

# Guanine Nucleotides: Base-Centered and Phosphate-Centered Valence-Bound Radical Anions in Aqueous Solution

Jiande Gu,<sup>\*,†</sup> Yaoming Xie,<sup>‡</sup> and Henry F. Schaefer, III<sup>\*,‡</sup>

Drug Design & Discovery Center, State Key Laboratory of Drug Research, Shanghai Institute of Materia Medica, Shanghai Institutes for Biological Sciences, CAS, Shanghai 201203 P. R. China, Center for Computational Quantum Chemistry, University of Georgia, Athens, Georgia 30602-2525

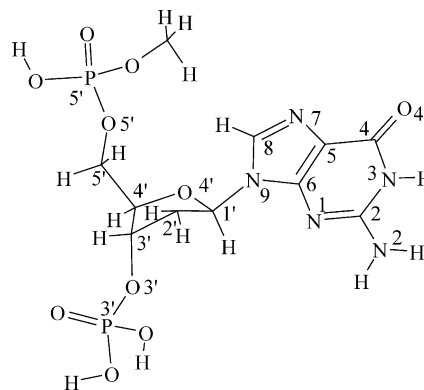
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To explore the nature of electron attachment to the guanine-centered DNA fragments in the presence of a polarizable medium, theoretical investigation of electron attachment to the guanine-related DNA single-strand fragments deoxyguanosine-3'-monophosphate (dGp), deoxyguanosine-5'-monophosphate (pdG), and deoxyguanosine-3',5'-diphosphate (pdGp) were performed using density functional theory with the polarizable continuum model. The electron distributions for the radical anions of pdGp in aqueous solution are extraordinarily different from those in the gas phase. In solution, the excess electron can covalently bind either to the base (forming  $\text{pdG}^{\bullet-\text{p}}$ ) or to the 3'-phosphate in the radical anion (forming  $\text{pdGp}^{\bullet-}$ ). The significant electron detachment energies found for these radical anions suggest that both  $\text{pdG}^{\bullet-\text{p}}$  and  $\text{pdGp}^{\bullet-}$  are electronically stable species in aqueous solution and are expected to be initiators in electron attachment-induced DNA damage in nature. In the presence of the polarizable medium, the base-centered radical anion  $\text{pdG}^{\bullet-\text{p}}$  is more stable than the phosphate-centered structure. By comparison with electron attachment to the monophosphated nucleotide models pdG and dGp, the existence of the phosphate-centered radical  $\text{pdGp}^{\bullet-}$  in pdGp is attributed to the cooperative influence of the two phosphate groups and the polarizable medium.

## Introduction

Electron attachment to DNA subunits is an exciting and important subject among biochemical processes.<sup>1,2</sup> The formation of base-centered or phosphate group-centered radical anions in DNA single strands may be the key step leading to strand breaking.<sup>3–8</sup> Among the four basic nucleobases (adenine A, guanine G, cytosine C, and thymine T), conventional A and conventional G have been found to have negative valence electron affinities (EA) in the gas phase.<sup>9</sup> The presence of the ribose moiety in nucleosides and the sugar–phosphate backbone in nucleotides increases their electron capture ability significantly.<sup>10–12</sup> However, geometrical features and molecular orbital analyses suggest that the excess electron of the anions of guanosine (dG) and its diphosphate compound (pdGp) is only dipole-bound to the molecular frame.<sup>12</sup> Experimental studies on uracil have demonstrated that solvation effects may transform the dipole-bound anions into valence-bound structures.<sup>13–15</sup> Theoretical investigations of nucleotides also suggest that the delocalized electron density of the anions of the purine nucleotides in the gas phase turns out to be well-localized due to the influence of the polarizable continuum medium.<sup>12</sup> In a previous study of the radical anions of nucleotides, the excess electron is found to reside on the phosphate group of pdGp due to solvation effects, and the corresponding electron affinity in aqueous solution is estimated to be 0.95 eV.<sup>12</sup> It is interesting to note that with PCM modeling for the hydration, the AEA of the hydrated G is predicted to be 1.33 eV.<sup>16,17</sup> As a corollary, it is reasonable to expect that there also might be a base-centered radical anion of guanosine diphosphate in aqueous solution.

