Hydration of the Carboxylate Group in Anti-Inflammatory Drugs: ATR-IR and Computational Studies of Aqueous Solution of Sodium Diclofenac

Elena O. Levina,† Nikita V. Penkov,‡ Natalia N. Rodionova,§ Sergey A. Tarasov,§ Daria V. Barykina,§ and Mikhail V. Vener*∥

†Department of Molecular and Chemical Physics, Moscow Institute of Physics and Technology, 7 Institutskiy per., 141700 Dolgoprudny, Russia
‡Department of Methods of Optical and Spectral Analysis, Institute of Cell Biophysics, Russian Academy of Sciences, 3 Institutskaya Street, 142292 Pushchino, Russia
§OOO “NPF” Materia Medica Holding, 47-1 Trifonovskaya Street, 129272 Moscow, Russia
∥Department of Quantum Chemistry, Mendeleev University of Chemical Technology, 9 Miusskaya Square, 125047 Moscow, Russia

*Supporting Information

ABSTRACT: Diclofenac (active ingredient of Voltaren) has a significant, multifaceted role in medicine, pharmacy, and biochemistry. Its physical properties and impact on biomolecular structures still attract essential scientific interest. However, its interaction with water has not been described yet at the molecular level. In the present study, we shed light on the interaction between the steric hindrance (the intramolecular N···H···O bond, etc.) carboxylate group (−CO2−) with water. Aqueous solution of sodium diclofenac is investigated using attenuated total reflection-infrared (ATR-IR) and computational approaches, i.e., classical molecular dynamics (MD) simulations and density functional theory (DFT). Our coupled classical MD simulations, DFT calculations, and ATR-IR spectroscopy results indicated that the −CO2− group of the diclofenac anion undergoes strong specific interactions with the water molecules. The combined experimental and theoretical techniques provide significant insights into the spectroscopic manifestation of these interactions and the structure of the hydration shell of the −CO2− group. Moreover, the developed methodology for the theoretical analysis of the ATR-IR spectrum could serve as a template for the future IR/Raman studies of the strong interaction between the steric hindrance −CO2− group of bioactive molecules with the water molecules in dilute aqueous solutions.

1. INTRODUCTION

Molecular solute–solvent interaction studies are of paramount importance in gaining a deeper understanding of the biomolecular function and the fundamental mechanisms involved in the conformational adaptation in solutions.1 Physicochemical properties of this interaction are widely studied in various environments (i.e., solvents), especially in water—usually referred to as the solvent of life.2

NMR spectroscopy is an important technique for investigating biomolecular interactions and protein dynamics.3 It is commonly used in combined experimental and theoretical studies to examine the interactions between polar solvents and biomolecular functional groups,4 the influence of solvent dynamics on the structure of hydrogen-bonded complexes,5,6 and the dynamics of side-chain amino groups in an aqueous solution.7 The applicability of NMR in the investigation of the first hydration shell of the −CO2− groups in aqueous solutions is not straightforward.8

IR spectroscopy is another basic tool used for elucidating the biomolecular interactions and solvent–solute interaction studies.9,10 Special focus is generally given to the so-called amide I, II, III, and A regions,11 but sometimes other regions are also investigated.12 Due to the strong absorbance of water in the mid-infrared spectral region, the IR difference spectra are used. The attenuated total reflection-infrared (ATR-IR) mode after subtraction of the water spectrum gives the IR spectrum of the solute,13 if it does not form relatively strong hydrogen bonds (H-bonds)14 with water molecules. As for specific strong interactions, the water spectrum should be subtracted except a few water molecules that form relatively strong H-bonds with a solute molecule. Their number can be estimated if the solute is highly soluble in water. Spectral analysis of a series of highly
concentrated solutions of known density and solute’s mass fraction allows obtaining a stoichiometric ratio of solutions under study\(^5,16\) and, consequently, estimating the number of water molecules in the first hydration shell.

Some peculiarities of active pharmaceutical ingredients can strongly affect the above research methodology. The first one is low solubility. For instance, the maximum plasma concentration of anti-inflammatory drugs\(^57–59\) is usually lower compared to their solubility. This stipulates the possibility of using substances with rather low solubility\(^20,21\) for medical purposes. The second is a relatively strong interaction between some bioactive molecules and water molecules.\(^22–24\) Due to rather low solubility of sodium diclofenac (\(\sim 3.5 \times 10^{-3} \text{ mol L}^{-1}\) at \(\text{pH} = 7.0\)\(^25\)) and its relatively strong specific interaction with water molecules, the interpretation of the ATR-IR spectrum of its aqueous solution is not straightforward. Thus, an adequate description of the structure of the first hydration shell and spectral features of the aqueous solution of sodium diclofenac (NaDN) could be achieved by combining ATR-IR and theoretical studies.

Density functional theory (DFT) based MD simulations\(^56,27\) give a reliable description of the local structural motifs of the hydration shell, and IR spectra\(^29–31\) of the bioactive molecules in water and polar solvents. It should be noted that these simulations are at present computationally demanding for systems of modest sizes (a solute and \(\sim 50\) water molecules), and currently intractable for larger sizes (a solute and \(\sim 1000\) water molecules).\(^32,33\) Due to low solubility of NaDN, the ratio of 1 solute molecule to 1000 water molecules corresponds to the highest possible concentration of the aqueous solution of NaDN\(^27\) that makes ab initio MD simulations hardly applicable.

Classical molecular dynamics (MD), Monte Carlo simulations,\(^34–40\) and the RISM integral equation method\(^41,42\) led to valuable insights into the hydration structure of bioactive molecules in water. In general, these approaches allow to compute the IR spectra.\(^43,44\) If nonpolarizable classical force fields are used in the simulations to obtain the IR spectra, the results should be treated with some caution, e.g., refer to ref \(^45\).

