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# Site Dependence of the Binding Energy of Water to Indole: Microscopic Approach to the Side Chain Hydration of Tryptophan

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A microscopic approach to the in vacuo energetics of the binding of a water molecule to the side chain of tryptophan, modeled by an indole molecule, is presented. Two binding sites have been studied, the most bound one, which corresponds to the conventional NH−$OH_2$ hydrogen bond and a slightly less bound, so-called $\pi$-type hydrogen bond in which the hydrogen atoms interact with the $\pi$ aromatic ring of indole. The structure of these two complexation sites as well as the potential energy surface has been obtained by a semiempirical model coupled with efficient procedures for the exploration of the surface. The H-bonded complex was observed in the supersonic expansion, and its binding energy (4.84 ± 0.23 kcal/mol) was measured using a laser two-color photofragmentation technique. The nonstandard H-bonded complex, not observed with indole, was observed with 1-methylindole, a substituted indole, in which the formation of the conventional bond is hindered. Its binding energy, measured with a similar accuracy (4.10 ± 0.14 kcal/mol), can be used as a fair estimate of the binding energy of the $\pi$-type complex of indole−water, as suggested by our calculations. The small difference in the binding energy between the two gas-phase complexes suggests that, although being traditionally considered as a highly hydrophobic residue, the side chain of tryptophan is not only able to established a H-bond with a proton acceptor but also can exhibit significant nonstandard interactions with an aqueous environment.

## 1. Introduction

Energetics data are rather scarce for gas-phase complexes, although they are important for validating the intermolecular model potentials that are currently used in chemistry or biochemistry.[1−3] In this respect these in vacuo measurements are often a unique alternative to ab initio calculations. Very recently, the binding energies of several hydrogen-bonded complexes of small aromatic organic molecules with water were measured. Two types of hydrogen bonds have been investigated. On one hand, typical H-bonds, in which the molecule acts as a proton donor, have been characterized, namely, the H-bonds of water with phenol,[4−8] with 1-naphthol,[9] and very recently with indole.[10−16] The binding energies found are all in the 200−250 meV (4.5−6.2 kcal/mol) energy range. On the other hand, evidence for a less conventional type of hydrogen bond in which the aromatic molecule acts as a proton acceptor via its $\pi$ cloud has been given for aromatic molecules that cannot be proton donors, for example, benzene.[17−20] In this latter case, the energy of this nonstandard H-bond was found to be about 110 meV (2.5 kcal/mol).[20]

However, all these binding energy measurements pertain to a well-defined geometry of the system: namely, that of the gas-phase complex, which corresponds to the most stable minimum of the potential energy surface. It is nevertheless interesting to access energetic data relative to other parts of the potential energy surface. In particular, with a planar aromatic molecule exhibiting a H-bond site, like indole for instance, one can expect that the two types of complexes evoked above can be observed (Figure 1). In other terms, a $\pi$-type complex can also occur for the indole−water complex, even if, so far, no direct experimental evidence of its existence has been given, probably because of its lesser stability compared to the H-bonded species and hence its very low population in a supersonic jet.

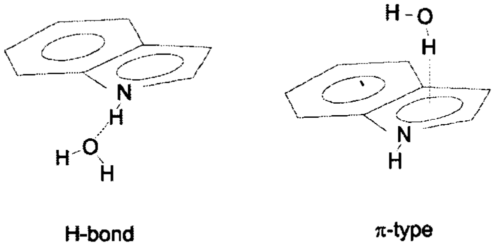

**Figure 1.** Schematic view of the two possible isomers H-bonded indole−water complexes: conventional H-bonded complex (H-bond) and nonstandard H-bonded complex ($\pi$ type).

Thus, an alternative way to approach the energetics of this $\pi$-type isomer consists of slightly modifying the molecule in order to inhibit the formation of the H-bond isomer. This can be achieved by substituting the H atom involved in the H-bond by a methyl group, for instance. In such a case, the H-bonded isomer does not exist any more and the complex with the substituted molecule will have another geometry, which should be $\pi$-type. Since the methylation does not affect strongly the electronic properties of the molecule, a comparison of the binding energy of the H-bonded and $\pi$-type complexes thus allows us to compare the energetics of two different binding sites of water to the indole molecule.

The choice of indole in the present study has been motivated by its relevance in the solvation study of biological molecules,

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since indole constitutes the side chain of the tryptophan amino acid. Because tryptophan is considered as one of the most hydrophobic residue by biochemists,[21] its interaction with water is often disregarded. Nevertheless, recent studies suggest that it actually seems to behave like an amphiphilic species having affinities for both hydrophobic and hydrophilic environments.[22] These properties would indeed allow it to play a key role in biological processes such as membrane translocation of proteins or protein anchoring.[22] This point is supported by a recent analysis of protein structural data that report an unusual high incidence of tryptophan residues in transmembrane proteins.[23] Another experimental study has provided evidence for the possibility of using 1-alkyl indoles as surfactant molecules to form vesicles.[24] In such a case the hydrophilic head of the molecule is the indole double ring itself, which suggests that its interaction with water is probably as large as in a conventional H-bond.

The present article reports a measurement of the binding energy of indole−water and 1-methylindole−water gas-phase complexes using a photoionization-based technique, already successfully employed on the complexes of water with benzene[20] and phenol.[8] These energetic data have been completed by semiempirical calculations in order to support the above assumptions, in particular the similar nature and energetics of the $\pi$-type indole−water and 1-methylindole−water complexes, and to provide a better understanding of the potential energy surface of the indole−water system.

## 2. Experimental Setup

The experimental setup, already described elsewhere,[25] combines a supersonic beam, dye lasers, and a time-of-flight mass spectrometer. The neutral complexes are formed by the expansion of a gas mixture containing the room-temperature vapor pressure of indole or 1-methylindole (Aldrich) with water in argon. The pulsed expansion is generated by a commercial pulsed valve (General Valve) of 0.3 mm diameter nozzle operating at a frequency of 10 Hz. The jet is skimmed before entering the mass spectrometer chamber (pressure less than $10^{-6}$ Torr during operation) perpendicular to the spectrometer axis. The complexes are excited and ionized by light from two separately pumped (Lambda Physik LPX 200 and EMG 201) dye lasers (Lambda Physik FL 3002 and 2002). The two pulsed laser beams were electronically synchronized in order to cross simultaneously the pulsed molecular beam in the interaction chamber of the mass spectrometer. The ions thus formed are accelerated in the spectrometer source and sent to the microchannel plate detector after transverse deflection in order to compensate for the jet velocity. The mass spectra are averaged on a numeric oscilloscope (LeCroy 9350) and then processed with a LabView based computer program.

