A Density Functional Investigation of Model Molecules for Ubisemiquinone Radical Anions

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We have applied density functional methods for the determination of structures, vibrational spectra, electron affinities, and isotropic hyperfine coupling constants of the radical anions of 2,3-dimethoxy-1,4-benzoquinone and 2,3-dimethoxy-5,6-dimethyl-1,4-benzoquinone. Our calculations predict three stable conformers with respect to the orientation of the two methoxy groups for both molecules. Different orientations of the methoxy groups are shown to affect force constants and vibrational spectra less dramatically than in the corresponding neutral quinone. The different mode decompositions predicted in symmetrical and nonsymmetrical conformers, respectively, suggest that site specific labeling of one carbonyl group might allow one to distinguish among these conformers. The presented calculations allow us to provide a mode assignment for the C=C and C=O modes which, in most cases, agrees well with previous assignments based solely on experimental data. Electron affinities are shown to vary by up to 40 kJ/mol for different orientations of the methoxy groups. Our calculations demonstrate that the two additional substituents at positions 5 and 6 have to be considered if the molecule is supposed to serve as a model compound for quinones participating in bacterial photosynthesis.

1. Introduction

Quinones play an important role in the electron-transfer processes of photosynthetic bacterial reaction centers. After light excitation of a chlorophyll dimer (*special pair*), an electron is transferred via a bacteriopheophytin molecule to a primary quinone Q_A and to a secondary quinone Q_B. The secondary quinone accepts two electrons through this pathway, and from the cytoplasmic side of the protein it receives two protons. Q_B H$_2$ thus formed leaves the reaction center. Detailed reviews of the primary electron-transfer steps of photosynthesis can be found in refs 1−3.

During photosynthesis, a sequence of electron-transfer reactions and protonation steps occurs at site Q_B, whereas the quinone at site Q_A undergoes only single reduction and oxidation steps. Infrared spectroscopy, which is highly sensitive to even small structural or electronic alterations, allows the detection of intermediate states of the reaction center, i.e., of states with quinones in various oxidation or protonation states. Applying the method of Q$^-$/Q difference spectroscopy, IR absorptions which are not affected through reduction can be eliminated. Using this method, Q_A-/Q_A[4,5] and Q_B-/Q_B[6] spectra could be precisely characterized for both *Rb. sphaeroides* and *Rp. viridis*. Since reduction or protonation of a quinone in a photosynthetic reaction center affects not only the IR spectrum of the isolated molecule in question but also that of its surrounding, such difference spectra cannot be easily interpreted. Reconstitution of reaction centers with isotopically labeled quinones, however, allows separation of the contributions of the quinones from those of the protein.[7−11]

The interpretation of such spectra requires various approaches such as comparison with IR spectra of model compounds in different redox states, investigation of isotope effects on the quinone vibrations, and normal mode calculations.[12−15] The combination of the electrochemistry of quinones in different solvents with FTIR spectroscopy allows generation of redox-induced IR difference spectra and in turn the assignment of the characteristic bands of semiquinone radical anions.[16] Long-living semiquinone radical anions of important classes of quinones involved in photosynthetic processes such as ubiquinones, plastoquinones, and naphthoquinones have recently been prepared through UV irradiation at low temperature and analyzed by FTIR spectroscopy.[17,18] In some of these experiments, it was concluded that the detected species corresponds to the protonated semiquinone and not to the semiquinone radical anion.[18] Additional information about the vibrational spectrum of semiquinone radical anions and protonated semiquinones come from time-resolved resonance Raman spectroscopic investigations of the photochemistry of ubiquinones.[19]

We have demonstrated in several contributions that density functional methods can accurately predict frequencies of C=C and C=O vibrations of *p*-benzoquinone[12] as well as of methyl- or methoxy-substituted *p*-benzoquinone.[15,20] In the case of 2,3-dimethoxy-1,4-benzoquinone, which serves as a model molecule for ubiquinones, we have shown that stable conformations predicted by quantum chemical methods, which account for correlation effects (DFT, MP2) and simple semiempirical calculations, were qualitatively different. We were furthermore able to explain differences in bond lengths and bond stretching force constants among the different stable conformers in terms of a resonance model which involves the two methoxy substituents. In recent contributions we have demonstrated that density functional methods can not only accurately predict vibrational modes of neutral quinones but also of quinones in various oxidation or protonation states.[21,22] In particular, the calculations on the radical anion of 2-methoxy-1,4-benzoquinone have predicted a less pronounced effect of the methoxy group orientation on the vibrational spectrum than in the neutral molecule.

In this contribution we investigate stable structures of the semiquinone radical anion of 2,3-dimethoxy-1,4-benzoquinone and their vibrational spectra. This molecule is supposed to serve as a model compound for ubisemiquinone radical anions which
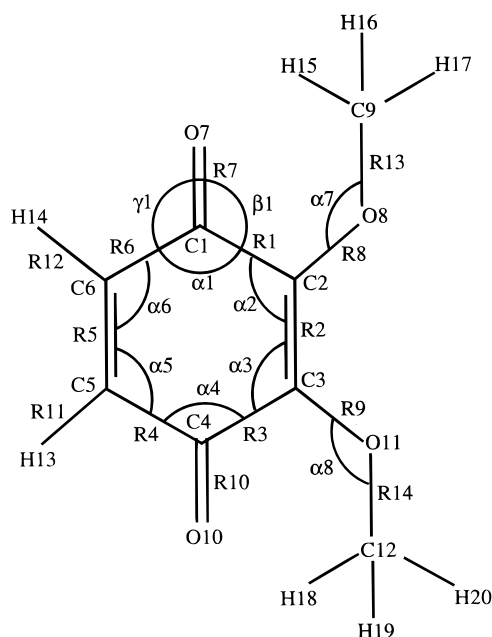
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Figure 1. Definition of valence internal coordinates of methoxy-substituted 1,4-benzoquinone. The drawn conformation shows dihedral angles of the two methoxy groups of 0°.

have been spectroscopically identified in vitro and in bacterial photosynthetic reaction centers. The effect of the orientation of the methoxy groups on structure, bond stretching force constants, and vibrational spectrum are studied. As in the case of the neutral quinone, we investigate whether the orientation of the methoxy groups in the protein could be determined with the help of quantum chemical calculations. For the neutral quinone, a better agreement with experimental data had been achieved after addition of two methyl groups at positions 5 and 6. We have, therefore, also included calculations on the radical anion of 2,3-dimethoxy-5,6-dimethyl-1,4-benzoquinone. Again, addition of the two methyl groups leads to a better agreement between experiment and calculation.

2. Computational Details

Density functional calculations were carried out with the program GAUSSIAN94.[23] The Becke exchange functional[24] combined with the gradient corrected correlation functional of Perdew[25] was applied and a standard 6-31G** basis set was chosen. In earlier contributions we referred to this method as BP86/6-31G**. During geometry optimization, no symmetry constraints were applied.

The calculations of open shell systems were performed by using separate orbitals for α and β electrons as used in the unrestricted Hartree–Fock formalism (UHF). The contribution of wave functions corresponding to states of higher multiplicity (spin contamination), monitored in terms of the expectation value $\langle S^2 \rangle$ before annihilation, was in all cases found to be negligible.