The solvation phenomenon can be described using implicit solvent models.\(^46\) In particular, the discrete-continuum model\(^47\) is often used for the evaluation of the vibrational frequencies of molecules in water.\(^48\) A serious drawback of the model is the uncertainty in the minimum number of water molecules required to simulate the first hydration shell in diluted aqueous solutions.\(^30\) This number can be estimated from the classical MD simulations. Another significant limitation of the discrete-continuum model consists in the absence of specific interactions between the molecules forming the first hydration shell and the adjacent water molecules. To the best of our knowledge, no approach that might overcome this limitation has been proposed so far. The microsolvation model provides a semiquantitative description of the spectral properties and the hydration shell’s structure of bioactive molecules.\(^30–32\) We conclude that classical MD simulations and DFT computations of complexes of a bioactive molecule with several water molecules may be considered as complimentary.

Sodium diclofenac is a pharmaceutically active substance with pronounced anti-inflammatory, analgesic, antipyretic, and anticancer activities.\(^53\) Its molecular effects are associated with the formation of hydrogen bonds between the carboxyl group of diclofenac and side chains of Tyr-385 and Ser-530 present in COX.\(^54,55\) However, now sodium diclofenac has limited use because of various side effects (including gastrointestinal and cardiovascular ones)\(^56\) arising from its nonselective action on both cyclooxygenase isozymes (COX-1 and COX-2). Diclofenac has currently been under intensive development by the pharmaceutical industry with respect to its efficacy and safety, aimed at creating improved forms of diclofenac-based drugs. For example, a released active form of diclofenac is being studied as a possible option. It is produced by means of a special process, which results in the high efficacy of diclofenac in the released active form, with almost no side effects.\(^57\) Because the pharmaceutical effects of diclofenac primarily occur due to the hydrogen-bond interaction between its carboxyl group and the targets (Tyr-385 and Ser-530 of the COX enzymes), it is important to look into the involvement of this carboxyl group in other processes, such as the formation of the hydration shell of diclofenac. Therefore, understanding the physicochemical properties and features of the hydration shell formation is essential for developing a form of diclofenac that could sterically facilitate a higher pharmaceutical efficacy (and safety). The NMR studies of NaDN solutions were used to detect related complexes with cyclodextrin\(^58,59\) and with calix[4]arene.\(^60\) NMR and IR spectroscopy techniques were used to evaluate the quality of generic NaDN tablets.\(^61\) The IR spectrum of the aqueous solution of NaDN has not been published yet, based on the available data.

The intramolecular \(\text{N}−\text{H}−\text{O}\) bond and chlorine atoms present in the adjacent benzene ring (Figure 1 and ref \(^62\))

Figure 1. Structure of the \(\text{DN}^-\) ion with the intramolecular \(\text{N}−\text{H}−\text{O}\) bond. The latter is denoted by the dotted line. The red, blue, green, gray, and white circles represent oxygen, nitrogen, chlorine, carbon, and hydrogen atoms, respectively.
facilitates the formation of a realistic model of the microsolvation, subsequently used in the DFT computations. Our goal was to completely isolate the interaction between $-\text{CO}_2-$ of the DN$^-$ ion and water, allowing for a detailed analysis of the most stable complexes and, therefore, a deeper understanding of the strong interaction between the steric hindrance $-\text{CO}_2-$ group of bioactive molecules with water molecules in aqueous solutions.

2. RESULTS

2.1. ATR-IR Studies. The ATR-IR spectrum of the powder of NaDN is given in Figure 2. It agrees well with the available literature data.\textsuperscript{67} The ATR-IR spectrum of the NaDN aqueous solution (0.4%) is also given in Figure 2. The spectra vary very much in the frequency region above 1700 cm$^{-1}$. Obviously, this difference is due to the solute–solvent interaction. For instance, the broad band around $\sim 3200$ cm$^{-1}$ corresponds to the Ow–H stretching and is shifted to the low-frequency region, which is typical to the systems with H-bonds between solute and solvent.\textsuperscript{68} A specific feature of the ATR-IR spectrum is a number of narrow Evans-type transmission windows (the Evans holes). The holes located at around 1000 and 1250 cm$^{-1}$ are associated with perturbations between superposing energy levels.\textsuperscript{69,70} The hole at around 1750 cm$^{-1}$ has a different nature because the IR intense bands in the 1000 frequency region are absent in the powder spectrum (Figure 2, and Figure 1 in ref 67). It is analyzed in Section 3.

The ATR-IR spectra of 0.4% NaDN show a good signal-to-noise ratio (Figures 2 and S1). The concentration of this solution is about 0.01 mol L$^{-1}$, approaching the ATR-IR detection limit for aqueous solutions.\textsuperscript{71} Indeed, the IR spectra of solutions with concentration <0.1% differ negligibly from the water spectrum (Figure S2). However, a higher detection limit could be used for individual contributions to broad bands, particularly for water-stretching vibrations, such as those exhibited by/observed for the quasi-free OH groups.\textsuperscript{72} The depth of the Evans hole, located at around 1750 cm$^{-1}$, decreases with decreasing concentration, but it is clearly visible at concentration 0.2% (Figure S1). This phenomenon is discussed in the section below.

2.2. Classical MD Simulations and DFT Computations. The O$_1$–O$_w$ radial distribution functions (RDFs) and cumulative distribution functions (CDFs), where O$_1$ is the oxygen atom of the $-\text{CO}_2-$ group and O$_w$ is the water oxygen, obtained by MD simulations, are given in Figure 3a. According to the CDF, the average number of water molecules around the oxygen (atom/atoms) concerned is $\sim 3$, which agrees with the literature data.\textsuperscript{73} To estimate the average number of water molecules around the $-\text{CO}_2-$ group, the C–O$_w$ RDF and CDF are also computed (Figure 3b). The target value is $\sim 6$. According to ref 64, the carboxylate ion has about 7 strongly bound water molecules in the first hydration layer. The reasons for this difference are the steric hindrance caused by the formation of an intramolecular H-bond in the DN$^-$ anion (Figure 1), and variations in the mutual orientation of the benzene rings. The latter is discussed in Section 3.