The experimental interrogation of the energetics of the complexes is focused on their spectroscopic behavior in the vicinity of the first electronic transition of indole (35 232 $\text{cm}^{-1}$)[10,11] and 1-methylindole (34 542 $\text{cm}^{-1}$)[26] and of the ionization threshold of indole (62 598 $\pm$ 5 $\text{cm}^{-1}$)[10] and 1-methylindole (60 754 $\pm$ 8 $\text{cm}^{-1}$, 7.533 $\pm$ 0.001 eV, present work). The fragmentation rate was carefully measured by integrating the parent and daughter ion signals and subtracting the signal baseline. The values of the ionization energy and fragmentation ratio given in the figures are not corrected for the electric field in the spectrometer (200 V/cm).

## 3. Experimental Results

**3.1. Principle of the Experiment.** The principle of the two-color experiment has been given elsewhere.[8] The first photon excites the complex in the $S_1$ state. The energy of the second photon is scanned above the photoionization region of the complex, and fragmentation is monitored in a time-of-flight mass spectrometer. Generally speaking, this type of experiment requires that photoionization is able to populate inter- and/or intravibrational (or vibronic) levels of the ion in the energy range corresponding to the ionic complex dissociation limit. Afterward, redistribution of the vibrational energy takes place toward the high density of intermolecular modes and fragmentation is detected as soon as the intermolecular vibrational energy overcomes the ionic binding energy.

Previous experiments on benzene−water[20] and phenol−water[8] systems have shown that significant and even large inter- and intramolecular geometry changes between the neutral $S_1$ state and the ion can greatly enhance the formation of highly vibrationally excited ions. In particular the photoionization of the $\pi$-type H-bonded benzene−water complex leads only to small population of stable, nondissociative benzene$^+$−water clusters. This observation has been interpreted as being a result of very poor Franck−Condon overlap for the intermolecular modes between the $S_1$ state complex and the low-lying vibrational ionic levels, due to a very large change of the minimum energy structure.[20] Photoionization efficiency becomes significant toward high-lying levels, very close to the dissociation limit of the ion. Thus, a high fragmentation ratio (85%) was obtained in the one-color two-photon ionization experiment in which the total two-photon energy is high enough to encompass the whole Franck−Condon envelop. In contrast, the fragmentation ratio observed with phenol−water[8] was much smaller (10%) because both inter- and intramolecular geometry changes between the neutral and the ion remain qualitatively small.

**3.2. One-Color Mass Spectra.** Owing to the above-mentioned considerations, one-color two-photon ionization spectra allow us to establish the feasibility of the two-color binding energy measurements.

Mass spectra of the 1:1 complexes of indole and 1-methylindole with water have been obtained by pumping an intense band of the $S_1 \leftarrow S_0$ complex transition, located respectively at −132 [10−12] (origin) and at −71 $\text{cm}^{-1}$ [13] (vibronic band) of the monomer origin.[10,11,26] Fragmentation ratios of 55% and 94% have been measured for the indole−water and 1-methylindole−water, respectively. This propensity of the ionic clusters to fragment suggests that ions are formed with significant excess energy compared to the complexes of phenol, for instance. This is easily understood, since indoles are known to present rich photoelectron spectra in the low internal energy region.[27] Significant vibrational activity is observed for the ground-state ion and also in the first electronic state whose origin lies at ca. 0.5 eV from the ground state.

The presence of a near-lying electronic state allows us to interpret the difference between two H-bonded complexes with water: that of phenol and indole. In the former case fragmentation is due to the tail of the intramolecular Franck−Condon envelop, while in the latter case, fragmentation arises mainly from excitation of the first excited electronic state followed by internal conversion and vibrational redistribution. However, the difference between 1-methylindole complexes and that of indole (whose photoelectron spectra are similar[27]), together with the similarity with that of benzene, suggests that in the 1-methylindole−water complex ionization mainly leaves the ion in very excited intramolecular vibrational dissociative levels. Fragmentation ratios as large as those measured with benzene supports a $\pi$-type structure of this complex.

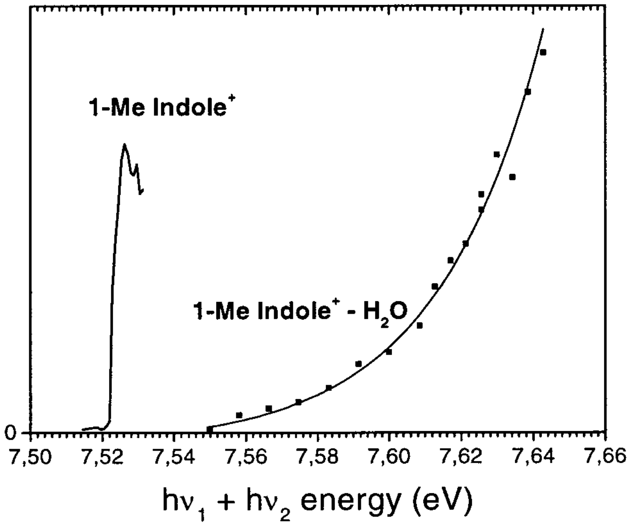

**Figure 2.** Comparison of the two-color photoionization efficiency spectra of 1-methylindole and 1-methylindole−water complex. The energy values are not corrected for the effect of field ionization.

**3.3. Photoionization Spectrum of the 1-Methylindole−Water Complex.** Previous studies of the photoionization of indole−water[10,15] have found that this complex exhibits a well-defined photoionization threshold, followed by successive thresholds assigned to the stretching of the H-bond in the ion, showing a large similarity to the phenol−water complex.[28]

For the sake of comparison we have investigated the photoionization of the 1-methylindole−water close to its threshold (Figure 2). No clear 1-methylindole$^+$−water signal could be observed until a two-photon energy of 7.55 eV was used, i.e., even above the ionization potential of the bare molecule. In contrast to the indole−water complex, a slowly increasing ion signal is observed, showing the same characteristics as the benzene−water complex.[20] As already discussed in this previous work, such an ill-defined ionization threshold is a signature of a nonadiabatic ionization threshold due to the large geometry changes between the minimum energy structure of the neutral and the ion. This feature supports the fact that in the complex of 1-methylindole the water molecule occupies a $\pi$-type hydration site.