## SCHEME 1: Model of a Guanine-Related DNA Single Strand: 2'-Deoxyguanosine-3',5'-diphosphate (pdGp)<sup>a</sup>



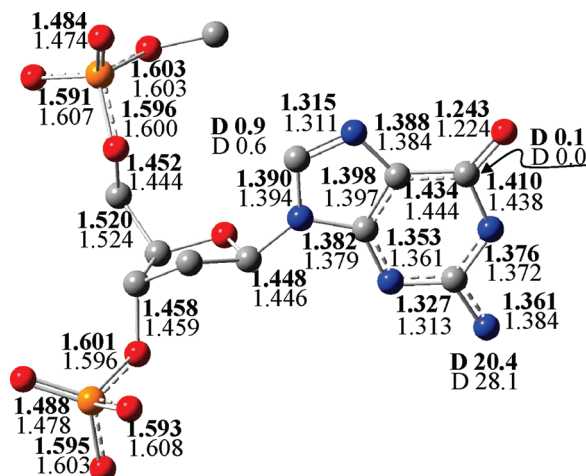
<sup>a</sup> For a better description of the influence of the 3'–5' phosphodiester linkage in DNA, the  $-\text{OPO}_3\text{H}$  moiety at the 5' position was terminated with a methyl group. This termination also prevents the biologically unrealistic proton transfer from  $-\text{OPO}_3\text{HH}$  to the guanine moiety in the radical anion.

Here, we report a theoretical investigation of electron attachment to the guanine-related DNA single-strand fragment deoxyguanosine-3',5'-diphosphate (pdGp; Scheme 1) in aqueous solution. To be consistent with our previous studies, only the conventional form of guanosine was included in this research, although the tautomeric forms of guanine might be important for the formation of the corresponding radical anions. The energetic properties, such as large electron vertical detachment energy (VDE as large as 2 eV), and geometrical features suggest that these covalent radical anions are electronically stable and might serve as precursors to trigger chemical reactions, leading to lesions in DNA and RNA.

\* Corresponding authors. E-mail: (J.G.) jiandegush@go.com, (H.F.S.) sch@uga.edu.

<sup>†</sup> Shanghai Institutes for Biological Sciences.

<sup>‡</sup> University of Georgia.



**Figure 1.** The optimized geometries of pdGp in the gas phase (ordinary print) and in aqueous solution (bold). Bond lengths in angstroms and dihedral angles, *D*, in degrees. Color conventions: gray, carbon; blue, nitrogen; red, oxygen; orange, phosphorus. Hydrogen atoms have been omitted for clarity.

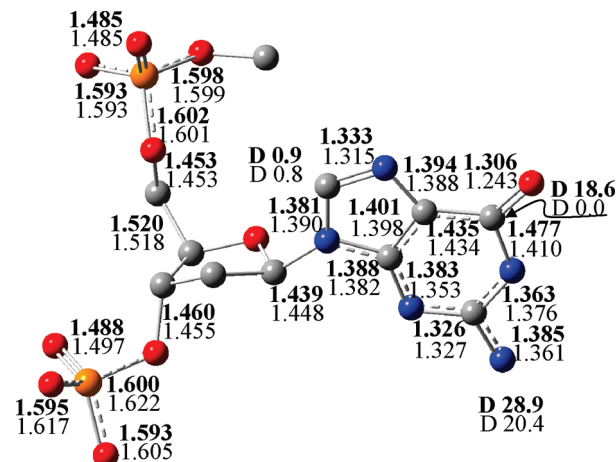
## Results and Discussion

The geometries were fully optimized at the B3LYP/DZP++ level,<sup>18</sup> which has been proven to be a reliable approach for describing the structures and energetics of radicals and anions related to DNA components.<sup>6–10,12</sup> The Barone–Tomasi polarizable continuum model (PCM)<sup>19</sup> with the dielectric constant of water ( $\epsilon = 78.39$ ) was used to simulate the solvated environment of an aqueous solution. The Gaussian03 program<sup>20</sup> was used for all computations.