In the next step, the microsolvation process of the DN$^-$ ion is studied using DFT computations. Metric parameters of the intramolecular N–H···O bond in the DN$^-$·nH$_2$O complexes, where n varies from 0 to 5, are given in Table 1. Due to the

![Figure 2. ATR-IR spectrum of the powder of NaDN vs the ATR-IR spectrum of its aqueous solution (0.4%) after subtraction of the water spectrum.](image)

![Figure 3. (a) O$_1$–O$_w$ radial distribution functions (RDFs) and cumulative distribution functions (CDFs) obtained for aqueous NaDN solutions ($\sim 1.7$ and $\sim 0.4$%), where O$_1$ is the oxygen atom of the $-\text{CO}_2-$ group and O$_w$ is the water oxygen. (b) C–O$_w$ RDF and CDF obtained for aqueous NaDN solutions ($\sim 1.7$ and $\sim 0.4$%), where C is the carbon atom of the $-\text{CO}_2-$ group and O$_w$ is the water oxygen.](image)
strength and targeted nature of intermolecular H-bonds, only a few low-energy structures for \( n > 2 \) are available. When \( n = 3 \) and 4, several conformers are localized; they differ by the mutual orientation of water molecules and the number of H-bonds (Figures S3 and S4). Increasing the number of the water molecules shifts the bridging proton from the oxygen to nitrogen atoms. This shift leads to an extension of the N···O distance and the distortion of the intramolecular H-bond. At \( n = 5 \), the intramolecular N−H···O bond becomes rather long (the N···O distance is larger than 2.75 Å), and strongly nonlinear (the N−H···O angle is ∼150°), see Table 1. At least one relatively short (strong) intermolecular O···H−Ow bond, where Ow is the oxygen atom of the water molecule, exists in the complexes with \( n > 1 \) (Table 1).

Table 1. Metric Parameters of the Intramolecular N···H···O Bond and the Shortest Intermolecular O···H−Ow Bond in the DN−·nH2O Complexes* Evaluated Using DFT Computations

| parameter          | \( n = 0 \) | \( n = 1 \) | \( n = 2 \) | \( n = 3 \) | \( n = 4 \) | \( n = 5 \) |
|-------------------|-------------|-------------|-------------|-------------|-------------|-------------|
| O···N, Å          | 2.627       | 2.547       | 2.722       | 2.737       | 2.782       | 2.808       |
| O−H (H−N), Å      | 1.030       | 1.071       | 1.041       | 1.037       | 1.031       | 1.028       |
| O−H−N, deg        | 166.4       | 167.9       | 154.6       | 153.4       | 151.1       | 150.2       |
| O−Ow, Å           | 2.916       | 2.713       | 2.704       | 2.658       | 2.658       | 2.686       |
| O−H−Ow, deg       | 157.7       | 173.6       | 175.5       | 176.5       | 176.5       | 175.8       |

*a* see Figure 4; ‘w’ denotes the oxygen atom of the water molecule. b at \( n > 1 \) the bridging proton locates near the nitrogen atom.

**Figure 4.** Global-minimum structures of the DN−·nH2O complexes: (a) \( n = 1 \); (b) \( n = 2 \); (c) \( n = 3 \); (d) \( n = 4 \); and (e) \( n = 5 \). Hydrogen bonds are highlighted with light blue dotted lines. Chlorine contacts are highlighted with red dotted lines. See the caption to Figure 1 for color coding.

According to the DFT computations, for \( n ≥ 2 \), two or three water molecules form a H-bond with the carboxyl group of the DN− ion (Figures 4 and S1). This is consistent with the results of MD simulations, which show that the average number of water molecules directly interacting with the oxygen atom of the −CO2− group is ∼3. Furthermore, the structure of H-bonded complexes demonstrated by the DFT computations agrees with the results of MD simulations (cf. Figures 4 and 5). Both cases indicate the occurrence of the R3(8) motif.
Figure 5. Snapshots of several H-bonded complexes of the DN\textsuperscript{−} ion and water molecules shown by the MD simulation of 0.4% aqueous solution. Trajectory length = 1 ns. The H-bonds are given by the dotted lines. The corresponding data for 1.7% solution is given in the Supporting Information (Figure S5). See the caption to Figure 1 for color coding.
Theoretical IR spectra of the DN⁻·3H₂O complex corresponding to the global-minimum structure with a zero-point energy correction are used for a tentative assignment of the ATR-IR spectrum of the aqueous solution of sodium diclofenac (Table S2). This approach gives reasonable description of the most IR intensive bands in the experimental spectrum (Figure 6). To interpret several low-intensive bands, the literature data should be used. The low intensive band at around 2330 cm⁻¹ is associated with the asymmetric stretch of the CO₂ molecules, which are absorbed by the solution from the air. The weak broad band near 2200 cm⁻¹ is caused by the asymmetric stretching vibration of the N−H−O bond.86−88 The wagging vibrations of water molecules may indicate the presence of the H-bonded water.88 The bending and wagging vibrations of water molecules may indicate the presence of the H-bonded water.88 The bending vibration of the first molecule and the wagging vibration of the second molecule shift to blue in comparison with the corresponding vibrations of the “free” water molecule, in accord with the literature data.89 Overtone of the wagging vibration is superposed with the bending vibration caused the hole in the IR spectrum. We attributed this hole to the relatively strong specific solute–solvent interaction. The point is that the R₃(8) motif does exist in the local-minimum structures of the DN⁻·3H₂O and DN⁻·3H₂O complexes (Figures S3 and S4).

Crystallographic data are a valuable source of information on the energy and geometric features of noncovalent interactions.90−92 Screening of the Cambridge Structure Database93 shows that the considered R₃(8) motif occurs in 31 crystals, including pharmaceutical solvates and co-crystals, e.g., norflaxacin dihydrate,94 sparflaxin trihydrate,95 and enrofloxacin dioxalate hexahydrate.96

Another specific feature of NaDN hydration is the steric hindrance effect. The MD simulations show that the quasi-linear N−H−O bond occurs frequently in the DN⁻ ion (the N−O distance is less than 2.75 Å, the N−H−O angle is greater than 160°) during MD simulations at both concentrations concerned (see Figure 8). The formation of intramolecular N−H−O bonds of ring size 7 is quite a rare event.98−99 Usually, intramolecular N−H−O bonds of ring size 6100 with the strongly nonlinear N−H−O moiety are presented.