The quantum chemically derived Cartesian force constant matrix was then read into the program GAMESS[26] which was employed to compute normal modes, force constants of internal coordinates, total energy distributions (TED) in order to assign the normal vibrations,[27,28] and frequencies of isotopically labeled compounds. Internal coordinates for the normal coordinate analysis are depicted in Figure 1 and are defined in Table 1. GAMESS was also used for the generation of molecular orbitals for visualization. Molecular orbitals have been visualized with the program MacMolPlt.[29]

Throughout this work calculated harmonic vibrational frequencies are compared to experimentally determined *anharmonic* fundamentals. A justification for this procedure and the impact of this comparison on the quality of the *harmonic* force field have been discussed in detail in ref 12. In short, the applied density functional method tends to slightly overestimate bond lengths. Simultaneously, force constants of bond stretchings are slightly underestimated. Good agreement between calculated and experimental harmonic frequencies, therefore, does not imply good agreement between calculated and experimental harmonic force fields.

3. Results and Discussion

3.1. Structures. To locate the stable conformations of the methoxy groups in the radical anion, we have determined potential energy profiles in the same way as we have previously done for the neutral molecule.[15] The energy profiles for the semiquinone radical anion are depicted in Figure 2. In contrast to our findings in the neutral molecule, we find a qualitative agreement between DFT and semiempirical PM3 calculations. In particular, both methods agree in that they predict only nonplanar orientations of the methoxy groups to be stable (see also ref 17). This agreement between DFT and PM3 methods suggests that correlation effects are less decisive in semiquinone radical anions than in neutral quinones. As in the case of the neutral molecule, complete optimization resulted in three stable conformations which are depicted in Figure 3. One of these stable structures has C_s symmetry (conformer a) and, thus, has both methoxy groups on the same side of the ring. This conformer has been obtained by a geometry optimization involving all degrees of freedom starting with the structure corresponding to the minimum of curve a. Semiempirical calculations had predicted a similar structure also for the neutral molecule.[17] Consideration of correlation effects by means of DFT methods, however, were in contradiction to this result and predicted a planar conformation (conformer A in ref 15). The results of the DFT calculations could be confirmed by the application of second-order perturbation theory calculations (MP2). The second stable radical structure (denoted as conformer b) resulted from the optimization of the minimum found on curve b in Figure 2. Interestingly, the methoxy group which was held at a dihedral angle of 180° during the calculation of the potential energy profile rotated away from the planar orientation and points toward the same side of the ring as the second methoxy group. In calculations on the radical of 2-methoxy-1,4-benzoquinone, we have shown that the orientation with a dihedral angle of 180° corresponds to an energy minimum.[22] The additional methoxy group at carbon C3, thus, seems to cause a repulsive interaction responsible for the predicted reorientation of the methoxy group attached to carbon C2 in the radical anion. The third stable conformation (conformer c) results after complete optimization of the minimum found on curve c and corresponds to structure C found in the neutral molecule. All dihedral angles of the methoxy groups of conformers a and c are very similar. We denote this orientation in the following as orientation I. In conformer b, however, the second methoxy group has a different dihedral angle which we denote as orientation II. The two dihedral angles are ∼53° (orientation I) and ∼ −133° (orientation II). In the neutral molecule, the planar conformer A had the lowest energy followed by conformers B and C. In the radical anion, conformer c has the lowest energy, followed by conformer a at only 0.87 kJ/mol higher energy. The nonsymmetric conformer b has the highest energy, 11.0 kJ/mol above that of conformer

TABLE 1: Numbering and Definition of Internal Coordinates. Valence Coordinates Are Defined in Figure 1

| number	coordinate	description
2, 5	R2, R5	C=C str
1, 3, 4, 6	R1, R3, R4, R6	C−C str
7, 10	R7, R10	C=O str
8, 9, 13, 14	R8, R9, R13, R14	C−O str
11, 12, 15, 16, 17, 18, 19, 20	R11, R12, R15, R16, R17, R18, R19, R20	C−H str
21, 22	$\alpha 7$, $\alpha 8$	C−O−C bend
23, 24, 25, 26, 27, 28	$\alpha 9$, $\alpha 10$, $\alpha 11$, $\alpha 12$, $\alpha 13$, $\alpha 14$	H−C−O bend
29, 33	$\tau 7$, $\tau 10$	C−O−C−C tors
30, 31, 32, 34, 35, 36	$\tau 15$, $\tau 16$, $\tau 17$, $\tau 18$, $\tau 19$, $\tau 20$	H−C−O−C tors
37, 40	$\delta 1$, $\delta 4$	C=O wag (out-of-plane)
38, 39	$\delta 2$, $\delta 3$	C−O wag (out-of-plane)
41, 42	$\delta 5$, $\delta 6$	C−H wag (out-of-plane)
43	$6^{-1/2}(\alpha 1 - \alpha 2 + \alpha 3 - \alpha 4 + \alpha 5 - \alpha 6)$	ring def (in-plane)
44	$12^{-1/2}(2\alpha 1 - \alpha 2 - \alpha 3 + 2\alpha 4 - \alpha 5 - \alpha 6)$	ring def (in-plane)
45	$2^{-1}(\alpha 2 - \alpha 3 + \alpha 5 - \alpha 6)$	ring def (in-plane)
46, 49	$2^{-1/2}(\beta 1 - \gamma 1)$, $2^{-1/2}(\beta - \gamma 4)$	C=O def (in-plane)
47, 48	$2^{-1/2}(\beta 2 - \gamma 2)$, $2^{-1/2}(\beta 3 - \gamma 3)$	C−O def (in-plane)
50, 51	$2^{-1/2}(\beta 5 - \gamma 5)$, $2^{-1/2}(\beta 6 - \gamma 6)$	C−H def (in-plane)
52	$6^{-1/2}(\tau 1 - \tau 2 + \tau 3 - \tau 4 + \tau 5 - \tau 6)$	ring tors
53	$2^{-1}(-\tau 2 + \tau 3 - \tau 5 + \tau 6)$	ring tors
54	$12^{-1/2}(-\tau 1 + 2\tau 2 - \tau 3 - \tau 4 + 2\tau 5 - \tau 6)$	ring tors
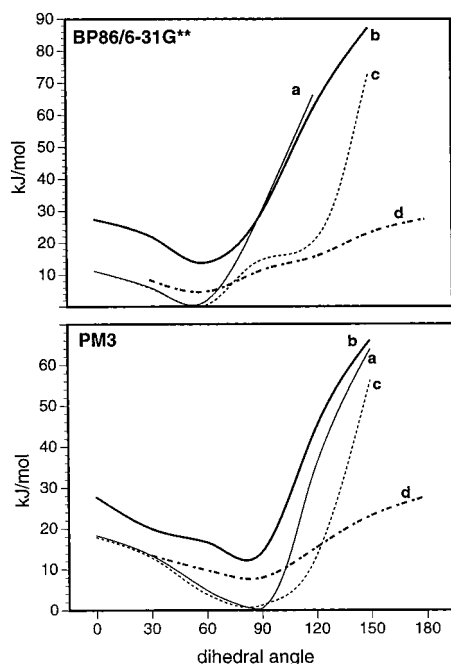
Figure 2. Potential energy profiles for rotation of methoxy groups of the semiquinone radical anion of 2,3-dimethoxy-1,4-benzoquinone: (a) one methoxy group is rotated clockwise, the other one anticlockwise; (b) one methoxy group is kept at a dihedral angle of 180°; (c) both methoxy groups are rotated clockwise; (d) one methoxy group is kept at a dihedral angle of 0°. For definition of dihedral angles, see Figure 1.

Figure 3. Optimized structures of the stable conformers of the semiquinone radical anion of 2,3-dimethoxy-1,4-benzoquinone.

c. The energy difference between a and c is thus very small and both conformers are most likely present in solution spectra. The finding of two orientations of the methoxy groups leads one to suspect that additional combinations of orientations I and II might be stable as well. We have therefore tested the following structures: two times orientation II on different sides of the ring, orientation I on one side and orientation II on the other side of the ring, and two times orientation II on the same side of the ring. All three structures have converged into one of the discussed conformers a, b, or c. We are thus confident, that no additional stable orientations of the methoxy group exist.