**3.4. Two-Color Fragmentation Spectra.** The binding energies $D_0$ of the indole−water and 1-methylindole−water complexes have been measured using the same procedure as previously,[8,20] from the appearance potential (AP) of the molecular ion (indole$^+$ or 1-methylindole$^+$) following the excitation of the $S_1 \leftarrow S_0$ complex band, according to the relation

$$D_0 = \mathrm{AP}(\mathrm{M}^+\cdots\mathrm{S} \rightarrow \mathrm{M}^+ + \mathrm{S}) - \mathrm{IP}(\mathrm{M})$$

where IP(M) is the ionization potential of the bare molecule.

Figure 3 shows the fragmentation probability of the indole$^+$−water and 1-methylindole$^+$−water complexes as a function of the two-photon energy. For both complexes studied, the fragmentation ratio is found to increase nearly linearly from a well-defined onset. The appearance energy has been taken at the intercept of the linear slope with the below-threshold background. The ionization energy of the bare molecules measured under the same experimental conditions, in particular in the same extraction electric field in the mass spectrometer, allows us to yield a value of the ground-state binding energy of the two complexes. The present values, 210 ± 10 meV for indole−water and 178 ± 6 meV for 1-methylindole−water (Table 1), have been obtained from an average of five different fragmentation spectra. The uncertainties mentioned account for the reproducibility of the experiment.

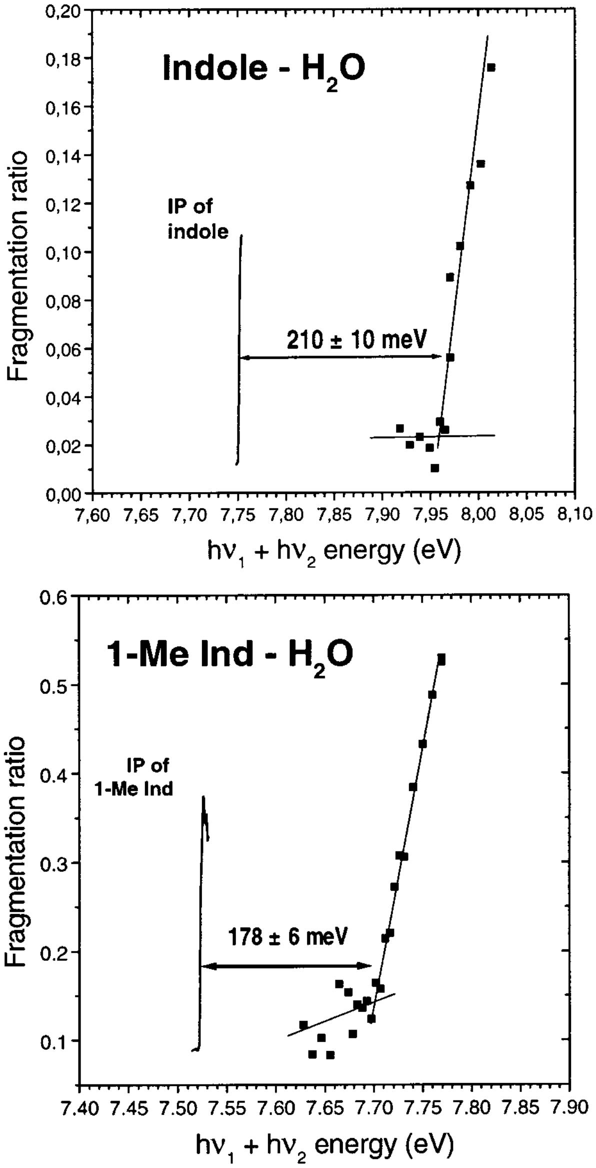

**Figure 3.** Fragmentation efficiencies of the ionized complexes obtained after two-color photoionization of the indole−water and 1-methylindole−water species, given as a function of the total photon energy $\nu_1 + \nu_2$. For reference, the two-color photoionization spectrum of the bare molecules is also given. The energy values are not corrected for the effect of field ionization.

The value obtained for the indole−water complex, although slightly less precise, is close to the value very recently reported by Neusser and co-workers,[15] using the mass-analyzed threshold ionization technique (Table 1). This agreement validates the present measurement. The $D_0$ value of the $\pi$-type complex of 1-methylindole is less than that of the H-bonded complex of indole, but only by 32 ± 16 meV, which represents only ~15% of the indole−water binding energy.

The present values can be used to derive binding energies of the ionic species, provided that the ionization threshold measured for the complex corresponds to the adiabatic ionization potential. This is the case for the indole$^+$−water complex. The IP shift of ref 15 allows us to give an ionic binding energy of 602 ± 11 meV, in agreement with the value of ref 15.

It should be noted that because of the nonadiabatic character of the ionization threshold measured for the 1-methylindole−

**TABLE 1: Experimental Measurements and Theoretical Estimates of the Binding Energy $D_0$ and Semiempirical and ab Initio Well Depths $D_e$ of the 1:1 Water Complexes of Indole and 1-Methylindole**

| | indole−water | | | | 1-methylindole−water | | |
|---|---|---|---|---|---|---|---|
| | experimental | | semiemp | ab initio | experimental | semiemp | ab initio |
| | this work | ref 15 | this work | ref 13 | this work | this work | ref 13 |
| $D_0$ (meV) | 210 ± 10 | 202 ± 2 | | | 178 ± 6 | | |
| $D_0$ (kcal/mol) | 4.84 ± 0.23 | 4.67 ± 0.04 | 3.56[a] | 3.62 | 4.10 ± 0.14 | 3.27[a] | 1.66 |
| $D_e$ (kcal/mol) | | | 4.99 | 5.05 | | 4.47 | 2.86 |

[a] Assuming the ZPE energy calculated in ref 13.

water complex, no precise evaluation of the ionic binding energy can be given. In particular, it should not be concluded from the observation of the ionization threshold at a higher energy than the IP of the bare molecule (Figure 2) that the ionic complex is less bonded than in the neutral state.