Mutual orientation of benzene rings might have a significant role in the COX-2 receptor inhibition.101 According to ref 62, the structure of diclofenac, which is able to inhibit COX-2

### Table 2. Theoretical Values of Metric Parameters of the Intermolecular H-Bonds Forming the Eight-Member Ring in the DN⁻·nH₂O Complexes,

Where n = 2, 3, 4, and 5

| R(0−Ow1) Å | R(0−Ow2) Å | R(0−Ow3) Å | R(Ow1−Ow2) Å |
|------------|------------|------------|--------------|
| 2.892 (169.8) | 2.704 (175.5) | 2.658 (176.5) | 2.686 (175.8) |
| 2.713 (173.6) | 2.789 (174.1) | 2.902 (165.2) | 2.922 (164.0) |
| 2.971 (157.4) | 2.890 (155.6) | 2.944 (149.4) | 2.946 (150.0) |

| σ(H−Ow1−H₃c) cm⁻¹ | σ(H−Ow2−H₃c) cm⁻¹ | σ(H−Ow3−H₃c) cm⁻¹ | σ(H−Ow1−H₃c) cm⁻¹ |
|-----------------|-----------------|-----------------|-----------------|
| 1717 (71) | 1682 (174) | 1671 (150) | 1671 (92) |
| 1690 (278) | 1737 (77) | 1730 (73) | 1726 (68) |
| 738 (136) | 902 (181) | 939 (127) | 901 (126) |
| 939 (87) | 823 (177) | 816 (157) | 801 (202) |

*See Figure 8. *Harmonic frequencies and IR intensities of the selected vibrations. σ and ω denote the bending and wagging vibrations, respectively. *IR intensities (kM mol⁻¹) are given in parentheses. *Strongly coupled to σ(H−Ow1−H₃c).
(with the C−C−N−C dihedral angle of the −60°, see Figure 2 in ref 62), is not stable in the gas-phase ab initio simulations. In the case of MD simulations of water solutions, various C−C−N−C dihedral angles were observed (see Figure S6), including oscillations around −60°. For a more accurate description of this effect on −CO2− solvation, which is beyond the scope of this study, further research is needed.

Very recently, the IR difference spectrum for the amide A band of Polyamide 6 has been evaluated using the MD simulations and quantum vibrational calculations. The combined theoretical approach has been successfully applied for the analysis of the hydration structures of the title compound. We conclude that the combined experimental and theoretical techniques provide significant insights into the spectroscopic manifestation of strong solute−water interactions and the structure of the hydration shell of bioactive molecules.

Figure 7. Eight-member ring in the global-minimum structure of the DN−·nH2O complexes: (a) n = 2, (b) n = 3, and (c) n = 4 and 5. H-bonds forming the R3(8) motif are given by dotted lines.

Figure 8. Formation of the intramolecular N−H⋯O bond in the DN− ion during the MD simulations for its 0.4% (upper panel) and 1.7% (lower panel) aqueous solutions. The trajectory lengths are 10 ns in both cases. Pairs of heavy atoms forming the H-bond are indicated on the right.
4. CONCLUSIONS

The classical MD simulations used in combination with the DFT computations in the microsolvation limit demonstrate a specific characteristic of the hydration shell structure of the carboxylate (−CO2−) group in the diclofenac anion. It is an eight-atom ring formed by two water molecules, and the −CO2− group linked by three intermolecular H-bonds (the R3(8) motif). The motif is generated by steric hindrance resulting from the structure of diclofenac anion (the intra-molecular N−H···O bond, etc.). Overtones of wagging vibrations in water molecules, which form the eight-atom ring, superpose the bending vibration of the water molecule, which forms the shortest H-bond with the −CO2− group. As a result of Fermi resonance, a deep hole is found in the ATR-IR spectrum of the aqueous solution of sodium diclofenac at approximately 1750 cm−1. We attribute this to the relatively strong specific interaction of water with the steric hindrance −CO2− group.

Screening of the Cambridge Structure Database shows that the R3(8) motif occurs in solvates and co-crystals of pharmaceutical compounds, which contain the steric hindrance −CO2− group. We do hope that the methodology developed for theoretical analysis of ATR-IR spectra could provide a model for further IR/Raman studies of the strong interaction between the steric hindrance −CO2− group of bioactive molecules and water molecules in dilute aqueous solutions.

5. EXPERIMENTAL SECTION

ATR-IR spectra were recorded with a Nicolet 6700 infrared Fourier spectrometer equipped with an attachment (Patent U.S. 5,172,182) with a zinc selenide crystal—single-reflection ATR accessory. The IR spectra were detected in the 550−3500 cm−1 range with the resolution of 8 cm−1. The concentration of the aqueous solution of NaDN varies from 0.4 to 0.02%. Each sample was measured in triplicate, with each measurement repeated five times. The choice of the upper concentration was determined by the following reasons. First, at concentrations close to the solubility limit, the dissolution was frequently incomplete, which could have led to a distortion of the spectrum due to sample inhomogeneity. Second, the problem with incorrect comparisons of spectra with substantially different concentrations might be due to differences in the refractive indexes. At the target concentrations, the refractive indexes were almost identical between experimental samples and as compared with the refractive index of water.

The measurement procedure details are given in the corresponding paragraph of the Supporting Information.