For the neutral molecules we have shown that the bond lengths of the CC and CO bonds depend considerably on the orientation of the two methoxy groups.[15] Table 2 clearly shows
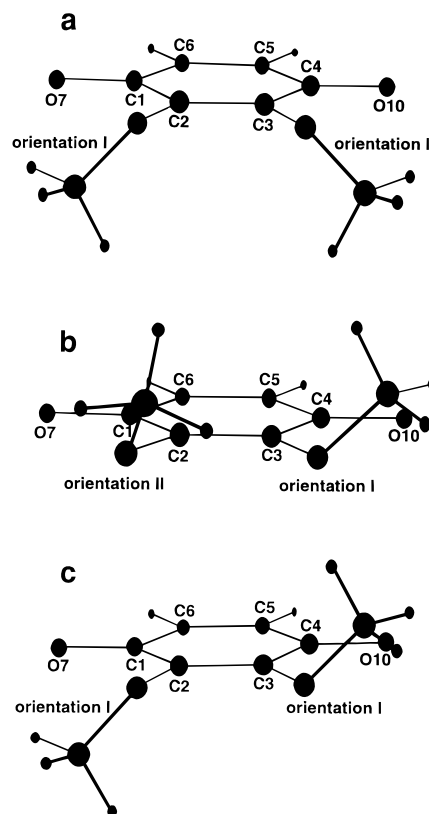
that this effect is much smaller in the semiquinone radical anion. The bond lengths of the three conformers deviate generally less than 0.01 Å. Compared to the semiquinone radical anion of 1,4-benzoquinone, substitution with two methoxy groups results in slightly longer bonds R1, R2, R3, and R4 and in a slightly shorter bond R6. These findings are to a very good approximation independent of the orientation of the methoxy groups. Upon rotation of one methoxy group from orientation I into orientation II, the following structural changes are noticed: the C=O bond of the carbonyl group in ortho position to the rotated methoxy group is shortened by 0.009 Å, and the O−C^{methyl} bond length of the rotated methoxy group is shortened by 0.008 Å. Moreover, the C^{ring}−O and O−C^{methyl} bond lengths of the

TABLE 2: Calculated Bond Lengths (in Å) for the Different Stable Conformers of the Radical Anion of 2,3-Dimethoxy-1,4-benzoquinone Compared to Calculated Structural Parameters of the Semiquinone Radical Anion of 1,4-Benzoquinone

parameter	conformer a	conformer b	conformer c	*p*-benzoquinone[a]
R1	1.472	1.477	1.472	1.462
R2	1.394	1.394	1.395	1.382
R3	1.472	1.473	1.472	1.462
R4	1.472	1.473	1.472	1.462
R5	1.381	1.382	1.381	1.382
R6	1.454	1.456	1.454	1.462
R7	1.280	1.271	1.280	1.277
R8	1.392	1.390	1.391	
R9	1.392	1.399	1.391	
R10	1.280	1.281	1.280	1.277
R13	1.433	1.425	1.433	
R14	1.433	1.437	1.433	

a Reference 21.

second methoxy group are reduced by 0.008 Å and increased by 0.004 Å, respectively. Still smaller changes are found for all other bond lengths. Possible reasons for some of these structural changes will be discussed in the next section.

3.2. Vibrational Frequencies. In a time-resolved resonance Raman investigation of ubiquinone photolysis in water bands at 1617, 1527, and 1474 cm^{-1} have been found.[19] A recent resonance Raman investigation of the radical anion of Q_{10} in ethanol reveals bands at 1610, 1525, and 1485 cm^{-1}.[30] This study also presents first resonance Raman data of semiquinones in the photosynthetic reactions center of *Rb. sphaeroides*. The following bands have been observed: 1605 cm^{-1} ($Q_A^{\bullet-}$) and 1613 cm^{-1} ($Q_B^{\bullet-}$), 1523 cm^{-1} ($Q_A^{\bullet-}$) and 1532 cm^{-1} ($Q_B^{\bullet-}$), and 1486 ($Q_A^{\bullet-}$) and 1489 cm^{-1} ($Q_B^{\bullet-}$). The bands at 1486 and 1489 cm^{-1} are shifted by 30 and 27 cm^{-1}, respectively, upon ^{13}C-labeling and, therefore, have been assigned as C=O stretching modes which are weakly coupled to C=C stretchings.[30]

Infrared data of ubiquinone radical anions in solution have been published by Bauscher and Mäntele.[16] For the semiquinone radical anion of Q_0 in water, absorptions at 1498 and 1476 cm^{-1} had been determined. Absorptions at 1486 (MeCN) and 1466 cm^{-1} (MeOH) (assigned as ν_{C-O}) and at 1424 (MeCN) and 1386 cm^{-1} (MeOH) (assigned as ν_{C-C}) have been measured for the semiquinone radical anion of Q_1. IR spectroscopic data of semiquinone radical anions in photosynthetic reaction centers have been made available by the application of FTIR spectroscopy. Infrared active vibrations have been detected at 1484 ($Q_A^{\bullet-}$, *Rb. sphaeroides*) and 1490 cm^{-1} ($Q_B^{\bullet-}$, *Rb. sphaeroides*, and $Q_B^{\bullet-}$, *Rp. viridis*) and at 1466 and 1448 ($Q_A^{\bullet-}$, *Rb. sphaeroides*), 1479 ($Q_B^{\bullet-}$, *Rb. sphaeroides*), and 1475 cm^{-1} ($Q_B^{\bullet-}$, *Rp. viridis*).[11,31] The ^{13}C shifts of the infrared absorptions of $Q_B^{\bullet-}$ at 1479 and 1475 cm^{-1} of 52 and 42 cm^{-1}, respectively,[11] differ clearly from the shift of 27 cm^{-1} found for the Raman band at 1489 cm^{-1} at the Q_B site.[30] Therefore, the IR bands around 1490 cm^{-1} should correspond to the $Q_B^{\bullet-}$ Raman band at 1489 cm^{-1} whereas the IR absorptions around 1475 cm^{-1} may have possibly not been observed in the resonance Raman investigation.

Selected calculated vibrational frequencies in the 1100 to 1600 cm^{-1} frequency region are listed in Table 3. The most intensive infrared absorptions are predicted around 1520 cm^{-1} (antisymmetric C=O stretching), around 1400 cm^{-1} (CH$_3$ bending), and around 1140−1150 cm^{-1} (combination of the two ring-O$^{\text{methoxy}}$ bond stretchings). At first glance, a straightforward assignment could be proposed as follows. The experimental Raman bands

TABLE 3: Selected Calculated Vibrational Frequencies (in cm^{-1}), Intensities (in km/mol), and Total Energy Distribution (in %) of the Stable Conformers of the Radical Anion of 2,3-Dimethoxy-1,4-benzoquinone

frequency	intensity	TED
	conformer a	
1597	7	18% R2 + 28% R5
1517	272	33% R7 + 33% R10
1502	30	43% R2 + 13% R7 + 13% R10
1465	5	39% R5 + 21% R7 + 21% R10
1402	158	CH$_3$ -bending
1144	151	16% R8 + 16% R9
	conformer b	
1596	16	15% R2 + 28% R5
1528	257	54% R7 + 14% R10
1502	57	39% R2 + 30% R10
1467	3	42% R5 + 14% R7 + 19% R10
1425	34	CH$_3$ -bending
1383	53	CH$_3$ -bending
1151	95	18% R8 + 13% R13
1130	98	21% R9 + 10% R14
	conformer c	
1596	7	17% R2 + 29% R5
1518	271	33% R7 + 33% R10
1498	29	43% R2 + 13% R7 + 13% R10
1465	4	39% R5 + 21% R7 + 21% R10
1402	163	CH$_3$ -bending
1144	169	16% R8 + 16% R9

above 1600 cm^{-1} are assigned to the C=C modes calculated at about 1600 cm^{-1}. This mode had previously been assigned as a C=C stretching[19] or as a C=C stretching which is weakly coupled to C=O stretchings.[30] Experimental frequencies at 1523 and 1532 cm^{-1}, previously assigned as C=C stretchings,[30] would then correspond to the predicted predominant C=O modes around 1515−1530 cm^{-1}. The modes at about 1490 cm^{-1} which had been found in Raman and IR investigations could then correspond to the mixed C=C/C=O modes predicted at about 1500 cm^{-1}, and the IR absorptions around 1475 cm^{-1} would correspond to the mixed C=C/C=O mode predicted around 1465 cm^{-1}.