The geometry of pdGp optimized in aqueous solution (modeled by the PCM) is found to be close to that in the gas phase. However, the presence of the polarizable continuum increases the C4=O4 and P=O double-bond lengths by 0.02 and 0.01 Å, respectively. Meanwhile, the single bond distances C4–C5, N3–C4, and C2–N2 are reduced by 0.01, 0.03, and 0.02 Å, respectively, in aqueous solution. Furthermore, due to the effects of the polarizable continuum, the dihedral angle around the pyramidalized N2 atom of guanine ( $D_{C2HH'N2}$ ) is 20.4°, about 8° less than that in the gas phase. The above geometric variations suggest that conjugation over the guanine frame is improved by the presence of the polarizable continuum.

Both base-centered (denoted as pdG<sup>•−</sup>p) and phosphate-centered radical (pdGp<sup>•−</sup>) anions of pdGp were located as local minima on the aqueous potential energy surface. Compared to the neutral species, noticeable geometric changes are predicted primarily on the guanine moiety for pdG<sup>•−</sup>p. The C4=O4 bond length increases to 1.306 Å in pdG<sup>•−</sup>p, 0.06 Å longer than that for the neutral compound. Other significant bond elongations are found for N3–C4 (1.477 vs 1.410 Å), C2–N2 (1.385 vs 1.361 Å), N1–C6 (1.383 vs 1.353 Å), and N7=C8 (1.333 vs 1.315 Å). Noticeable bond distance *decreases* are found for C2–N3 (1.363 vs 1.376 Å) and C8–N9 (1.381 vs 1.390 Å). It should be mentioned that the glycosidic bond length N9–C1' also decreases by 0.01 Å (1.439 vs 1.448 Å) due to the excess electron residing on the base.

Perhaps most surprisingly, the pyramidalization of the C4 atom of G is detected for the base-centered radical anion. The dihedral angle  $D_{N3C5O4C4}$  is 18.6° in pdG<sup>•−</sup>p, whereas it is essentially zero in pdGp and pdGp<sup>•−</sup>. Considering that this pyramidalization is found only for the C atom next to the glycosidic bond in other nucleotides<sup>6,8</sup> and nucleosides<sup>10</sup> (C8 in A and C6 in T and C), the nonplanar purine ring of pdG<sup>•−</sup>p



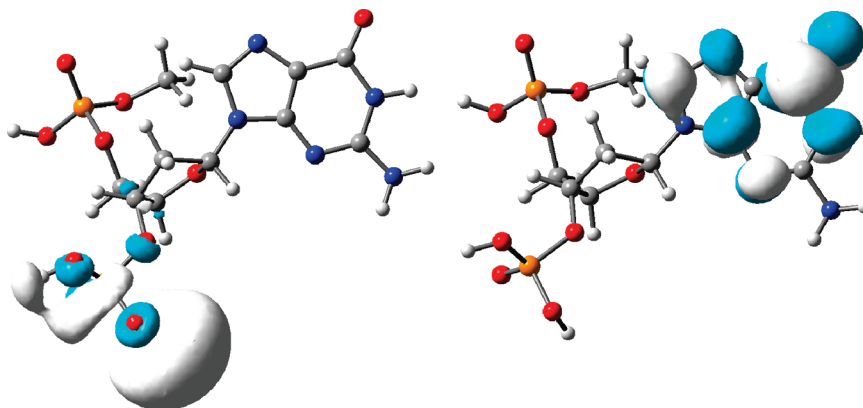
**Figure 2.** Optimized geometries for pdGp<sup>•−</sup> (ordinary print) and pdG<sup>•−</sup>p (bold) in aqueous solution. Bond lengths in angstroms and dihedral angles, *D*, in degrees. Color conventions: gray, carbon; blue, nitrogen; red, oxygen; orange, phosphorus. Hydrogen atoms have been omitted for clarity.