6. COMPUTATIONAL METHODS

6.1. Molecular Dynamics Simulations. The GROMACS code103−106 was used to perform the MD simulations. The MD simulations were performed for two systems corresponding to available experimental concentrations.107 The first one included 1 molecule of NaDN and 4400 water molecules (~0.4%) placed in a cubic cell. The other used 1 molecule of NaDN and 1000 water molecules (~1.7%) placed in cubic cell. For NaDN, the potentials based on the parameters of the force field OPLS-AA108 were used together with the SPC/E water model.109 Atomic partial charges and van der Waals parameters of DN anion are given in Table S1 of the Supporting Information. The simulations were carried out in the NVT (constant number, constant volume, and constant temperature) ensemble. The volumes of cubic cells were chosen according to experimental densities of 1.7 and 0.4% aqueous solutions of sodium diclofenac (1.00475 and 1.00040 g mL−1, respectively). The temperature maintained at 298 K employing the velocity-rescaling temperature coupling110 with the time constant of 0.5 ps. The equations of motion were integrated using the leap-frog algorithm111 with a time step of 0.5 fs. Long-range electrostatic interactions were calculated using the particle mesh Ewald method112,113 (the cutoffs were set at 15 Å for ~1.7% solution modeling and 25 Å for ~0.4% solution); van der Waals and short-range interactions were truncated at 14 Å for ~1.7% solution and 24 Å for ~0.4% solution. The fluctuations of kinetic, potential, and total energy around some mean values serve as a criterion of the equilibration of the systems. For information purposes, 10 ns (a time step was 0.5 fs) simulations were performed.

6.2. DFT Computations. The structure and IR spectrum of the DN− anion and its complexes with nH2O molecules, DN−·nH2O, where n varies from 0 to 5, were computed at the B3LYP/6-311++G** level using Gaussian 03 software package.114 This level of approximation is efficiently used to establish a relation between experimental spectra and the underlying conformations of bioactive compounds in the microsolvation limit.51 The scaled factors115 have not been used for harmonic frequencies. The starting structures of complexes with a particular n were borrowed from the literature.65 London dispersion interactions were taken into account by using the B97D/6-311++G** approximation.116 In accord with the literature data,117,118 these interactions were found to have little influence on the geometrical and IR spectroscopic parameters of intermolecular H-bonds in the target/analyzed complexes.

The energy of O···Cl and H2O···Cl noncovalent interactions Eint was evaluated by the Bader analysis of electron density119 in conjunction with the Espinosa scheme120

\[ E_{\text{int}} [\text{kJ mol}^{-1}] = 1124G_b [\text{au}] \]  

(1)

where \(G_b\) is the local electronic kinetic energy density at the O···H−Cl or H2O···Cl bond critical point in electron density.121 Equation 1 yields reasonable \(E_{\text{int}}\) values for Cl···Cl and Hal···O noncovalent interactions, where Hal = F or Cl, in gas and condensed phases.122−124