While this assignment is derived solely from a comparison of calculated and experimental frequencies, it entails mode assignments which in many cases are opposed to previous ones. Most unsatisfactory, however, seems the finding that the mode at about 1520 cm^{-1}, which is predicted to show the largest IR activity, does not correspond to a vibration observed in the FTIR difference spectra.

We, therefore, conclude that the energy sequence of the C=C and C=O modes is not correctly predicted by our model calculation and, correspondingly, that the tentative assignment given above is invalid. We perform our reassignment by focusing on the intensive C=O mode. Correspondingly we assign the strong IR band around 1475 cm^{-1} to our calculated C=O mode at about 1520 cm^{-1} which entails the assumption that the C=O frequency is overestimated by 45 cm^{-1} in our calculation. In previous calculations of the 1,4-benzosemiquinone radical anion,[21] we had found that the calculation overestimates the corresponding frequency by approximately 22 cm^{-1}. Thus, a part of the 45-cm^{-1} deviation between experimental and calculated frequencies might be explained with a systematic computational artifact. Given the C=O mode assignment, the IR and Raman active modes around 1490 cm^{-1} should most likely correspond to the mixed C=O/C=C modes predicted at 1465 cm^{-1}. The experimental Raman bands around 1610 and 1525 cm^{-1} can finally be assigned to the modes predicted around 1600 cm^{-1} (C=C) and around 1500 cm^{-1} (mixed C=O/C=C), respectively.

Ubisemiquinone Radical Anions

J. Phys. Chem. B, Vol. 102, No. 21, 1998 **4221**

With this reassignment we end up with considerable deviations between experimental and calculated frequencies. On the other hand, we find a good agreement between the present calculations and the results obtained for the 1,4-benzosemiquinone radical anion. In that molecule, we had calculated the b_{1u} C=O vibration at 1526 cm^{-1}[21] which corresponds to the experimental band at 1504 or 1508 cm^{-1}.[16] Similarities between the vibrational spectra of the two molecules are to be expected since the additional methoxy substituents have shown to weakly affect only the structure of the 1,4-benzosemiquinone radical anion.

Several reasons can be responsible for the strong deviations of calculated and experimental frequencies noted above. First, the chosen quantum chemical level of theory might not be appropriate for a quantitative prediction of vibrational modes in the case of the open shell systems at hand. Second, as indicated by the experimental data given further above, protein−quinone interactions considerably affect quinone frequencies and are neglected in the calculations. Third, the chosen model molecule, which lacks substituents at carbon atoms C5 and C6, might simply not be appropriate as a model molecule for ubiquinone compounds. The latter point will be discussed in section 3.6 below. In contrast to the case of the neutral quinone, the orientation of the methoxy groups can be excluded as a source of the noted deviations.

For the neutral 2,3-dimethoxy-1,4-benzoquinone, we had found a distinct effect of the methoxy group orientation on the vibrational energies and the mode assignments of the C=C and C=O modes.[15] Inspection of Table 3 makes clear that these effects are considerably smaller in the case of the radical anion. The infrared spectra of conformers a and c are essentially identical concerning vibrational energies and mode assignments. The predicted spectrum of conformer b is only slightly different. Apart from an upshift of the antisymmetric C=O vibration by 11 cm^{-1}, we find a splitting of the infrared active CH$_3$ bending mode at 1402 cm^{-1} into two modes at 1425 and 1383 cm^{-1} and, similarly, also two intensive ring−O^{methoxy} stretching vibrations at 1149 and 1129 cm^{-1}. Analysis of the total energy distribution shows that the slight asymmetry caused by the different orientations of the methoxy groups affects the mode decomposition of several vibrations. We will discuss in the next section whether these differences could be considered as a tool for a spectroscopic identification of the different conformers.

In summary, the presented calculations demonstrate that the vibrational spectrum of the 2,3-dimethoxy-1,4-benzosemiquinone radical anion depends only weakly on the orientation of the two methoxy groups. Experimentally found differences of characteristic modes in various reaction centers or binding sites are therefore presumably caused by differences of electrostatic protein−quinone interactions.

In resonance Raman investigations of ubisemiquinone radical anions in photosynthetic reaction centers and in solution, a strong band around 1605 cm^{-1} had been detected.[30] No corresponding infrared absorption could, however, be found in FTIR investigations.[8,11] According to our calculations, the low energy C=C mode of the neutral 2,3-dimethoxy-5,6-dimethyl-1,4-benzoquinone is at 1590−1595 cm^{-1} and is very intensive. For the semiquinone radical anion of this molecule, we calculate a weak IR absorption around 1590 cm^{-1} which corresponds to the high-energy C=C mode. The C=C modes of the neutral quinone and of the semiquinone radical anion are thus most likely very close in energy, and the weak absorption of the semiquinone

TABLE 4: Calculated Bond Stretching Force Constants (in mdyn/Å) of the Stable Conformers of the Radical Anion of 2,3-Dimethoxy-1,4-benzoquinone

element	a	b	c	1,4-benzoquinone[a]
R1	4.777	4.601	4.778	4.879
R2	7.108	7.078	7.019	7.405
R3	4.776	4.773	4.775	4.879
R4	5.099	5.147	5.102	4.879
R5	7.473	7.455	7.472	7.405
R6	5.099	5.068	5.101	4.879
R7	8.958	9.370	8.963	9.141
R8	5.530	5.560	5.546	
R9	5.530	5.315	5.543	
R10	8.958	8.909	8.963	9.141
R11	4.941	5.046	4.939	
R12	4.941	4.848	4.939	

[a] Reference 21.

TABLE 5: Calculated Isotope Shifts (cm^{-1}) for Conformers a, b, and c

mode	$^{13}C_{all}$	$^{13}C_{1,4}$	$^{13}C_{5,6}$	$^{18}O_{7,10}$	$^{13}C_1$	$^{13}C_4$
			conformer a			
1597	55	11	20	2	5	5
1517	39	35	2	18	9	9
1502	42	8	2	6	9	9
1465	34	18	27	17	11	11
			conformer b			
1596	56	13	20	4	6	6
1528	40	29	2	20	22	6
1501	41	16	0	8	2	10
1467	40	17	32	16	8	12
			conformer c			
1596	56	12	21	3	5	5
1518	39	35	3	19	11	11
1498	41	7	1	6	7	7
1465	34	18	27	17	11	11

radical anion is, therefore, presumably hidden under the intensive signal of the neutral molecule in FTIR difference spectra.[32]

3.3. Isotope Shifts. Only a few experimental data on isotope shifts of semiquinone radical anions in solution are available. In the case of UQ$_{10}$, shifts of 41 and 15 cm^{-1} of the carbonyl mode at 1483 cm^{-1} have been measured upon ^{13}C and ^{18}O labeling, respectively.[16] These shifts agree well with the shifts of ~40 and 18−20 cm^{-1} (see Table 5) which we have calculated for the IR intensive mode around 1520 cm^{-1} after ^{13}C labeling of the ring and ^{18}O labeling of the two carbonyl groups.

