-31g** method, showing that the values for the isotropic shielding O2, O1, O3 atoms in metalated CMP, UMP, dTMP are higher but the isotropic shielding of C6 atom is less than in nonmetalated CMP, UMP, dTMP. On the other hand, absolute isotropic TMS of C6, H6 atoms in the metalated compounds is higher (Tables 12, 14). It was found that the isotropic shielding O8 atom in metalated UMP, dTMP is less than in nonmetalated CMP, UMP, dTMP, but isotropic shielding O7 atom in metalated CMP was increased.

Table 10

Computed absolute isotropic shielding values for TMS

Method	^{13}C	^1H
B3LYP/6-31g** : GIAO	191. 8644	31. 7532
B3LYP/6-31g** : CSGT	193. 0751	29. 3957
HF/6-31g** : GIAO	203. 1548	32. 3360

The results calculated by using HF/6-31g** level at GIAO to predict the NMR shielding tensor of σ_{iso} in Table 15, 16 show that the isotropic shielding of C6, H6 atoms in magnesium hydrate CMP, UMP, dTMP have been increased; on the other hand we have found a higher deshielding of the C6 and H6 to TMS in magnesium hydrate pyrimidine nucleotides than in the pyrimidine nucleotides. Isotropic shielding tensor of N1, N8, N3, O7 atoms in Mg(H₂O)₄CMP and N1, N3, O7, O8 atoms in Mg(H₂O)₅UMP and N1, N3, O3 atoms in Mg(H₂O)₅dTMP have been decreased as compared to the CMP, UMP, dTMP values.

Table 11

B3LYP/6-31g** Calculations of the σ_{iso} in ppm, for Atoms in CMP, UMP, dTMP

Atom	Method	CMP σ_{iso}	UMP σ_{iso}	dTMP σ_{iso}
O2	GIAO	-514. 3770	-513. 0544	-14. 0452
	CSGT	-511. 8219	-510. 5122	-0. 1908
P	GIAO	366. 2695	366. 4302	364. 1990
	CSGT	293. 7501	293. 8301	293. 1711
O1	GIAO	-590. 5471	-587. 5774	-720. 9073
	CSGT	-587. 4588	-584. 3941	-722. 2328
O3	GIAO	15. 5493	12. 7500	-535. 5780
	CSGT	30. 8019	27. 9149	-535. 5553
O5'	GIAO	233. 7576	233. 6205	232. 8569
	CSGT	239. 4151	239. 0236	239. 5009
C5'	GIAO	125. 1186	125. 0175	125. 8444
	CSGT	125. 0595	24. 9494	125. 8703

C4'	GIAO	100. 5310	100. 3551	106. 8920
	CSGT	102. 2623	102. 0066	108. 0079
C3'	GIAO	121. 1567	120. 5738	117. 1316
	CSGT	120. 2270	119. 7958	116. 3437
C2'	GIAO	121. 6867	119. 7113	152. 4803
	CSGT	119. 1631	117. 3912	154. 0507
C1'	GIAO	95. 1572	97. 5965	99. 9643
	CSGT	94. 8540	97. 3514	100. 2233
O1'	GIAO	240. 3374	245. 0119	230. 6627
	CSGT	232. 8270	237. 3346	224. 5652
N1	GIAO	132. 1447	136. 7890	129. 6839
	CSGT	128. 9853	134. 3059	126. 7020
C2	GIAO	39. 9668	45. 6711	45. 8944
	CSGT	34. 8292	39. 1514	39. 9325
O7	GIAO	14. 3922	40. 1028	62. 3734
	CSGT	8. 9886	39. 7758	60. 8320
N3	GIAO	67. 9164	113. 2957	118. 0751
	CSGT	60. 5725	106. 7640	110. 5459
C4	GIAO	48. 8156	45. 0549	49. 0805
	CSGT	44. 0677	39. 2903	42. 6477
C5	GIAO	109. 6685	110. 6851	109. 0991
	CSGT	104. 8438	105. 6880	105. 4772
C6	GIAO	94. 6256	98. 9236	99. 1797
	CSGT	92. 0962	96. 4334	93. 7506
O8	GIAO		46. 4841	79. 9393
	CSGT		41. 6251	71. 4099

Table 12

Relative (to TMS) Shifts in ppm for ^{13}C -NMR and ^1H -NMR of CMP, UMP, dTMP using GIAO and CSGT Method at B3LYP/6-31g**. The NMR Chemical Shift ($\delta = \sigma_{\text{iso}} \text{ TMS} - \sigma_{\text{iso}} \text{ Sample}$).