## 4. Theoretical Calculations

**4.1. Methodology.** Molecular clusters generally exhibit complicated potential energy surfaces (PES's) that present several significant minima, and thus, an extensive exploration of the PES's must be performed in order to understand the overall shape of the surface. Since the local minimization techniques can favor some minima or disregard some others, one has to use nonlocal methods that require a direct and rapid calculation of the interactions in the system. In the traditional quantum chemistry approach, the calculation of interactions between neutral molecules requires highly sophisticated wave functions, especially for the dispersive forces, and is thus too CPU time consuming for the systems we are interested in. With this approach, only limited parts of the PES can be explored. Furthermore, taking explicitly into account the basis set superposition error (BSSE), a well-known artifact of this technique, during the optimization process is not easy to carry out. For this reason, we have chosen an exploration strategy that combines both a semiempirical model,[29,30] especially devised for the calculation of intermolecular interactions, and a global minimization method[30c] based on the simulated annealing technique. Such a strategy already turned out to be successful for the study of several molecular clusters.[8,31] We will present here only its main features.

*4.1.a. Intermolecular Interactions.* In the model, all the important contributions to the interaction energy (electrostatic, polarization, repulsion, and dispersion) are taken into account. The different contributions are expressed by simplified analytical formulas, which are derived from expressions obtained at second order in the exchange perturbation treatment and which give a reliable description of interactions for all intermolecular distances.[29] The electrostatic term is the sum of all two-body multipole−multipole interactions. The polarization term is an *n*-body term that is the sum of the polarization energy of each molecule due to the field created by the multipoles of all the other molecules. The polarizabilities of centers of molecular subunits involved in the calculation of the polarization term are derived from mean bond experimental polarizabilities. The multipole distribution of each molecular subunit (a charge, a dipole, and a quadrupole on atoms and one point per chemical bond) is generated from the multipolar, multicentric development of its electronic distribution through a systematic procedure of the reduction of the number of centers.[32] This development is derived from the wave function obtained via an ab initio calculation in which an extended basis set has to be used and electronic intramolecular correlation must be introduced.[30] In the present calculations, the multipole distributions of the indole, 1-methylindole, and water molecules are obtained using the basis sets and level of theory indicated in ref 8, which gives a good agreement between the calculated and experimental permanent dipole moments of molecules. In this way, we expect that the electrostatic interactions and electric fields are calculated very accurately. The repulsion and dispersion terms are sums of atom−atom terms. The repulsion term takes into account the variation of the electronic population of each atom in the molecules and its influence on the van der Waals radius. The dispersion term includes contributions up to $C_{10}/R^{10}$ as well as an exchange contribution.

*4.1.b. Exploration of the Potential Energy Surface.* The six intermolecular coordinates used to describe the relative positions of the two molecules are the three Euler angles for the rotation and the three coordinates of translation (position of the mass center). The molecules are assumed to be frozen in their equilibrium geometries. The procedure for finding the PES minima is an extension of the simulated annealing method. First of all, a random search on the surface is performed by the Metropolis algorithm.[33] Second, the conformations obtained from this exploration are sorted out. Finally, the resulting conformations are optimized by a local minimization method (quasi- Newton method, BFGS[34]), each minimum being checked by the Hessian. For the determination of the saddle point between minima, the method developed by Liotard[35] has been used. It consists of drifting on the surface path connecting the two minima, the starting path being generated from an estimated saddle point and the two minima considered. The energetic relaxation of the path to the highest point that can no more be relaxed leads to the determination of the saddle point. Owing to the local character of the method, several starting paths are needed in order to assess the reliability of the result.

**4.2. Results of the Indole−Water System.** Two pairs of minima are found, with similar geometries and structures within each pair. The structure of the conformations in the first pair (well depth $D_e$ of 4.99 and 4.78 kcal/mol, respectively) is found to involve the conventional H-bond, the indole molecule acting as proton donor via its NH group and the water molecule as proton acceptor via its oxygen atom (parts a and b of Figure 4, respectively). The hydrogen bond is found to be nearly translinear, and the plane of the water molecule is perpendicular to the plane of the indole molecule. The two structures differ mainly by the orientation of the plane of the water molecule with respect to the N−H bond. The two other minima found ($D_e$ of 4.27 and 4.20 kcal/mol, respectively) correspond to a structure in which the water molecule is located above the plane of indole, the planes of both molecules forming an angle of 68° for the first minimum and 82° for the second one (parts a and b of Figure 5, respectively). These structures correspond to the nonstandard hydrogen bond ($\pi$-type) and differ mainly in the ring where the molecule of water is located. In the first minimum, the oxygen atom of the water molecule stands above the five-membered ring with only one hydrogen atom prefer-

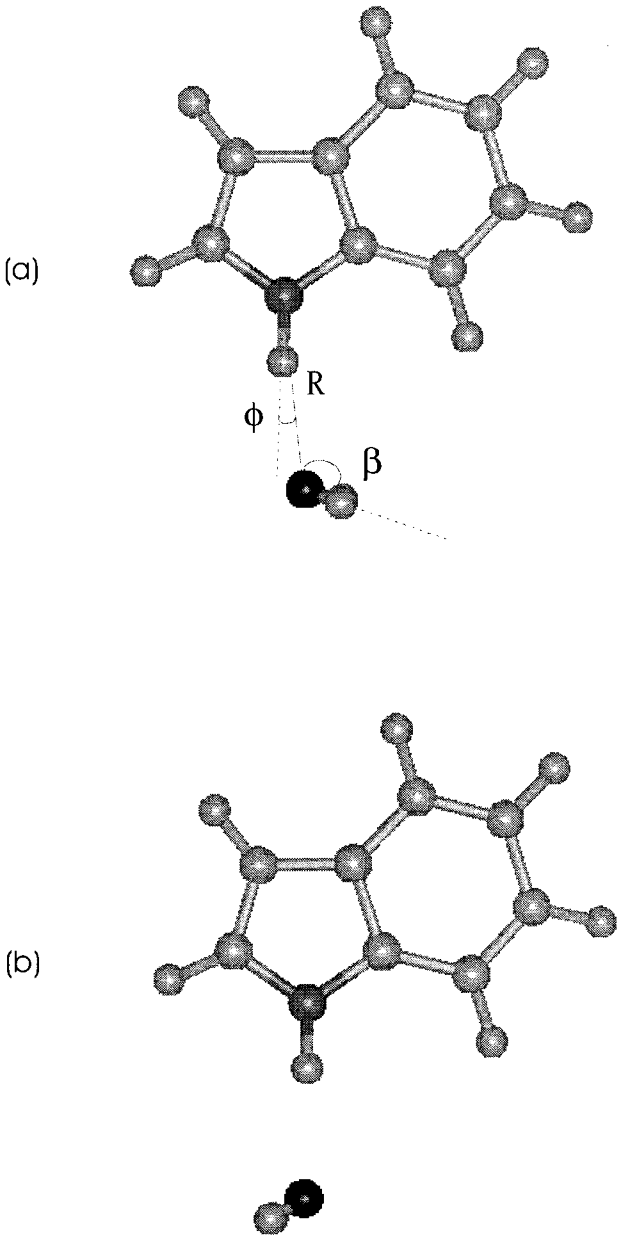

**Figure 4.** Calculated structures (top views) of the two H-bond minima of the indole−water complex: (a) $R = 3.00$ Å, $\phi = 7°$, $\beta = 114°$ and (b) $R = 3.02$ Å, $\phi = 4°$, $\beta = 123°$.