Experimental data on isotope shifts of semiquinone radical anions have furthermore been reported in bacterial reaction centers.[30,31] Upon complete ^{13}C-labeling, shifts of 49 and 58 cm^{-1} have been found for the modes above 1600 cm^{-1} at sites Q$_A$ and Q$_B$ of *Rb. sphaeroides*, respectively.[30] The same labeling resulted in shifts of the bands around 1485 cm^{-1} of 30 (Q$_A$) and 27 (Q$_B$) cm^{-1}, respectively.[30] FTIR Q$_B^{\bullet-}$/Q$_B$ difference spectroscopy reveals ^{18}O shifts of 34 (*Rb. sphaeroides*) and 22 cm^{-1} (*Rp. viridis*) and ^{13}C shifts of 52 (*Rb. sphaeroides*) and 42 cm^{-1} (*Rp. viridis*) for bands found at 1479 (*Rb. sphaeroides*) and 1475 cm^{-1} (*Rp. viridis*), respectively.[11]

Calculated isotope shifts for ^{18}O- and ^{13}C-labeling are listed in Table 5. For all conformers, shifts of about 55 cm^{-1} are predicted for the highest energy mode upon ^{13}C-labeling of the ring. Thus, we find a good agreement with experimental data for this mode. Previously we have suggested that the IR active modes observed at 1475 cm^{-1} should most likely correspond to the calculated IR intensive C=O modes around 1520 cm^{-1}. For these modes ^{18}O shifts around 18 cm^{-1} and ^{13}C shifts around 40 cm^{-1} are predicted. Experimental isotope shifts of this mode differ in *Rp. viridis* and *Rb. sphaeroides*. Consequently, this

vibration seems to be considerably perturbed by the protein environment. Note that we find a good agreement of our calculated ^{13}C and ^{18}O shifts for this mode with the data found in *Rp. viridis*.

Previously we have assigned the band at about 1490 cm^{-1} as detected by Raman and IR spectroscopy to the mixed C=C/C=O mode calculated around 1465 cm^{-1}. For this mode we calculate shifts of 34 (ring ^{13}C) and 17 cm^{-1} (^{18}O). The experimental ^{13}C shifts of 30 (Q_A) and 27 cm^{-1} (Q_B) found in *Rb. sphaeroides* agree well with the calculated shift.

Inspection of Table 5 makes clear that conformers a and c cannot be distinguished on the basis of isotope shifts. "Symmetric" labeling such as that of all ring carbon atoms, of both carbonyl oxygens, or of carbons 1 and 4 or 5 and 6 do also not allow us to distinguish between conformers a/c and b. Site specific labeling of only one carbonyl group (only ^{13}C-labeling is technically possible[32]), however, might at least help to discriminate conformers a and c from conformer b. Labeling of carbon C1 results in a shift of 22 cm^{-1} of the mode at 1528 cm^{-1} of conformer b, whereas the corresponding shifts in conformers a and c are only 9 and 11 cm^{-1}, respectively. Carbon C1 belongs to the carbonyl group in ortho position to the methoxy group in orientation II. Labeling of carbon C4 results in a smaller shift of the most intensive IR band at 1528 cm^{-1} of only 6 cm^{-1}. Single ^{13}C-labeling results in a splitting of 15 (conformer a) and 16 cm^{-1} (conformer c) of the two modes around 1500 cm^{-1}, whereas we find splittings of 7 (labeling of C1) and 31 cm^{-1} (labeling of C4) for the corresponding modes of conformer b.

3.4. Force Constants. Calculated bond stretching force constants are listed in Table 4. In accordance with the preceding discussion of structural parameters, only small differences are found for the various stable conformers. Whereas the force constants of conformers a and b are essentially identical, a few differences are found for conformer c. The force constant of C=O bond R7 is ~0.4 mdyn/Å larger and that of C−O bond R9 is ~0.2 mdyn/Å smaller than in conformers a and b. Changing the orientation of the methoxy group at C2 from orientation I to orientation II thus affects the force constants of bonds R7 and R9. We generally find much smaller effects of the methoxy group orientation on the force constants of bond stretchings than in the neutral quinone.[15]

In previous calculations of the radical anion of 2-methoxy-1,4-benzoquinone,[22] we had determined two stable orientations for the methoxy group. In the conformer with the methoxy group in an out-of-plane orientation, the force constants of the C=O groups in ortho and meta positions, respectively, were reduced by 0.279 mdyn/Å and increased by 0.076 mdyn/Å, respectively, as compared to the C=O force constant calculated for the 1,4-benzosemiquinone radical anion. We suggest that the changes of these force constants are caused mainly by an inductive effect of the electronegative oxygen of the methoxy group which pulls electron density out of the ring system. In the second stable conformation with the methoxy group located in the ring plane, a resonance effect becomes effective which mainly results in a strengthening of the carbonyl bond in ortho position and of the C−O bond of the corresponding methoxy group and in a decrease of the force constant of bond R1.

With these results in mind, we can note that the carbonyl force constants of the 2,3-dimethoxy-1,4-benzoquinone radical anions with both methoxy groups in orientation I can in good approximation be predicted as an additive effect of two independent methoxy groups in their nonplanar conformation. The force constant is then calculated to 9.141 (1,4-benzo-
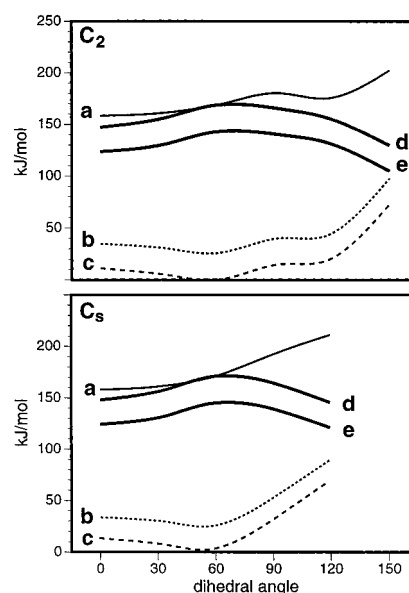

Figure 4. Potential energy profiles for structures of C_2 and C_s symmetry of the neutral quinone (a), nonrelaxed radical anion (b), and fully relaxed semiquinone radical anion (c). Curves d and e represent the resulting adiabatic and vertical electron affinities, respectively.

quinone•$^-$) + 0.067 (effect of a methoxy group on the C=O group in meta position) − 0.279 (effect of a methoxy group on the C=O group in ortho position) = 8.929 mdyn/Å which is close to the values of 8.958 and 8.963 mdyn/Å calculated for conformers a and b, respectively. Different effects are, however, found when the methoxy group at C2 is in orientation II. The force constants of the carbonyl bond in ortho position (R7) and of the C−O bond of the neighboring methoxy group (R9) are mainly affected; the force constant of R7 is increased by ~0.4 mdyn/Å, whereas that of R9 is reduced by ~0.2 mdyn/Å.