Atom	Method	CMP	UMP	dTMP
C5'	GIAO	66.7458	66.8469	66.0200
	CSGT	68.0156	68.125	67.2048
C4'	GIAO	91.3334	91.5093	84.9724
	CSGT	90.8128	91.0685	85.0672
C3'	GIAO	70.7077	71.2906	74.7328
	CSGT	72.8481	73.2793	76.7314
C2'	GIAO	70.1777	72.1531	39.3841
	CSGT	73.9120	75.6839	39.0244
C1'	GIAO	96.7072	94.2679	91.9001
	CSGT	98.2211	95.7237	92.8518
C2	GIAO	151.8976	146.1933	145.9700
	CSGT	158.2459	153.9237	153.1426
C4	GIAO	143.0488	146.8095	142.7839
	CSGT	149.0074	153.7843	150.4274
C5	GIAO	82.1959	81.1793	82.7653
	CSGT	88.2313	87.3871	87.5979
C6	GIAO	97.2388	92.9408	92.6847
	CSGT	100.9789	96.6417	99.3245
H6	GIAO	4.1373	3.8745	4.0302
	CSGT	1.914	1.8724	1.9107
H5	GIAO	3.7528	3.2124	
	CSGT	1.363	0.8072	
H1'	GIAO	3.6604	3.7883	4.3442
	CSGT	1.6391	1.8535	2.1017
H5'	GIAO	3.5612	3.5541	3.5052
	CSGT	1.6425	1.6358	1.7646
H5''	GIAO	3.5662	3.5665	3.7272
	CSGT	1.2322	1.1845	1.8797

Table 13

B3LYP/6-31g** Calculations of the σ_{iso} and η in ppm for Atoms in Mg-Nucleotides of CMP, UMP, dTMP

Atom	Method	Mg(H ₂ O)4CMP		Mg(H ₂ O)5UMP		Mg(H ₂ O)5 dTMP	
		σ_{iso}	η	σ_{iso}	η	σ_{iso}	η
O2	GIAO	226.5585	-46.9672	224.1038	3.5428	218.7024	4.3389
	CSGT	245.7534	-2.9947	244.7279	3.6289	236.7522	11.228
P	GIAO	386.6372	-0.5633	385.6625	19.2799	382.4083	3.2854
	CSGT	325.6397	-0.5702	325.3632	-2086.1338	327.6836	3.9113
O1	GIAO	197.3560	2.0219	197.0063	-0.1012	215.0692	4.0250
	CSGT	210.9127	4.7907	212.1973	0.0291	227.2497	1.2191
O3	GIAO	215.5189	-0.3773	219.6031	2.2073	196.0184	-115.6696
	CSGT	232.8156	0.6239	236.2105	1.2732	211.6728	-13.1598
O5'	GIAO	244.2142	-3.5982	240.129	-8.5761	245.8752	1.1966
	CSGT	245.0896	-2.3198	242.0614	-5.2685	250.4434	0.5286
C5'	GIAO	130.4883	-17.5763	131.7728	-7.2811	128.0928	-2.3850
	CSGT	129.0078	11.2064	130.3865	-11.7384	127.9573	-2.2781
C4'	GIAO	108.5125	1.6408	108.3150	1.1493	103.1658	-26.0886
	CSGT	108.7888	1.4643	108.1458	1.1457	104.3985	69.0538
C3'	GIAO	120.0705	0.8833	123.2455	0.9566	118.0323	2.2854
	CSGT	117.9747	0.7957	120.9372	0.82990	117.5202	2.1381
C2'	GIAO	119.9059	-0.0243	119.6933	0.9584	156.9011	-5.9778
	CSGT	117.9733	-0.0026	117.7392	1.1354	157.9196	-7.2084
C1'	GIAO	92.3321	-1.40396	94.8099	-1.2215	97.7546	-6.2002
	CSGT	91.6214	-1.2893	94.4952	-1.0589	97.1814	-5.1565
O1'	GIAO	256.4880	0.4694	250.8207	0.2404	246.4044	-1.4148
	CSGT	250.0488	0.4707	245.8660	0.2418	239.8925	-1.2924
N1	GIAO	101.0952	54.2223	117.2015	-6.0647	117.0489	0.6819
	CSGT	97.8561	-54.9369	113.1909	-5.0327	113.5592	0.5794
C2	GIAO	49.0915	70.7041	53.7386	-2.8224	57.7475	-1.7563
	CSGT	42.8417	-111.0038	47.6462	2.8930	48.5867	-1.7628
O7	GIAO	32.2392	-8.8977	38.3589	-1.6768	40.0617	0.2607
	CSGT	26.8208	-8.5985	36.1594	1.6072	36.5339	0.1914
N3	GIAO	59.0252	-8.7095	108.1047	0.3558	107.1225	-4.1925
	CSGT	52.0032	10.6351	101.8760	0.3222	101.0808	-3.5567
C4	GIAO	38.7826	-5.2799	44.9328	0.4644	43.1285	0.4340
	CSGT	33.8865	-5.4036	39.2421	0.3676	37.5025	0.3913
C5	GIAO	106.1726	0.04226	94.6694	0.7960	86.8756	0.2144
	CSGT	100.4281	-0.0926	88.6740	-0.8128	81.9921	0.1228