is unique. The nonplanar feature of the amine group of G is also intensified by electron attachment to the base. The dihedral angle  $D_{C2HH'N2}$  is 28.9° in pdG<sup>•−</sup>p, about 8° larger than that for pdGp and pdGp<sup>•−</sup>.

Similar geometric characteristics are also observed in the radical anions of the nucleoside guanosine and the nucleobase guanine. On the other hand, visible geometric alterations can be detected mainly on the phosphate group attached to the 3' position of pdGp<sup>•−</sup>. Specifically, the P=O double bond increases to 1.497 Å in pdGp<sup>•−</sup>, 0.01 Å longer than that in pdGp. The three P–O single bonds also lengthen to 1.605, 1.617, and 1.622 Å, the distance changes amounting to 0.01, 0.02, and 0.02 Å, respectively. In the present investigation, we find that the radical anion with electron attached to the phosphate group at the 5'-position of the nucleotide is unstable. It is interesting to note that the excess electron is distributed around both the base moiety and the 3'-phosphate group of the dipole-bound anion of pdGp in the gas phase; radical anions with an excess electron covalently bound to these two areas should not be unexpected in solution.

To understand the physical background of these different distributions of the unpaired electron on pdGp<sup>•−</sup> and pdG<sup>•−</sup>p, two more model molecules, deoxyguanosine-3'-phosphate (dGp) and deoxyguanosine-5'-phosphate (pdG), were also studied in the same polarizable medium. However, the optimized structures of the radical anions of these two nucleotides (dG<sup>•−</sup>p and pdG<sup>•−</sup>) exhibit only the base-centered radical characteristics. Specifically, the significant dihedral angle  $D_{N3C5O4C4}$  (19.4° and 19.3°) and the elongated C4=O4 (1.306 Å), N3–C4 (1.478 Å), C2–N2 (1.386 Å and 1.384 Å), N1–C6 (1.383 Å and 1.382 Å), and N7=C8 (1.333 Å) bond lengths are found in both dG<sup>•−</sup>p and pdG<sup>•−</sup>. Therefore, the existence of the phosphate-centered radical pdGp<sup>•−</sup> in pdGp is due in part to the cooperative influence of the diphosphate groups and the polarizable medium.

The predicted energy of the base-centered radical anion is 8.7 kcal/mol (0.38 eV) lower than that of the phosphate-centered structure. Both pdG<sup>•−</sup>p and pdGp<sup>•−</sup> lie below the corresponding neutral species (pdGp) from an energetic point of view. The AEA is predicted to be 1.31 eV for forming pdG<sup>•−</sup>p and 0.93 eV for forming pdGp<sup>•−</sup> in aqueous solution. The AEA of the former is close to that predicted for the G<sup>•−</sup> and dG<sup>•−</sup> in aqueous solution (1.27 eV for G and 1.30 eV for dG, on the basis of the structures here optimized with the PCM model); the excess



**Figure 3.** The plots of the SOMOs of the radical anions  $\text{pdGp}^{\bullet-}$  and  $\text{pdG}^{\bullet-}\text{p}$ .