The differences in structure and force constants caused by the rotation of a methoxy group from orientation I to orientation II are related to the changes of the shape of the corresponding singly occupied highest occupied molecular orbitals. Corresponding orbitals are depicted in Figure 5 for a structure having two methoxy groups in orientation I (a) and for the nonsymmetric structure with the methoxy group at C2 in orientation II and that at C3 in orientation I (b). Rotation from orientation I to II primarily reduces the antibonding character of the carbonyl group in the ortho position which agrees with the shorter C=O bond and its larger stretching force constant. We also find a smaller π-density around bond R1 which agrees with the found slightly longer bond length. Most of the effects caused by a reorientation of a methoxy group from orientation I to orientation II like the strengthening of the C=O bond in ortho position, the weakening of the proximal C−C "single bond", and the strengthening of the ring−O^{methoxy} bond of the corresponding group can be explained with a resonance effect caused by the respective methoxy group.

3.5. Electron Affinities. For 1,4-benzoquinone it has been shown that the chosen BP86/6-31G** method underestimates the adiabatic electron affinity by 10%.[33] We have found that this error of the calculation is systematic and is also valid in 1,4-naphthoquinone and duroquinone.[22] The BP86/6-31G** method which has been chosen because it allows an accurate prediction of vibrational frequencies at an affordable requirement of computing resources, thus allows us to predict electron affinities of quinones at a well-defined level of accuracy. Relative energies of all optimized structures of the neutral
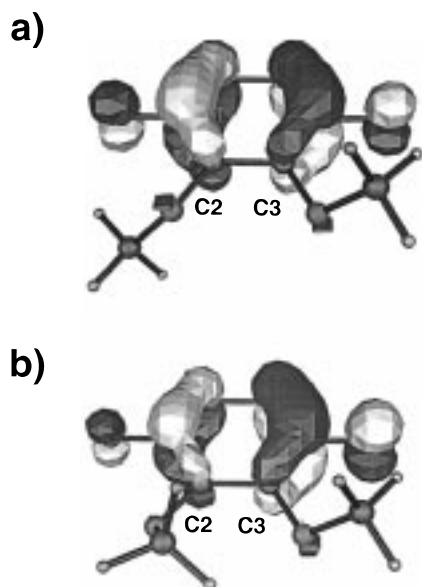
Ubisemiquinone Radical Anions

J. Phys. Chem. B, Vol. 102, No. 21, 1998 **4223**

Figure 5. Highest occupied molecular orbitals for two methoxy groups in orientation I (a) and for one methoxy group in orientation II (at C2) and one in orientation I (at C3).

TABLE 6: Relative Energies and Adiabatic Electron Affinities (in kJ/mol)

radical anion	neutral quinone[a]		
	A	B	C
	159.06	176.46	163.02
a 0.87	158.19	175.59	162.16
b 11.00	148.06	165.46	152.02
c 0.0	159.06	176.46	163.02

[a] For definition of conformers A, B, and C of the Neutral Quinone See ref 15.

molecule and of the radical anion, together with the corresponding adiabatic electron affinities, are listed in Table 6. The values range between 148 and 176.5 kJ/mol or, after correction for the systematic error of 10%, 1.68−2.01 eV. They are thus in the range of values found for 1,4-benzoquinone and of methyl substituted 1,4-benzoquinones.[33] A good agreement is also found with experimental electron affinities of 2,6-dimethoxy-1,4-benzoquinone (1.72 eV) and 2,3-dimethoxy-6-methyl-1,4-benzoquinone (1.86 eV).[34] Furthermore, the smaller calculated values agree well with our calculated adiabatic electron affinity of 147.8 kJ/mol for 2-methoxy-1,4-benzoquinone. The finding that a second methoxy group does not drastically affect electron affinities is also in good agreement with the very similar $E_{1/2}$ values of 2-methoxy-1,4-benzoquinone and 2,3-dimethoxy-5,6-dimethyl-1,4-benzoquinone.[35]

The variation of adiabatic electron affinities as a function of the methoxy group orientation (curves d in Figure 4) is somewhat larger than the range of 144−168 kJ/mol calculated for 2-methoxy-1,4-benzoquinone.[22] Vertical (nonadiabatic) electron affinities are considerably smaller than the values in Table 6. We calculate vertical electron affinities of 123.71, 130.60, and 123.17 kJ/mol for conformers A, B, and C, respectively. A distinct dependence of electron affinities on the orientation of the methoxy groups has already been demonstrated in previous calculations.[22,36,37] For illustration, potential energy profiles for structures of C_2 and C_s symmetry are presented in Figure 4. The curves of the neutral molecules correspond to energy profiles of fully optimized structures. The very same structures have been used for the determination of the energies of nonrelaxed semiquinone radical anions and of

TABLE 7: Calculated Vibrational Frequencies (in cm-1), Intensities (in km/mol), and Total Energy Distribution (in %) of the Stable Conformers of the Radical Anion of 2,3-Dimethoxy-5,6-dimethyl-1,4-benzoquinone

frequency	intensity	TED	^{13}C	^{18}O
		conformer a		
1590	13	25% R2 + 30% R5	58	1
1505	10	38% R2 + 23% R5	46	1
1501	264	36% R7 + 36% R10	37	21
1490	42	18% R7 + 18% R10	33	23
1489	4	15% R7 + 15% R10		
		conformer b		
1586	23	22% R2 + 32% R5	58	2
1512	192	70% R10	39	25
1501	22	34% R2 + 24% R5	45	−1
1492	81	62% R7	34	22
		conformer c		
1588	12	25% R2 + 30% R5	58	1
1503	7	38% R2 + 23% R5	45	1
1500	266	36% R7 + 36% R10	37	21
1489	45	18% R7 + 18% R10	33	23
1487	8	16% R7 + 16% R10		

vertical electron affinities represented in curves b and d, respectively. Curves c finally represents adiabatic electron affinities; i.e., all structural parameters of the semiquinone radical anion except the two torsional angles of the methoxy groups have been fully optimized.

In solution, the small torsional barriers allow a fast structural relaxation after reduction of a quinone. Therefore, experimental electron affinities most likely correspond to the energy difference of fully relaxed quinone and semiquinone radical anion structures.[36] In contrast, at a catalytic site in a protein, the orientations of the methoxy groups of the quinone and the semiquinone radical anion are most likely constrained. A partial relaxation, mainly of bond lengths, can however be expected to occur rapidly. We can thus expect that experimentally found electron affinities are between the values of vertical and adiabatic electron affinities. Figure 4 demonstrates that the two curves are to a good approximation parallel and that complete relaxation increases electron affinities by about 25 kJ/mol. Variations of the dihedral angles of the methoxy groups cause variations in the electron affinities of up to about 40 kJ/mol. Considerably smaller variations have been previously predicted using semiempirical methods.[37]

3.6. Inclusion of Methyl Substituents at Carbon Atoms C5 and C6. In the case of the neutral quinone we had found that addition of two methyl groups at positions 5 and 6 results in a better agreement of calculated and experimental vibrational frequencies and isotope shifts.[15] Our attempts in sections 3.2 and 3.3 to compare calculated frequencies and isotope shifts of the 2,3-dimethoxy-1,4-benzosemiquinone radical anion with experimental data of the semiquinone radical anions of methylated 2,3-dimethoxy-1,4-benzoquinone or of ubiquinone molecules turned out to be to some extent unsatisfactory. We have therefore decided to carry out additional calculations on the semiquinone radical anion of a 2,3-dimethoxy-1,4-benzoquinone with additional methyl substituents at carbons C5 and C6.