C6	GIAO	51. 6989	-0. 8031	56. 1445	-0. 4309	58. 3169	-5. 1678
	CSGT	47. 5190	-0. 8601	51. 8109	-0. 4227	53. 3347	-1. 1616
O8	GIAO			-53. 3664	5. 2413	-35. 2001	0. 4340
	CSGT			-58. 6325	4. 7164	-40. 6903	0. 2002

Table 14

Relative (to TMS) Shifts in ppm for ^{13}C -NMR and ^1H -NMR of CMP, UMP, dTMP using GIAO and CSGT Method at B3LYP/6-31g**. The NMR Chemical Shift ($\delta = \sigma_{\text{iso}} \text{ TMS} - \sigma_{\text{iso}} \text{ Sample}$)

Atoms	Method	Mg(H ₂ O) ₄ CMP	Mg(H ₂ O) ₅ UMP	Mg(H ₂ O) ₅ dTMP
C5'	GIAO	61. 3761	60. 0916	63. 7712
	CSGT	64. 0673	62. 6886	64. 9823
C4'	GIAO	83. 3519	83. 5494	88. 6986
	CSGT	84. 2863	84. 9293	88. 6766
C3'	GIAO	71. 7939	68. 6189	73. 8321
	CSGT	75. 1004	72. 1379	75. 5549
C2'	GIAO	71. 9585	72. 1711	34. 9633
	CSGT	75. 1018	75. 3359	35. 1555
C1'	GIAO	99. 5323	97. 0545	94. 1098
	CSGT	101. 4537	98. 5799	95. 8937
C2	GIAO	142. 7729	138. 1258	134. 1169
	CSGT	143. 9836	145. 4289	144. 4884
C4	GIAO	153. 0818	146. 9316	148. 7359
	CSGT	159. 1886	153. 8330	155. 5726
C5	GIAO	85. 6918	97. 1950	104. 9888
	CSGT	92. 6470	104. 4011	111. 0830
C6	GIAO	140. 1655	135. 7199	133. 5475
	CSGT	145. 5561	141. 2642	139. 7404
H6	GIAO	6. 8675	6. 5400	6. 2692
	CSGT	4. 5447	4. 2703	3. 9418
H5	GIAO	5. 0146	5. 1211	
	CSGT	2. 8509	2. 8774	
H1'	GIAO	4. 2588	4. 1879	4. 8331
	CSGT	2. 0505	1. 9026	2. 7834
H5'	GIAO	3. 7881	3. 8469	3. 1857
	CSGT	2. 0462	2. 0789	1. 7267
H5''	GIAO	3. 6275	3. 3817	3. 4009
	CSGT	1. 7449	1. 7436	1. 5303

Table 15

HF/6-31g** Calculations of the σ_{iso} in ppm for Atoms in Metalated or Nonmetalated CMP, UMP, dTMP
(Charge and Multiplicity -2, 3)