entially pointing toward this ring, whereas in the second one, the oxygen atom stands above the six-membered ring with both H atoms pointing to it.

The similarity in the two pairs of minima is also suggested by the analysis of the components of the interaction energy. For the two most stable minima (H-bond), the electrostatics is responsible for a large part of the attractive interaction (65% in both cases) whereas dispersion represents only 24% and 23% of it, respectively (Table 2). In contrast, for the two $\pi$-type minima (Table 3), the percentages of the electrostatic and dispersive contributions are almost identical, 45% and 47% for the first minimum and 42% and 50% for the second one, providing evidence for the hybrid character of this $\pi$-type hydrogen bond. Furthermore, for the H-bond minima that present strong local electrostatic interactions and therefore a few short interatomic distances, the sum of the dispersion−repulsion contributions is repulsive by about 2.00 kcal/mol whereas for the two $\pi$-type minima, this term is attractive by about 0.8 kcal/mol.

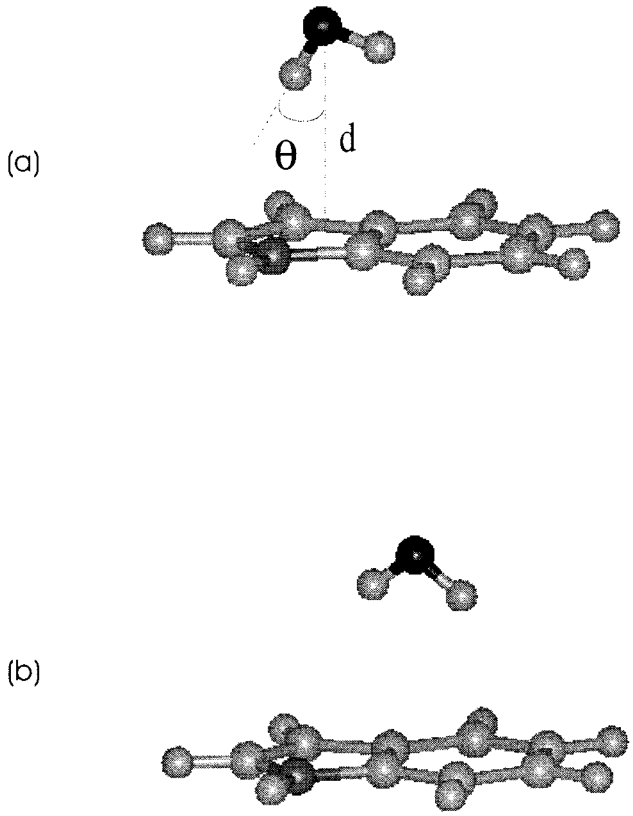

**Figure 5.** Calculated structures (side views) of the two $\pi$-type minima of the indole−water complex: (a) $d = 3.19$ Å, $\theta = 33°$ and (b) $d = 3.22$ Å, $\theta = 51°$.

**TABLE 2: Components of the Interaction Energy (kcal/mol) of the Conventional H-Bond Minima of the 1:1 Water Complexes with Indole and Phenol**

| | components of the interaction energy | | | | |
|---|---|---|---|---|---|
| | electrostatic | repulsion | polarization | dispersion | total |
| indole−water | | | | | |
| min 1 | −5.91 | 4.17 | −1.00 | −2.25 | −4.99 |
| min 1′ | −5.73 | 3.98 | −1.00 | −2.03 | −4.78 |
| phenol−water | −6.60 | 4.45 | −0.96 | −2.29 | −5.40 |

In a further attempt to characterize the indole−water PES, the saddle points between several minima have been determined. The energy of the saddle point connecting the two H-bond minima is found to be only 0.08 kcal/mol higher than that the least stable minimum. A still smaller barrier ($3 \times 10^{-4}$ kcal/mol) is found for the saddle point between the two $\pi$-type minima. Thus, these barriers are of the order of or lower than the accuracy of the calculation and are obviously not significant. In contrast, the saddle point corresponding to the isomerization of the more stable $\pi$-type H-bonded complex toward the more stable H-bonded one is found to be significant: 0.63 kcal/mol.

These results thus suggest a picture of the PES surface as made of two large attraction wells separated by a significant barrier, one of them corresponding to the conventional H-bonded complex, with a shallow profile along the wagging coordinate of the water molecule, and the other one, located 0.72 kcal/mol above, corresponding to the nonstandard H-bonded complex with a shallow profile for the displacement of the water molecule above the two rings.

**4.3. Results of the 1-Methylindole−Water System.** In this system, only one minimum is found (Figure 6). It corresponds to a $\pi$-type structure very similar to that found for the deepest well of the indole−water complex (Figure 5a). The water

**TABLE 3: Components of the Interaction Energy (kcal/mol) of the Nonstandard H-Bond Minima of the 1:1 Water Complexes with Indole, 1-Methylindole, and Benzene**

| | components of the interaction energy | | | | |
|---|---|---|---|---|---|
| | electrostatic | repulsion | polarization | dispersion | total |
| indole−water | | | | | |
| min 2 | −3.08 | 2.60 | −0.58 | −3.21 | −4.27 |
| min 2′ | −2.65 | 2.10 | −0.51 | −3.14 | −4.20 |
| 1-methylindole−water | −3.18 | 2.60 | −0.58 | −3.31 | −4.47 |
| benzene−water | −1.96 | 2.08 | −0.47 | −2.95 | −3.30 |