Again we find three stable conformers which correspond to the conformers a, b, and c of the nonmethylated semiquinone radical anion. Selected calculated vibrational frequencies together with total energy distributions and ^{13}C and ^{18}O shifts are listed in Table 7. In the symmetric structures a and c we find that the additional methyl groups alter the sequence of the C=C and C=O modes. The IR intensive C=O mode is now predicted at about 1500 cm^{-1}, i.e., only 25 cm^{-1} above the strong experimental IR absorption at about 1475 cm^{-1}. Taking

TABLE 8: Calculated Isotropic Hyperfine Coupling Constants (G)

atom	nonmethylated			methylated		
	a	b	c	a	b	c
H13, H$_3$ (C(13))	−2.96	−2.78	−2.97	2.18	1.96	2.19
H14, H$_3$ (C(14))	−2.96	−3.27	−2.97	2.18	2.44	2.19
C1	0.83	−0.16	0.91	1.42	0.48	1.51
C2	0.75	1.41	0.36	0.95	1.54	0.57
C3	0.75	−0.56	0.36	0.95	−0.15	0.57
C4	0.83	1.36	0.91	1.42	1.98	1.51
C5	1.78	1.44	1.78	1.67	1.20	1.67
C6	1.78	2.35	1.78	1.67	2.27	1.67
O7	5.10	5.13	5.12	5.04	5.10	5.07
O8	0.83	1.27	1.00	0.98	1.33	1.21
C9	0.04	1.32	0.03	−1.32	−1.47	−1.32
O10	5.10	4.94	5.12	5.04	4.92	5.07
O11	0.83	0.10	1.00	0.98	0.47	1.21
C12	0.04	−0.41	0.03	−1.32	−1.22	−1.32
C13				0.18	0.19	0.16
C14				0.18	−0.24	0.16

the previously noted computational artifact of ∼20 cm^{-1} for this mode into consideration, we now achieve good agreement with the position of the intensive bands found in the FTIR investigations of *Rb. sphaeroides* and *Rp. viridis*. After addition of the methyl groups, the mixed C=O/C=C mode at 1465 cm^{-1} is shifted to 1490 cm^{-1} and agrees well with bands at about 1490 cm^{-1} as observed in Raman and FTIR investigations of Q$_6$ in photosynthetic reaction centers. The addition of the two methyl groups removes the C=O/C=C coupling and the mode corresponds now to a predominant C=O mode with only small contributions from other vibrations. This mode decomposition disagrees, however, with a previous assignment of this mode as a predominant C=C vibration[31] which was based on the study of isotope shifts. The found disagreement between experimental data and calculation could either be caused by the different substituents at C5 or C6 or by protein−chromophore interactions.

Besides a better agreement with experimental frequencies, we also find a better agreement for some isotope shifts. The C=O mode at lower energy, now predicted at 1490 cm^{-1}, exhibits ^{13}C and ^{18}O shifts of 33 and 23 cm^{-1}, respectively. The ^{13}C shift agrees well with the shifts of 30 and 27 cm^{-1} as found in *Rb. sphaeroides* at sites Q$_A$ and Q$_B$, respectively.[30] Furthermore, ^{13}C and ^{18}O shifts of the IR active C=O mode of 37 and 21 cm^{-1}, respectively, agree well with the FTIR data (42 and 22 cm^{-1}) found in *Rp. viridis*.[11]

3.7. Isotropic Hyperfine Coupling Constants. Besides vibrational spectroscopy, EPR experiments are a very useful additional technique for the investigation of photosynthetic processes.[38−40] The capabilities of density functional and hybrid Hartree−Fock/density functional methods for the calculation of hyperfine coupling constants of cyclic delocalized neutral or ionic radicals has been demonstrated in several recent publications.[41−43] A good agreement between experimental and theoretical proton hyperfine coupling constants has been found, for example, in the phenol radical cation with BLYP, B3LYP, and BP86 methods. We have listed calculated isotropic hyperfine coupling constants determined by the use of the BP86 functional in Table 8. In our previous investigation on radicals of 1,4-benzoquinone,[21] we have found that hyperfine coupling constants calculated using the BP86 and BLYP functionals generally agree well with experimental data in the case of hydrogen atoms. On the other hand, calculated hyperfine coupling constants of carbon or oxygen varied strongly among different methods. Experimental hyperfine coupling constants of semiquinone radical anions of ubiquinone-0 (UQ-0) and

ubiquinone-10 (UQ-10) have been determined in protic and aprotic solutions.[44−47] Isotropic hyperfine coupling constants of 2.25 G had been measured for the proton connected to the ring and for the protons attached to the methyl group (UQ-0).[47] This latter constant is reduced to 2.11[47] or 2.15 G[46] when the isoprene chain is added. We have calculated coupling constants of about −2.95 for the proton connected to the ring and of about 2.2 G for the protons of the methyl groups in conformers a and c. We thus find a good agreement for the methyl protons. The larger deviation in the case of the proton connected to the ring might be a shortcoming of our model molecules which have either two protons or two methyl groups attached to carbon atoms C5 and C6. Conformer b has clearly different hyperfine coupling constants of −2.78 and −3.27 G for the two protons connected to the ring or, in the case of the dimethylated molecule, of 1.96 and 2.44 G for the protons of the two methyl groups. Nonsymmetrical conformers should, thus, most likely be distinguished with the help of EPR spectroscopy.

Experimental ^{13}C hyperfine coupling constants furthermore show that the coupling constants of carbons C1 and C4 are clearly larger than those of the other ring carbon atoms. Our calculations predict larger constants for carbons C1 and C4 than for C2 and C3 most pronounced for conformer c. A large coupling constant is, however, also predicted for carbons C5 and C6 for all structures with or without additional methyl groups. In agreement with experimental data, coupling constants of the methoxy groups are very small and are not included in Table 8.

4. Conclusions

We have presented density functional calculations of structures, force constants, vibrational spectra, electron affinities, and isotropic hyperfine coupling constants of the semiquinone radical anions of 2,3-dimethoxy-1,4-benzoquinone and 2,3-dimethoxy-5,6-dimethyl-1,4-benzoquinone which both had been supposed to serve as model molecules for ubisemiquinone radical anions. The calculations made clear that inclusion of two substituents at carbons C5 and C6 is necessary in order to achieve a satisfactory agreement with experimental vibrational frequencies and isotope shifts. The calculations moreover demonstrate that the effect of the methoxy group orientation on the vibrational spectrum is less pronounced than in the corresponding neutral molecules. Correspondingly, frequency differences observed in various reaction centers of binding sites are most likely due to differences of electrostatic quinone−protein interactions. Although only a very limited set of experimental vibrational frequencies and isotope shifts is available, one may conclude that the chosen BP86/6-31G** method provides a useful tool for the prediction of molecular properties of quinone radical anions.

The ability of the methoxy group to tune the electron affinity by changing its orientation has been shown in previous calculations at the ab initio Hartree−Fock[36] and semiempirical PM3[17,37] level of theory. The DFT calculations, which are expected to give more accurate predictions than the methods used in previous investigations, predict this effect to be even more pronounced.

Calculations at the chosen level of theory can give useful estimates of proton isotropic hyperfine coupling constants. Besides the frequencies of the intensive C=O stretching mode and, more promisingly, the isotope shifts of this mode after site specific carbonyl labeling, hyperfine coupling constants of ring protons or methyl group protons may be able to distinguish among different conformers with regard to the orientation of the methoxy groups.

References and Notes

(1) Kirmaier, C.; Holten, D. *Photosynth. Res.* **1987**, *13*, 225.

(2) Parson, W. W. In *New Comprehensive Biochemistry: Photosynthesis*; Amesz, J., Ed.; Elsevier: Amsterdam, 1987; p 43.

(3) Boxer, S. G.; Goldstein, R. A.; Lockhart, D. J.; Middendorf, T. R.; Takiff, L. *J. Phys. Chem.* **1989**, *93*, 8280.