### ASSOCIATED CONTENT

#### Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.omega.7b01034. Measurement procedure details; ATR-IR spectra of aqueous solution of sodium diclofenac at different concentrations; differences in the microsolvation of the −CO2− group in the DN− and acetate (CH3CO2−) anions; low-energy structures of DN−·3H2O and DN−·4H2O complexes computed using DFT; structures of several H-bonded complexes of the DN− ion and water molecules obtained during MD simulation; atomic partial charges and van der Waals parameters of DN anion in MD simulation; model with atom types of the diclofenac anion; provisional bond assignment in the experimental spectrum of aqueous solution of NaDN; evolution of C−C−N−C dihedral angle of the DN− ion (PDF)
trifluoroacetic acid-water system. Zh. Phys. Khim. 1971, 45, 1488–1492 (in Russian).
(16) Spitzer, U. A.; Toone, T. V.; Stewart, R. Aqueous trifluoroacetic acid as a medium for organic reactions. I. Acidity functions and the identity of the manganese(VII) species found in powerfully acidic media. Can. J. Chem. 1976, 54, 440–447.
(17) Drago, S.; Imboden, R.; Schlatter, P.; Bulaert, M.; Krähenbühl, S.; Drew, J. Pharmacokinetics of Transdermal Etofenteamate and Diclofenac in Healthy Volunteers. Basic Clin. Pharmacol. Toxicol. 2017, 121, 423–429.
(18) Bedada, S. K.; Yelü, N. R.; Neerati, P. Effect of Resveratrol Treatment on the Pharmacokinetics of Diclofenac in Healthy Human Volunteers. Phytother. Res. 2016, 30, 397–401.
(19) Hartlieb, K. J.; Ferris, D. P.; Holcroft, J. M.; Kandel, I.; Stern, C. L.; Nassar, M. S.; Botros, Y. Y.; Stoddart, J. F. Encapsulation of Ibuprofen in CD-MOF and Related Bioavailability Studies. Mol. Pharm. 2017, 14, 1831–1839.
(20) Moutasim, M. Y.; ElMeshad, A. N.; El-Nabarawi, M. A. A pharmaceutical study on lornoxicam fast disintegrating tablets: formulation and in vitro and in vivo evaluation. Drug Delivery Transl. Res. 2017, 7, 450–459.
(21) Colombo, M., Staufenbiel, S.; Rühl, E.; Bodmeier, R. In situ determination of the saturation solubility of nanocrystals of poorly soluble drugs for dermal application. Int. J. Pharm. 2017, 521, 156–166.
(22) Bondar, A.-N.; Dau, H. Extended protein/water H-bond networks in photosynthetic water oxidation. Biochim. Biophys. Acta 2012, 1817, 1177–1190.
(23) Díaz, N.; Suarez, D.; Sordo, T. L.; Merz, K. M. Molecular dynamics study of the IIA binding site in human serum albumin: influence of the protonation state of Lys193 and Lys199. J. Med. Chem. 2001, 44, 250–260.
(24) Díaz, N.; Sordo, T. L.; Merz, K. M., Jr.; Suárez, D. Insights into the acylation mechanism of class A beta-lactamases from molecular dynamics simulations of the TEM-1 enzyme complexed with benzylpenicillin. J. Am. Chem. Soc. 2003, 125, 672–684.
(25) Fini, A.; Laus, M.; Orienti, I.; Zecchi, V. Dissolution and partition thermodynamic functions of some nonsteroidal anti-inflammatory drugs. J. Pharm. Sci. 1986, 75, 23–25.
(26) Iftīmīe, R.; Tuckerman, M. E. Decomposing total IR spectra of aqueous systems into solute and solvent contributions: A computational approach using maximally localized Wannier orbitals. J. Chem. Phys. 2005, 122, No. 214508.
(27) Gaigeot, M. P. Theoretical spectroscopy of floppy peptides at room temperature. A DFTMD perspective: gas and aqueous phase. Phys. Chem. Chem. Phys. 2010, 12, 3336–3339.
(28) Troittsch, R. Z.; Tulip, P. R.; Crain, J.; Martyna, G. J. A simplified model of local structure in aqueous proline acid revealed by first-principles molecular dynamics simulations. Biophys. J. 2008, 95, 5014–5020.
(29) Sun, J.; Bousquet, D.; Forbert, H.; Marx, D. Glycine in aqueous solution: solvation shells, interfacial water, and vibrational spectroscopy from ab initio molecular dynamics. J. Chem. Phys. 2010, 133, 114508–114518.
(30) Sun, J.; Niehues, G.; Forbert, H.; Decka, D.; Schwab, G.; Marx, D.; Havenith, M. Understanding THz spectra of aqueous solutions: glycine in light and heavy water. J. Am. Chem. Soc. 2014, 136, 5031–5038.
(31) Yadav, V. K.; Klein, M. L. Probing the dynamics of N-methylacetamide in methanol via ab initio molecular dynamics. Phys. Chem. Chem. Phys. 2017, 19, 12868–12875.
(32) Sterpone, F.; Strimeman, G.; Hynes, J. T.; Laage, D. Water hydrogen-bond dynamics around amino acids: the key role of hydrophilic hydrogen-bond acceptor groups. J. Phys. Chem. B 2010, 114, 2083–2089.
(33) Rocchi, C.; Bizzarri, A. R.; Cannistraro, S. Water dynamical anomalies evidenced by molecular-dynamics simulations at the solvent-protein interface. Phys. Rev. E 1998, 57, 3315–3325.
(44) Semrouni, D.; Sharma, A.; Dognon, J.-P.; Ohanessian, G.;
(45) Vener, M. V.; Odinokov, A. V.; Wehmeyer, C.; Sebastiani, D.
(46) Tomasi, J.; Mennucci, B.; Cammi, R. Quantum Mechanical
(47) Praprotnik, M.; Janežič, D.; Marić, V. Temperature Dependence of Water Vibrational Spectrum: A Molecular Dynamics Simulation Study. J. Phys. Chem. A 2004, 108, 11056–11062.
(48) Semrouni, D.; Sharma, A.; Doganov, J.-P.; Ohanessian, G.; Clavgaviera, C. Finite Temperature Infrared Spectra from Polarizable Molecular Dynamics Simulations. J. Chem. Theory Comput. 2014, 10, 3190–3199.
(49) Vener, M. V.; Odinokov, A. V.; Wehmeyer, C.; Sebastiani, D. The structure and IR signatures of the arginine-glutamate salt bridge. Insights from the classical MD simulations. J. Chem. Phys. 2015, 142, No. 215106.
(50) Tomasi, J.; Mennucci, B.; Cammi, R. Quantum Mechanical Continuum Solvation Models. Chem. Rev. 2005, 105, 2999–3044.
(51) Tušnö, L.; Rinaldi, D.; Ruiz-López, M. F.; Rival, J. L. Hydroxide ion in liquid water: structure, energetics, and proton transfer using a mixed discrete-continuum ab initio model. J. Phys. Chem. 1995, 99, 3798–3805. (52) Barone, V.; Carmimeo, I.; Scalmanni, G. Computational Spectroscopy of Large Systems in Solution: The DFTB/PCM and TD-DFTB/PCM Approach. J. Chem. Theory Comput. 2013, 9, 2052–2071 and references therein.
(53) Vener, M. V.; Shendedovich, I. G.; Rykoumov, A. A. A qualitative study of the effect of a co-solute and polar environment on the structure and spectroscopic signatures of a hydrated hydroxyl anion. Theor. Chem. Acc. 2013, 132, No. 1361.
(54) Chowdhry, B. Z.; Dines, T. J.; Jábeen, S.; Withnall, R. Vibrational Spectra of α-Amino Acids in the Zwitterionic State in Aqueous Solution and the Solid State: DFT Calculations and the Influence of Hydrogen Bonding. J. Phys. Chem. A 2008, 112, 10333–10347.
(55) Zhu, H.; Blom, M.; Compagnon, I.; Rius, A. M.; Roy, S.; von Helden, G.; Schmidt, B. Conformations and vibrational spectra of a model tripeptide: change of secondary structure upon micro-solvation. Phys. Chem. Chem. Phys. 2010, 12, 3415–3425 and references therein.
(56) de Tudela, R. P.; Marx, D. Water-Induced Zwitterionization of Glycine: Stabilization Mechanism and Spectral Signatures. J. Phys. Chem. Lett. 2016, 7, 5137–5142.
(57) Scholer, D. W.; Ku, E. C.; Boettcher, I.; Schweizer, A. Pharmacology of diclofenac sodium. Am. J. Med. 1986, 80, 34–38. (58) Rowlinson, S. W.; Kiefer, J. R.; Prusakiewicz, J. J.; Pawlitz, J. L.; Kozak, K. R.; Kalogkar, A. S.; Stallings, W. C.; Kurumbail, R. G.; Marnett, L. J. A Novel Mechanism of Cyclooxygenase-2 Inhibition Involving Interactions with Ser-530 and Tyr-385. J. Biol. Chem. 2003, 278, 45763–45769.
(59) Sidhu, R. S.; Lee, J. Y.; Yuan, C.; Smith, W. L. Comparison of Cyclooxygenase-1 Crystal Structures: Cross-Talk between Monomers Comprising Cyclooxygenase-1 Homodimers. Biochemistry 2010, 49, 7069–7079.
(60) Ng, S. C.; Chan, F. K. NSAID-induced gastrointestinal and cardiovascular injury. Curr. Opin. Gastroenterol. 2010, 26, 611–617.
(61) Sakat, S. S.; Mian, K.; Demidenko, Y. O.; Gorbulov, E. A.; Tarasov, S. A.; Mathur, A.; et al. Release-active dilutions of diclofenac enhance anti-inflammatory effect of diclofenac in carrageenan-induced rat paw edema model. Inflammation 2014, 37, 1–9.
(62) Arancibia, J. A.; Boldrini, M. A.; Escandar, G. M. Spectrofluorimetric determination of diclofenac in the presence of α-cyclodextrin. Talanta 2000, 52, 261–268.
(63) Sahra, K.; Dinari, K.; Seridi, A.; Kadri, M. Investigation on the inclusion of diclofenac with β-cyclodextrin: a molecular modeling approach. Struct. Chem. 2015, 26, 61–69.
(64) de Namor, A. D.; Nejad, M. A.; Salas, J. A. V.; Bryant, S.; Howlin, B. A calix[4]arene derivative and its selective interaction with drugs (clofibric acid, diclofenac and aspirin). Eur. J. Pharm. Sci. 2017, 100, 1–8.
(65) Mubengayi, C. K.; Ramli, Y.; Routaboul, C.; Gilard, V.; Karbene, M. E.; Cherrah, Ya; Malet-Martino, M.; Essass El, M. Quality Evaluation of Diclofenac Formulations Manufactured in DR Congo. Pharm. Anal. Chem. 2016, 2, 112.
(66) Kozlowska, M.; Rodziewicz, P.; Kaczmarek-Kedziera, A. Structural stability of diclofenac vs. inhibition activity from ab initio molecular dynamics simulations. Comparative study with ibuprofen and ketoprofen. Struct. Chem. 2017, 28, 999–1008.
(67) Pouplana, R.; Perez, C.; Sanchez, J.; Lozano, J. J.; Puig-Pallaret, P. The structural and electronical factors that contribute affinity for the time-dependent inhibition of PGHS-1 by indomethacin, diclofenac and fenamates. J. Comput.-Aided Mol. Des. 1999, 13, 297–313.
(68) Jorgensen, W. L.; Gao, J. Monte Carlo simulations of the hydration of ammonium and carboxylic acids. J. Phys. Chem. 1986, 90, 2174–2182.
(69) Gojlo, E.; Śmiejchowski, M.; Panuszko, A.; Stangret, J. Hydration of Carboxylic Anions: Infrared Spectroscopy of Aqueous Solutions. J. Phys. Chem. B 2009, 113, 8128–8136 and references therein.
(70) Vovk, M. A.; Pavlova, M. S.; Chizhik, V. I.; Vorontsova, A. A. Hydrate Shell Models of Acetate Ions According to Nuclear Magnetic Relaxation Data and Quantum–Chemical Calculations. Russ. J. Phys. Chem. A 2011, 85, 1597–1602.
(71) Mazurek, S.; Sioztoł, R. Comparison of infrared attenuated total reflection and Raman spectroscopy in the quantitative analysis of diclofenac sodium in tablets. Vib. Spectros. 2011, 57, 157–162.
(72) Sammon, C.; Mura, C.; Yardwood, J.; Everall, N.; Swart, R.; Hodge, D. FTIR–ATR Studies of the Structure and Dynamics of Water Molecules in Polymeric Matrices. A Comparison of PET and PVC. J. Phys. Chem. B 1998, 102, 3402–3411.
(73) Evans, J. C. Further studies of unusual effects in the infrared spectra of certain molecules. Spectrochim. Acta 1960, 16, 994–1000. (74) Evans, J. C. Narrow transmission regions within broad infrared absorption bands in solids. Spectrochim. Acta 1962, 18, 507–512.
(75) Max, J.-J.; Chapados, C. Infrared spectroscopy of aqueous ionic salt mixtures at low concentrations: Ion pairing in water. J. Chem. Phys. 2007, 127, No. 114509.
(76) Reimenschneider, J. Spectroscopic Investigations on Pure Water and Aqueous Salt Solutions in the Mid Infrared Region; University of Rostock: Rostock, 2011; pp 1–112.
(77) del Val, C.; Bondar, A. N. Charged groups at binding interfaces of the PsbO subunit of photosystem II: a combined bioinformatics and simulation study. Biochim. Biophys. Acta, Bioenerg. 2017, 1858, 432–441.
A. D. Inelastic neutron scattering studies of some intramolecular infrared line shapes of H-bonds within the strong anharmonic coupling