**TABLE 1: Electron Affinities of G, dG, pdG, dGp, and pdGp in Aqueous Solution (in eV)**

process	AEA	VDE <sup>a</sup>
$\text{pdGp} \rightarrow \text{pdGp}^{\bullet-}$	0.93 (0.95) <sup>b</sup>	1.01
$\text{pdGp} \rightarrow \text{pdG}^{\bullet-}\text{p}$	1.31	1.88
$\text{pdG} \rightarrow \text{pdG}^{\bullet-}$	1.35 <sup>c</sup>	1.91 <sup>c</sup>
$\text{dGp} \rightarrow \text{dGp}^{\bullet-}$	1.36 <sup>c</sup>	1.90 <sup>c</sup>
$\text{dG} \rightarrow \text{dG}^{\bullet-}$	1.30 <sup>c</sup>	1.90 <sup>c</sup>
$\text{G} \rightarrow \text{G}^{\bullet-}$	1.27 <sup>c</sup> (1.33) <sup>d</sup>	1.88 <sup>c</sup>

<sup>a</sup>  $\text{VDE} = E(\text{neutral}) - E(\text{anion})$ , in which the energies are evaluated using the optimized anion structures. <sup>b</sup> Ref 12, with the PCM model, based on the gas phase optimized structures. <sup>c</sup> This research. <sup>d</sup> Refs 16 and 17.

electron is expected to reside on the base. Similar AEA values are also found for the monophosphate nucleosides pdG and dGp (1.35 eV to form  $\text{pdG}^{\bullet-}$  and 1.36 eV to form  $\text{dG}^{\bullet-}\text{p}$ ). The AEA of  $\text{pdGp}^{\bullet-}$  is similar to that predicted by PCM modeling of pdGp with the structure optimized in the gas phase (0.95 eV).<sup>12</sup> Therefore,  $\text{pdGp}^{\bullet-}$  should be recognized as the phosphate-centered radical anion. The VDEs of  $\text{pdG}^{\bullet-}\text{p}$  and  $\text{pdGp}^{\bullet-}$  are estimated to be 1.88 and 1.01 eV, respectively. The radical anions of pdGp thus seem to be the electronically stable species in aqueous solution.

Even more direct evidence for the distributions of the excess electron comes from molecular orbital analysis of the singly occupied molecular orbitals (SOMO). This analysis provides an electronic structure-based rationale for the electron-attracting capabilities of the nucleotides. Plots of the SOMOs of  $\text{pdG}^{\bullet-}\text{p}$  and  $\text{pdGp}^{\bullet-}$  are shown in Figure 3. The most striking feature revealed by the SOMOs of these two anions is that the excess electron density is well-localized on either the guanine base moiety or the phosphate group. Although  $\text{pdG}^{\bullet-}\text{p}$  is 8.7 kcal/mol more stable than  $\text{pdGp}^{\bullet-}$ , converting from  $\text{pdG}^{\bullet-}\text{p}$  to  $\text{pdGp}^{\bullet-}$  in aqueous solution seems difficult because the corresponding SOMOs are well-separated. Instead, charge transfer from the phosphate to the base moiety, or vice versa, may trigger chemical reactions with low activation energy barriers, such as C3'–O3' and C5'–O5'  $\sigma$  bond breaking<sup>8</sup> and intramolecular proton transfer.<sup>16,17</sup>

## Conclusions

In summary, the electron distributions for the radical anions of pdGp in aqueous solution are extraordinarily different from those in the gas phase. The excess electron can covalently bind to either the base or the 3'-phosphate moiety of the radical anion. Both  $\text{pdG}^{\bullet-}\text{p}$  and  $\text{pdGp}^{\bullet-}$  are expected to be electronically stable

species in aqueous solution and to be initiators in electron-attachment-induced DNA damages in nature. However, in the presence of the polarizable medium, the base-centered radical anion  $\text{pdG}^{\bullet-}\text{p}$  is more stable than the phosphate-centered structure. By comparison with electron attachment to the monophosphated nucleotide models pdG and dGp, the existence of the phosphate-centered radical  $\text{pdGp}^{\bullet-}$  in pdGp should be attributed to the cooperative influence of the two phosphate groups and the polarizable medium.

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