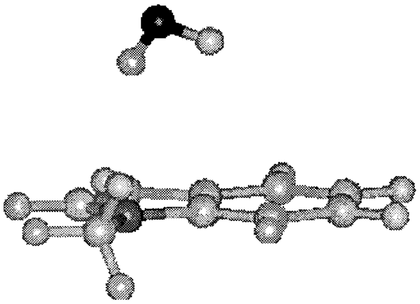

**Figure 6.** Calculated structure (side views) of the $\pi$-type minimum of the 1-methylindole−water complex: $d = 3.19$ Å and $\theta = 36°$.

molecule is located above the five-membered ring of the 1-methylindole, with one hydrogen atom preferentially directed toward this ring, the planes of both molecules forming an angle of 68°. This system does not exhibit any minimum corresponding to a position of the water molecule above the six-membered ring. The total interaction energy is −4.47 kcal/mol, and the contribution of the electrostatic and dispersive forces to the overall attractive interactions are 45% and 47%, respectively (Table 3). As a striking fact, the two features considered, the interaction energy and its components, are very similar to those found for the $\pi$-type minimum of the indole−water complex. The electrostatic and dispersion contributions differ only by 0.10 kcal/mol. The picture brought by this analysis suggests that methylation of the indole does not influence significantly the geometry or the energetics of the system, which validates our approach of the 1-methylindole−water system as a model system for the nonstandard H-bonded conformer of indole−water.

## 5. Discussion

**5.1. Theoretical Results of the 1:1 Water Complexes of Indole and 1-Methylindole.** The comparison between the calculated values of the energetic data is unfortunately not straightforward. Indeed, our experience in similar systems,[8,20] namely, phenol−water and benzene−water, suggests that the present semiempirical model gives fair values for nonstandard H-bonded complexes but significantly underestimates the well of the H-bonded complexes. In addition, ab initio calculations can provide very different results depending on the theory level they have been performed at.[36] We have nevertheless tried to compare the present calculations with those reported in the adjoigning paper by Carney and Zwier[13] (Table 1). A fair agreement is reached for the well depth of indole−water, but the above consideration suggests that these binding energies are underestimated. For the 1-methylindole−water complex, a significant disagreement is observed, which suggests that the modest basis set used in the ab initio calculation is not suitable to obtain reliable energetic data on the system. In both calculations, however, the same type of equilibrium geometry is found for the lowest conformation of each system, in particular, an intermolecular H···O distance of 1.97 and 2.01 Å is found in the ab initio calculations and the semiempirical model, respectively, for the NH···O bond of indole−water.

**5.2. Comparison of the Theoretical Results of the 1:1 Water Complexes of Indole and Phenol.** Our previous work on the phenol complexes has shown that the semiempirical model gives reliable relative well depths for complexes of similar nature, even if the absolute values might be only rough estimates. We thus believe that the difference found in the well depth between the H-bonded phenol−water and indole−water complexes is quite significant. Moreover, the measured binding energies also exhibit a difference that cannot be explained only by the variation of the zero-point energy (ZPE) owing to the similarity between these two complexes. Taking advantage of the possibility offered by the semiempirical model to provide a detailed analysis of the nature of the interaction energy within the complex, Table 2 shows that the dispersion and polarization forces are equivalent and that the difference is due to the contribution of the electrostatic and repulsion forces that are both more important in the phenol complex. A more detailed analysis in which the contribution of each subunit can be individualized shows that, in both complexes, the major part of the electrostatic and repulsion forces are due to the OH group in the phenol complex (−5.00 and 4.12 kcal/mol) and to the NH group in the indole complex (−5.34 and 4.07 kcal/mol). The rest of the aromatic molecule accounts only for −1.60 and 0.33 kcal/mol, respectively, with phenol and for −0.57 and 0.10 kcal/mol with indole, which shows that additional interactions, involving hydrogen atoms of the ring in particular, are much larger with phenol than with indole.

**5.3. Comparison of the Theoretical Results of the 1:1 Water Complexes of Indole, 1-Methylindole, and Benzene.** The structure of the $\pi$-type minimum of the indole−water complex where the molecule of water stands above the six-membered ring (Figure 5b) is very similar to the structure determined for the only minimum of the benzene−water complex.[20] Indeed, the minimum of the benzene−water complex corresponds to a structure where the water is located above the benzene ring in such a way that the oxygen atom stands on the benzene $C_6$ axis, the planes of both molecules being perpendicular and the geometrical parameters $\theta$ and $d$ being equal to ±45° and 3.22 Å. However, the total interaction energy of this complex is much less larger than that of the complex with indole: −3.30 kcal/mol compared to −4.20 kcal/mol. This difference can be explained by the analysis of the different contributions of the interaction energy (Table 3). The major part of this difference, 0.70 kcal/mol, is due to the electrostatic contribution, which is larger in the indole−water complex. The repulsion and the polarization contributions are identical, and the dispersion contributions differ only by 0.20 kcal/mol. A similar comparison between the two $\pi$-type minima of the indole−water complex shows that the similarity of their interaction energies is due to a subtle compensation between two contributions, the electrostatic and the repulsion, which vary in an opposite way in these two minima. These variations reflect directly the changes observed in the orientation of the molecule of water in these two minima. When the water molecule stands above the six-membered ring, the electrostatic contribution seems to be a maximum for the two hydrogen atoms directed toward the ring. Now, when the water molecule stands above the five-membered ring, the electrostatic contribution seems to be a maximum and larger for only one hydrogen atom preferentially directed toward the ring and then the repulsion contribution is increased. The 1-methylindole−water complex presents a minimum in both energy and structure that is very close to the most stable $\pi$-type minimum of the complex with indole. The difference in the interaction energy with respect to the benzene−water complex (−1.17 kcal/mol) is then mainly explained by the larger electrostatic contribution (−1.22 kcal/mol), which is not completely compensated by the larger repulsion term (+0.52 kcal/mol), and to a small extent by the larger dispersion contribution (−0.36 kcal/mol).