Kent, P. R. C.; Kubicki, J. D.; Wesolowski, D. J.; Sofo, J. O. Vibrational Hydrogen Bonding: Functionality and Graph Set Analysis in Crystals. ACS Omega 2007, 3, 2601–2611.

B. J.; Li, Y.; Homan, E. A.; Lampkins, A. J.; Marin, I. V.; Abboud, K. A. Seven-Membered Intramolecular Hydrogen Bonding of Phenols: Database Analysis and Phenolucin Model Compounds. Eur. J. Org. Chem. 2012, 2012, 4483–4492.

Filarowski, A.; Koll, A.; Sobczyk, L. Intramolecular Hydrogen Bonding in o-hydroxy Aryl Schiff Bases. Curr. Org. Chem. 2009, 13, 172–193.

Filarowski, A. Intramolecular hydrogen bonding in o-hydroxyaryl Schiff bases. J. Phys. Org. Chem. 2005, 18, 686–698.

Thomsen, B.; Kawakami, T.; Shigemoto, I.; Sugita, Y.; Yagi, K. Weight-Averaged Anharmonic Vibrational Analysis of Hydration Structures of Polyamide 6. J. Phys. Chem. B 2017, 121, 6050–6063.

Berendsen, H. J. C.; van der Spoel, D.; van Drunen, R. GROMACS: A message-passing parallel molecular dynamics implementation. Comput. Phys. Commun. 1995, 91, 43–56.