(4) Breton, J.; Thibodeau, D.; Berthomieu, C.; Mäntele, W.; Verméglio, A.; Nabedryk, E. *FEBS Lett.* **1991**, *278*, 257.

(5) Nabedryk, E.; Berthomieu, C.; Verméglio, A.; Breton, J. *FEBS Lett.* **1991**, *293*, 53−58.

(6) Breton, J.; Berthomieu, C.; Thibodeau, D.; Nabedryk, E. *FEBS Lett.* **1991**, *288*, 109.

(7) Brudler, R.; de Groot, J.; van Liemt, W.; Steggerda, W.; Esmeijer, R.; Gast, P.; Hoff, A.; Lugtenburg, J.; Gerwert, K. *EMBO J.* **1994**, *13*, 5523−5530.

(8) Breton, J.; Burie, J.-R.; Berthomieu, C.; Berger, G.; Nabedryk, E. *Biochemistry* **1994**, *33*, 4953.

(9) Breton, J.; Boullais, C.; Burie, J.-R.; Nabedryk, E.; Mioskowski, C. *Biochemistry* **1994**, *33*, 14378−14386.

(10) Breton, J.; Burie, J.-R.; Boullais, C.; Berger, G.; Nabedryk, E. *Biochemistry* **1994**, *33*, 12405−12415.

(11) Breton, J.; Boullais, C.; Berger, G.; Mioskowski, C.; Nabedryk, E. *Biochemistry* **1995**, *34*, 11606−11616.

(12) Nonella, M.; Tavan, P. *Chem. Phys.* **1995**, *199*, 19.

(13) Burie, J.-R.; Nonella, M.; Nabedryk, E.; Tavan, P.; Breton, J. In *Fifth International Conference on the Spectroscopy of Biological Molecules*; Theophanides, T., et al., Eds.; Kluwer Academic Publishers: Dordrecht, Boston, London, 1993; p 27, 28.

(14) Nonella, M. *J. Mol. Struct. (THEOCHEM)* **1996**, *362*, 7−22.

(15) Nonella, M.; Brändli, C. *J. Phys. Chem.* **1996**, *100*, 14549−14559.

(16) Bauscher, M.; Mäntele, W. *J. Phys. Chem.* **1992**, *96*, 11101.

(17) Burie, J.-R. Ph.D. Thesis, University of Paris, Paris IV, 1994.

(18) Burie, J.; Boussac, A.; Boullais, C.; Berger, G.; Mattioli, T.; Mioskowski, C.; Nabedryk, E.; Breton, J. *J. Phys. Chem.* **1995**, *99*, 4059−4070.

(19) Parker, A. W.; Hester, R. E.; Phillips, D.; Umapathy, S. *J. Chem. Soc., Faraday Trans.* **1992**, *88*, 2649−2653.

(20) Nonella, M. *J. Phys. Chem.* **1996**, *100*, 20148−20155.

(21) Nonella, M. *J. Phys. Chem. B* **1997**, *101*, 1235−1246.

(22) Nonella, M. *Photosynth. Res.*, in press.

(23) Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Gill, P. M. W.; Johnson, B. G.; Robb, M. A.; Cheeseman, J. R.; Keith, T.; Petersson, G. A.; Montgomery, J. A.; Raghavachari, K.; Al-Laham, M. A.; Zakrzewski, V. G.; Ortiz, J. V.; Foresman, J. B.; Peng, C. Y.; Ayala, P. Y.; Chen, W.; Wong, M. W.; Andres, J. L.; Replogle, E. S.; Gomperts, R.; Martin, R. L.; Fox, D. J.; Binkley, J. S.; Defrees, D. J.; Baker, J.; Stewart, J. P.; Head-Gordon, M.; Gonzalez, C.; Pople, J. A. GAUSSIAN94, Revision d.2.; Gaussian, Inc.: Pittsburgh, PA, 1995.

(24) Becke, A. D. *Phys. Rev. A* **1988**, *38*, 3098.

(25) Perdew, J. P. *Phys. Rev. B* **1986**, *33*, 8822.

(26) Schmidt, M. W.; Baldridge, K. K.; Boatz, J. A.; Jensen, J. H.; Koseki, S.; Gordon, M. S.; Nguyen, K. A.; Windus, T. L.; Elbert, S. T. *QCPE Bull.* **1990**, *10*, 52.

(27) Pulay, P.; Török, F. *Acta Chim. Hung.* **1965**, *47*, 273.

(28) Keresztury, G.; Jalsovszky, G. *J. Mol. Struct.* **1971**, *10*, 304.

(29) Bode, B. *MacMolPlt*; Department of Chemistry, Iowa State University, 1996.

(30) Zhao, X.; Ogura, T.; Okamura, M.; Kitagawa, T. *J. Am. Chem. Soc.* **1997**, *119*, 5263−5264.

(31) Breton, J.; Nabedryk, E. *Biochim. Biophys. Acta* **1996**, *1275*, 84−90.

(32) Breton, J. Private communication.

(33) Boesch, S. E.; Grafton, A. K.; Wheeler, R. A. *J. Phys. Chem.* **1996**, *100*, 10083−10087.

(34) Heinis, T.; Chowdhury, S.; Scott, S. L.; Kebarle, P. *J. Am. Chem. Soc.* **1988**, *110*, 400−407.

(35) Morrison, L. E.; Schelhorn, J. E.; Cotton, T. M.; Bering, C. L.; Loach, P. A. In *Function of Quinones in Energy Conserving Systems*; Trumpower, B. L., Ed.; Academic Press: New York, London, 1982; pp 35−58.

(36) Robinson, H. H.; Kahn, S. D. *J. Am. Chem. Soc.* **1990**, *112*, 4728.

(37) Burie, J.-R.; Boullais, C.; Nonella, M.; Mioskowski, C.; Nabedryk, E.; Breton, J. *J. Phys. Chem. B* **1997**, *101*, 6607−6617.

(38) Pedersen, J. A. *Handbook of EPR Spectra from Quinones and Quinols;* CRC Press: Boca Raton, 1985.

(39) Feher, G.; Okamura, M. Y.; McElroy, J. D. *Biochim. Biophys. Acta* **1983**, *267*, 222.

(40) Gast, P.; de Groot, A.; Hoff, A. J. *Biochim. Biophys. Acta* **1983**, *723*, 52.

(41) Quin, Y.; Wheeler, R. A. *J. Chem. Phys.* **1995**, *102*, 1689.

(42) Quin, Y.; Wheeler, R. A. *J. Phys. Chem.* **1996**, *100*, 10554.

(43) Walden, S. E.; Wheeler, R. A. *J. Phys. Chem.* **1996**, *100*, 1530.

(44) Samoilova, R. I.; van Liemt, W.; Steggerda, W. F.; Lugtenburg, J.; Hoff, A. J.; Spoyalov, A. P.; Tyryshkin, A. M.; Gritzan, N. P.; Tsvetkov, Y. D. *J. Chem. Soc., Perkin Trans. 2* **1994**, 609−614.

(45) Samoilova, R. I.; Gritzan, N. P.; Hoff, A. J.; van Liemt, W. B. S.; Lugtenburg, J.; Spoyalov, A. P.; Tsvetkov, Y. D. *J. Chem. Soc., Perkin Trans. 2* **1995**, 2063−2068.

(46) MacMillan, F.; Lendzian, F.; Lubitz, W. *Magn. Res. Chem.* **1995**, *33*, S81−S93.

(47) Kropacheva, T. N.; van Liemt, W. B. S.; Raap, J.; Lugtenburg, J.; Hoff, A. J. *J. Phys. Chem.* **1996**, *100*, 10433.