Atom	CMP	Mg(H ₂ O)4CMP	UMP	Mg(H ₂ O)5UMP	dTMP	Mg(H ₂ O)5dTMP
C5'	145. 7379	146. 0515	145. 7585	147. 2284	144. 6637	142. 7135
C4'	125. 0030	126. 0659	124. 2624	125. 3747	116. 5172	122. 3503
C3'	138. 5291	136. 9841	138. 7352	139. 6812	139. 4666	135. 8682
C2'	136. 5700	135. 8006	135. 2260	135. 6797	168. 1820	169. 3802
C1'	114. 3494	112. 2896	115. 4666	114. 7350	125. 1421	117. 5060
C6	17. 7335	48. 3862	22. 5274	59. 4509	-2. 8103	58. 9970
C4	20. 8724	32. 9057	35. 1250	45. 3533	46. 5828	42. 6675
C5	105. 0896	117. 7746	90. 2017	106. 07750	58. 1032	100. 2425
C2	46. 5906	49. 1639	52. 6562	54. 3845	58. 28024	55. 2619
H1'	28. 5188	28. 1328	29. 0910	29. 0370	28. 6257	27. 8413
H6	22. 8798	24. 6581	23. 0883	25. 1749	22. 0930	25. 4905
H5	26. 6215	26. 9912	26. 4179	26. 8249		
H5'	28. 8930	28. 8101	28. 8735	28. 9776	29. 7987	29. 0149
H5"	28. 3595	28. 5846	28. 3058	28. 4569	28. 6257	28. 9690
N8	219. 2447	197. 7547				
N1	168. 1530	128. 8930	170. 4110	147. 8460	159. 7488	148. 1081
N2						
N3	124. 1017	81. 8900	149. 9611	132. 6250	136. 2581	131. 7112
O3	-399. 1824	244. 3538	-3373.0419	248. 1015	252. 1960	227. 5930
O2	26. 1615	255. 0413	263. 5201	253. 7420	245. 0883	250. 5244
O1	279. 8879	230. 3126	279. 8830	229. 6790	256. 7207	244. 6401
O5'	284. 4205	281. 5102	284. 3592	278. 3393	276. 9306	280. 5709
O1'	289. 7209	295. 0279	291. 1317	291. 301	286. 6974	286. 9013
O7	100. 6183	62. 7614	109. 6324	63. 3416	42. 2072	63. 6221
O8			131. 8189	-36. 5383	-50. 8827	-20. 0441
P	421. 4834	422. 8997	421. 8483	422. 4350	415. 2184	421. 3731

Table 16

Relative (to TMS) Shifts in ppm for ^{13}C -NMR and ^1H -NMR of Nucleotides using GIAO Method
at HF/6-31g**. The Chemical Shifts ($\delta = \sigma_{\text{iso TMS}} - \sigma_{\text{iso Sample}}$)

Atom	CMP	Mg(H ₂ O)4CMP	UMP	Mg(H ₂ O)5UMP	dTMP	Mg(H ₂ O)5dTMP
C5'	57. 4169	57. 1033	57. 3963	55. 9264	58. 4911	60. 4413
C4'	78. 1518	77. 0889	78. 8924	77. 7801	86. 6376	80. 8045
C3'	64. 6257	66. 1707	64. 4196	63. 4736	63. 6882	67. 2866
C2'	66. 5848	67. 3542	67. 9288	67. 4751	34. 9728	33. 7746
C1'	88. 8054	90. 8652	87. 6882	88. 4198	78. 0127	85. 6488
C6	185. 4213	154. 7686	180. 6274	146. 7039	205. 9651	144. 1578
C5	98. 0652	85. 3802	112. 9531	97. 0773	145. 0516	102. 9123
C4	182. 2824	170. 2491	168. 0298	157. 8015	198. 5720	160. 4873
C2	156. 5642	153. 9909	150. 4986	148. 7703	144. 8724	147. 8929
H6	9. 4562	7. 6779	9. 2477	7. 1611	10. 2430	6. 8455
H5	5. 7145	5. 3448	5. 9181	5. 5111		
H1'	3. 8172	4. 2032	3. 2450	3. 2990	3. 7103	4. 4947
H5'	3. 443	3. 5259	3. 4625	3. 3584	2. 5373	3. 3211
H5''	3. 9765	3. 7514	4. 0302	3. 8791	3. 7103	3. 3670

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