Lindahl, E.; Hess, B.; van der Spoel, D. GROMACS 3.0: a package for molecular simulation and trajectory analysis. J. Mol. Model. 2001, 7, 306–317.

van der Spoel, D.; Lindahl, E.; Hess, B.; Groenhof, G.; Mark, A. E.; Berendsen, H. J. C. GROMACS: fast, flexible, and free. J. Comput. Chem. 2005, 26, 1701–1718.

Hess, B.; Kutzner, C.; van der Spoel, D.; Lindahl, E. GROMACS 4: algorithms for highly efficient, load-balanced, and scalable molecular simulation. J. Chem. Theory Comput. 2008, 4, 435–461.

Llinas, A.; Burley, J. C.; Box, K. J.; Glen, R. C.; Goodman, J. M. Diclofenac solubility: independent determination of the intrinsic solubility of three crystal forms. J. Med. Chem. 2007, 50, 979–983.

Cornell, W. D.; Cieplak, P.; Bayly, C. I.; Gould, I. R.; Merz, K. M.; Ferson, D. M.; Spellmeyer, D. C.; Fox, T.; Caldwell, J. W.; Kollman, P. A. A second generation force field for the simulation of proteins, nucleic acids, and organic molecules. J. Am. Chem. Soc. 1995, 117, 5179–5187.

Berendsen, H. J. C.; Grigera, J. R.; Sarama, T. P. The missing term in effective pair potentials. J. Phys. Chem. 1987, 91, 6269–6271.

Bussi, G.; Donadio, D.; Parrinello, M. Canonical sampling through velocity rescaling. J. Chem. Phys. 2007, 126, No. D4101.

Hockney, R. W.; Goel, S. P.; Eastwood, J. Quiet high resolution computer models of a plasma. J. Comput. Phys. 1974, 14, 148–158.

Darden, T.; York, D.; Pedersen, L. Particle mesh Ewald: An N·log(N) method for Ewald sums in large systems. J. Chem. Phys. 1993, 98, 10089–10092.

Esmann, U.; Perera, L.; Berkowitz, M. L.; Darden, T.; Lee, H.; Pedersen, L. G. A smooth particle mesh Ewald method. J. Chem. Phys. 1995, 103, 8577–8593.

Frisch, M. J.; Trucks, G. W.; Schlegel, H. B.; Scuseria, G. E.; Robb, M. A.; Cheeseman, J. R.; Zakrzewski, V. G.; Montgomery, J. A.; Stratmann, R. E.; Burant, J. C.; Dapprich, S.; Millam, J. M.; Daniels, A. D.; Kudin, K. N.; Strain, M. C.; Farkas, O.; Tomasi, J.; Barone, V.; Cossi, M.; Cammi, R.; Mennucci, B.; Pomelli, C.; Adamo, C.; Clifford, S.; Ochterski, J.; Petersson, G. A.; Ayala, P. Y.; Cui, Q.; Morokuma, K.; Malick, D. K.; Rabuck, A. D.; Raghavachari, K.; Foresman, J. B.; Cioslowski, J.; Ortiz, J. V.; Stefanov, B. B.; Liu, G.; Liashenko, A.; Piskorz, P.; Komaromi, I.; Gomperts, R.; Martin, R. L.; Fox, D. J.
Keith, T.; Al-Laham, M. A.; Peng, C. Y.; Nanayakkara, A.; Gonzalez, C.; Challacombe, M.; Gill, P. M. W.; Johnson, B. G.; Chen, W.; Wong, M. W.; Andres, J. L.; Head-Gordon, M.; Replogle, E. S.; Pople, J. A. Gaussian Inc.: Pittsburgh, PA, 2003.

(115) Merrick, J. P.; Moran, D.; Radom, L. An Evaluation of Harmonic Vibrational Frequency Scale Factors. J. Phys. Chem. A 2007, 111, 11683−11700.

(116) Grimme, S. Semiempirical GGA-type density functional constructed with a long-range dispersion correction. J. Comput. Chem. 2006, 27, 1787−1799.

(117) Katsyuba, S. A.; Vener, M. V.; Zvereva, E. E.; Brandenburg, J. G. The role of London dispersion interactions in strong and moderate intermolecular hydrogen bonds in the crystal and in the gas phase. Chem. Phys. Lett. 2017, 672, 124−127.

(118) Vener, M. V.; Chernyshov, I. Y.; Rykounov, A. A.; Filarowski, A. Structural and Spectroscopic Features of Proton Hydrates in the Crystalline State. Solid-state DFT Study on HCl and Triflic Acid Hydrates. Mol. Phys. 2018, 116, 251−262.

(119) AIMAll, version 17.01.25; TK Gristmill Software: Overland Park, KS, 2017.

(120) Mata, I.; Alkorta, I.; Espinosa, E.; Molins, E. Relationships between interaction energy, intermolecular distance and electron density properties in hydrogen bonded complexes under external electric fields. Chem. Phys. Lett. 2011, 507, 185−189.

(121) Bader, R. F. W. Atoms in Molecules - A Quantum Theory; Oxford University Press: Oxford, 1990.

(122) Nelyubina, Y. V.; Antipin, M. Y.; Lyssenko, K. A. Are Halide···Halide Contacts a Feature of Rock-Salts Only? J. Phys. Chem. A 2007, 111, 1091−1095.

(123) Katsyuba, S. A.; Vener, M. V.; Zvereva, E. E.; Fei, Z.; Scopelliti, R.; Laurenczy, G.; Yan, N.; Paunescu, E.; Dyson, P. J. How Strong Is Hydrogen Bonding in Ionic Liquids? Combined X-ray Crystallographic, Infrared/Raman Spectroscopic, and Density Functional Theory Study. J. Phys. Chem. B 2013, 117, 9094−9105.

(124) Vener, M. V.; Shishkina, A. V.; Rykounov, A. A.; Tsirelson, V. G. Cl···Cl Interactions in Molecular Crystals: Insights from the Theoretical Charge Density Analysis. J. Phys. Chem. A 2013, 117, 8459−8467.