**5.4. Comparison between Experiment and Theory for the 1:1 Water Complexes of Indole and 1-Methylindole.** *Structures*. The conventional H-bond structure theoretically found for the most stable minimum of the indole−water complex is in line with the experimental data available, namely, resonant ion-dip infrared spectroscopy (RIDIRS)[12] and rotationally resolved UV spectroscopy[14,16] data, which both predict a hydrogen bond structure where the indole molecule acts as a proton donor. However, the present calculation that gives a NH···O bond of 2.01 Å corresponding to a N···O heavy-atom separation of 3.00 Å, together with the ab initio results of Carney and Zwier (1.97 and 2.98 Å, respectively, for NH···O and N···O distances[13]), are in agreement with the N···O value, 3.07 Å, given by Pratt and co-workers[16] but in total disagreement with the NH···O bond value given by Neusser and co-workers, namely, 2.93 Å.[14] This surprisingly large value appears to be also very different from the distances found both experimentally and theoretically in a similar H-bonded system, namely, phenol−water,[5−8] which suggests that the analysis of the rotational structure reported by these authors has to be reconsidered.

The nonstandard H-bond structure found theoretically for 1-methylindole−water is in qualitative agreement with the relatively small spectral shifts, the nonadiabatic ionization potential measured, the high fragmentation ratios observed after 1C photoionization, normally expected for these structures,[20] and finally the decisive resonant ion-dip infrared spectroscopy data reported by Carney and Zwier in the adjoining paper.[13]

The theoretical evidence for the existence of a second minimum for the indole water complex is not supported by the experiment. Generally speaking, the relaxation between conformers depends on both the interconversion barrier and the energy difference between forms. Microwave-based experiments have shown that room-temperature (or slightly lower) ratios are usually observed for rotamers of molecules such as alcohols in an helium jet and that, with heavier gases, barriers lower than 400 $\text{cm}^{-1}$ (1.1 kcal/mol) are needed for relaxation to occur.[37] This result is nevertheless not general, since, very recently, aryl alcohols (having therefore similar barriers to internal rotation about the C−O bond) have been found to relax very efficiently to specific rotamers in helium.[38] These considerations suggest that in the present system both the low barrier to isomerization (0.62 kcal/mol) and the significant energy difference between the species (0.74 kcal/mol) compared to $kT$ at room temperature (0.58 kcal/mol) contribute together to favor the relaxation process to the lowest-energy structure.

*Energetics*. A quantitative comparison between experiment and semiempirical calculations requires the knowledge of the zero-point energy (ZPE) in order to evaluate $D_0$ from $D_e$. For this purpose, we have chosen to take the ab initio ZPE value calculated by Carney and Zwier,[13] and the results are given in Table 1. As already mentioned, the quantitative comparison between experiment and theory is often disappointing. Both ab initio and semiempirical models fail to reproduce quantitatively the experimental data for the indole−water complex. Better results are obtained by the semiempirical calculation for the 1-methylindole−water complex, in line with the fair agreement already reported in the case of the similar benzene−water complex.

## 6. Summary and Concluding Remarks

**H-Bonded Systems: Phenol vs Indole.** The indole−water complex appears to be a conventional "translinear" H-bonded species that compares well both in shape and in energetics with another conventional H-bonded species, namely, phenol−water. The two species exhibit binding energies in the same energy range (5.60 ± 0.11 kcal/mol with phenol). The larger value for the phenol complex can be explained at least partially by the ZPE; the rest is likely to be due to the extra interaction between the water molecule and the phenol ring, as suggested by the analysis of the $D_e$ values in section 5.2. Indeed, the calculated ZPE value given by Carney and Zwier is 1.43 kcal/mol [13] for indole−water compared to 1.7 kcal/mol for phenol−water.[36] Thus, the difference in ZPE between the two species is much less than the difference between the measured binding energies (0.76 kcal/mol).

**Nonstandard H-Bonds: 1-Methylindole−Water and the Second Minimum of Indole−Water vs Benzene−Water.** The 1-methylindole−water structure is theoretically found to be very similar to the unobserved second minimum of indole−water. This structure is reminiscent of the benzene−water structure, the so-called $\pi$-type hydrogen bond in which the indole molecule is a proton acceptor via its aromatic cloud. The analysis of the components of the interactions in the 1-methylindole−water and the second minimum of indole−water systems shows that electrostatic and dispersion forces are the main attractive components and that they have the same order of magnitude. The influence of the change of the permanent dipole moment between indole and 1-methylindole is negligible as well as the increase of dispersion interaction due to the presence of the methyl group because of the relatively large distance between this group and the water molecule. This result allows us to validate our initial assumption and thus to consider the binding energy value obtained for 1-methylindole−water as a reliable estimate of the binding energy of a water molecule above the indole ring.

The experimental binding energy of 1-methylindole−water is much larger than that found for the benzene−water system (2.44 ± 0.09 kcal/mol with $H_2O$ and 2.67 ± 0.11 kcal/mol with $D_2O$).[20] The analysis suggests that this difference is explained by a larger electrostatic contribution to the interaction energy in the complex with the indole.

**Comparison between the Binding Sites of Water to Indole.** The present article allows us to propose both experimentally and theoretically an estimate of the interaction energy of a water molecule for its two main binding sites around the indole molecule: the conventional and the nonstandard hydrogen bonds. The former of these sites corresponds to the gas-phase complex, as observed in the supersonic expansion. The other one, a less bound conformer, is not observed experimentally, and its energetics is obtained by comparison with the gas-phase complex of the *N*-methylated indole molecule. This strategy, successful in the case of indole, can also be applied to other similar cases, like for instance the binding site (not observed experimentally in the gas phase) of a water molecule to the O atom of phenol, with this oxygen atom playing the role of proton acceptor.

As a surprising result, the binding energy of the site in which water is above the $\pi$ aromatic cloud of indole is nearly as strong as the conventional H-bond. The energy difference is small (24 $\pm$ 8 meV or 0.55 $\pm$ 0.18 kcal/mol, if one takes the value of ref 15 for indole−water) relative to the binding energy itself; the $\pi$-type hydrogen bond is only 12% less strong than the conventional hydrogen bond. In other words, the binding energy of a water molecule to indole, and presumably around the side chain of the tryptophan, is not very dependent on the location of the water, even if the nature of the binding differs totally.

The present gas-phase energetic study suggests that although traditionally assumed to have hydrophobic character, a tryptophan residue can establish several types of strong interactions with water. These characteristics might play a role in the amphiphilic properties of the tryptophan residue invoked to interpret, on a microscopic scale, biological processes such as protein membrane translocation.

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## References and Notes

(1) Perutz, M. F. *Philos. Trans. R. Soc. London A* **1993**, *345*, 105.
(2) Zwier, T. S. *Annu. Rev. Phys. Chem.* **1996**, *47*, 205.
(3) Thornton, J. M.; MacArthur, M. W.; McDonald, I. K.; Jones, D. T.: Mitchell, J. B. O.; Nandi, C. L.; Price, S. L.; Zvelebil, J. J. M. *Philos. Trans. R. Soc. London A* **1993**, *345*, 113.
(4) Tanabe, S.; Ebata, T.; Fujii, M.; Mikami, N. *Chem. Phys. Lett.* **1993**, *215*, 347.
(5) Berden, G.; Meerts, W. L.; Schmitt, M.; Kleinermanns, K. *J. Chem. Phys.* **1996**, *104*, 972.
(6) Gerhards, M.; Schmitt, M.; Kleinermanns, K.; Stahl, K. *J. Chem. Phys.* **1996**, *104*, 967.
(7) Helm, R. M.; Vogel, H.-P.; Neusser, H. J. *J. Chem. Phys.* **1998**, *108*, 4496.
(8) Courty, A.; Mons, M.; Dimicoli, I.; Piuzzi, F.; Brenner, V.; Millié, P. *J. Phys. Chem. A* **1998**, *102*, 4890.
(9) Burgi, T.; Droz, T.; Leutwyler, S. *Chem. Phys. Lett.* **1995**, *246*, 291.
(10) Hager, J. W.; Ivanco, M.; Smith, M. A.; Wallace, S. C. *Chem. Phys.* **1986**, *105*, 397.
(11) Huang, Y.; Sulkes, M. *J. Chem. Phys.* **1996**, *100*, 16479.
(12) Carney, J. R.; Hagmeister F. C.; Zwier, T. S. *J. Chem. Phys.* **1998**, *108*, 3379.
(13) Carney, J. R.; Zwier, T. S. *J. Phys. Chem. A* **1993**, *103*, 9943.
(14) Helm, R. M.; Clara, M.; Grebner, Th. L.; Neusser, H. J. *J. Phys. Chem. A* **1998,** *102*, 3268.
(15) Braun, J. E.; Grebner, Th. L.; Neusser, H. J. *J. Phys. Chem. A* **1998,** *102*, 3273.
(16) Korter, T. M.; Pratt, D. W.; Kuepper, J. *J. Phys. Chem. A* **1998,** *102*, 7211.
(17) Pribble, R. N.; Zwier, T. S. *Science* **1994**, *265*, 75.
(18) Gruenloh, C. J.; Carney, J. R.; Arrington, C. A.; Zwier, T. S.; Fredericks, S. Y.; Jordan, K. D. *Science* **1997**, *276*, 1678.
(19) Cheng, B. M.; Grover, J. R.; Walters, E. A. *Chem. Phys. Lett.* **1995**, *232*, 364.
(20) Courty, A.; Mons, M.; Dimicoli, I.; Piuzzi, F.; Gaigeot, M.-P.; Brenner, V.; de Pujo, P.; Millié, P. *J. Phys. Chem. A* **1998**, *102*, 6590.
(21) Makhatadze, G. I.; Privalov, P. L. *J. Mol. Biol.* **1990**, *213*, 375 and references therein.
(22) Jacobs, R. E.; White, S. H. *Biochemistry* **1989**, *28*, 3421.
(23) Schiffer, M.; Chang, C.-H.; Stevens, F. J. *Protein Eng.* **1992**, *5*, 213.
(24) Abel, E.; Fedders, M. F.; Gokel, G. W. *J. Am. Chem. Soc.* **1995**, *117*, 1265.
(25) Uridat, D.; Brenner, V.; Dimicoli, I.; Le Calvé, J.; Millié, Ph.; Mons, M.; Piuzzi, F. *Chem. Phys.* **1998**, *239*, 151.
(26) Hager, J. W.; Wallace, S. C. *Anal. Chem.* **1988**, *60*, 5.
(27) Guesten, H.; Klasinc, L.; Ruscic, B. *Z. Naturforsch.* **1976**, *31a*, 1051.
(28) Müller-Dethlefs, K.; Dopfer, O.; Wright, T. G. *Chem. Rev.* **1994**, *94*, 1847.
(29) (a) Claverie, P. In *Intermolecular Interactions from Diatomics to Biopolymers*; Pullman, B., Ed.; Wiley: New York, 1978. (b) Hess, O.; Caffarel, M.; Langlet, J.; Caillet, J.; Huiszoon, C.; Claverie, P. In *Proceedings of the 44th International Meeting on Modelling of Molecular Structures and Properties in Physical Chemistry and Biophysics*, Nancy, France, Sept 11−15, 1989; Rivail, J. L., Ed.; Elsevier: Amsterdam, 1990.
(30) (a) Brenner, V.; Millié, Ph. *Z. Phys. D* **1994**, *30*, 327. (b) Millié, Ph.; Brenner, V. *J. Chim. Phys*. **1995**, *92*, 428. (c) Brenner, V. Thèse de l'Université de Paris-sud, 1993.
(31) (a) Brenner, V.; Martrenchard, S.; Millié, Ph.; Jouvet, C.; Lardeux-Dedonder, C.; Solgadi, D. *J. Phys. Chem.* **1995**, *99*, 5848. (b) Le Barbu, K.; Brenner, V.; Millié, Ph.; Lahmani, F.; Zehnacker-Rentien, A. *J. Phys. Chem. A* **1998**, *102*, 128.
(32) Vigne-Maeder, F.; Claverie, P. *J. Chem. Phys.* **1988**, *88*, 4934.
(33) Metropolis, N.; Rosenbluth, A.; Rosenbluth, M.; Teller, A.; Teller, E. *J. Chem. Phys.* **1953**, *21*, 1087.
(34) (a) Flether, R. *Comput. J.* **1970**, *13*, 317. (b) Goldfarb, D. *Math. Comput*. **1970**, *24*, 23. (c) Shanno, D. F. *Math. Comput*. **1970**, *24*, 647.
(35) Liotard, D. *Int. J. Quantum Chem*. **1992**, *44*, 723.
(36) Schutz, M.; Burgi, T.; Leutwyler, S. *J. Chem. Phys.* **1993**, *98*, 3763.
(37) Ruoff, R. S.; Klots, T. D.; Emilsson, T.; Gutowsky, H. S. *J. Chem. Phys.* **1990**, *93*, 3142.
(38) Mons, M.; Robertson, E. G.; Snoek, L. C.; Simons, J. P. *Chem. Phys. Lett.*, in press.