

# Exploring Ion-Ion Interactions in Aqueous Solutions by a Combination of Molecular Dynamics and Neutron Scattering

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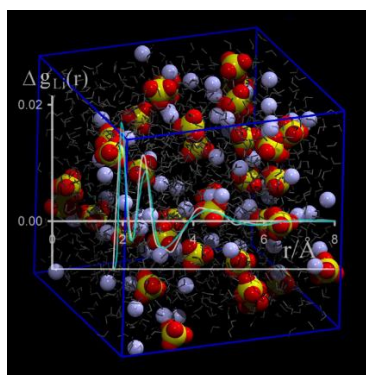
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## ABSTRACT

Recent advances in computational and experimental techniques have allowed for accurate description of ion pairing in aqueous solutions. Free energy methods based on ab initio molecular dynamics, as well as on force fields accounting effectively for electronic polarization, can provide quantitative information about the structures and occurrences of individual types of ion pairs. When properly benchmarked against electronic structure calculations for model systems and against structural experiments, in particular neutron scattering, such force field simulations represent a powerful tool for elucidating interactions of salt ions in complex biological aqueous environments.

*TOC graphic*



KEYWORDS: ion pairing, molecular dynamics, neutron scattering, density functional theory, charge rescaling.

Many inorganic salts readily dissolve in water, implying that the cation-anion attraction in the crystal is replaced by favorable ion-water interactions. Nevertheless, ion pairing does not vanish completely in water except for the ideal case of solutions at infinite dilution. At realistic situations of finite concentrations the question is not whether ion pairing is present but rather how strong it is. For each salt the particular outcome is a result of a fine balance between direct Coulomb attraction between the ions and the solvation forces. At a given concentration, one way (which is not covered in this Perspective) to tip the scale toward strong ion pairing is to reduce the dielectric constant either by going to supercritical water<sup>1</sup> or switching to other polar solvents.<sup>2</sup> Another possibility, discussed in more detail below, is to move from monovalent to multivalent salt ions. This often tends to strengthen ion pairing in water which is, however, already non-negligible for monovalent ions starting from moderate concentrations, such as those present at physiological conditions.<sup>2-4</sup> This is also true for interactions of biologically relevant salt ions with charged side chain groups of aqueous proteins, which are of key importance for salting out and salting in of proteins as embodied in the Hofmeister series of ions.<sup>5-10</sup>

Within the Pitzer's model of electrolytes,<sup>11</sup> which is widely used to predict activity coefficients of salt solutions, ion-ion interactions are recognized only implicitly without direct reference to ion pairing. This is despite the fact that already in 1920s Bjerrum<sup>12</sup> postulated the existence of associated ions of opposite sign in the solution. While at that time there was no direct evidence available, today we have ample experimental and computational data pointing to the existence of

contact, as well as solvent-shared and solvent-separated ion pairs in aqueous solutions,<sup>2, 13</sup> as originally suggested by Eigen and Tamm,<sup>14</sup> despite the fact that ion pairing in water can be weak in many cases.

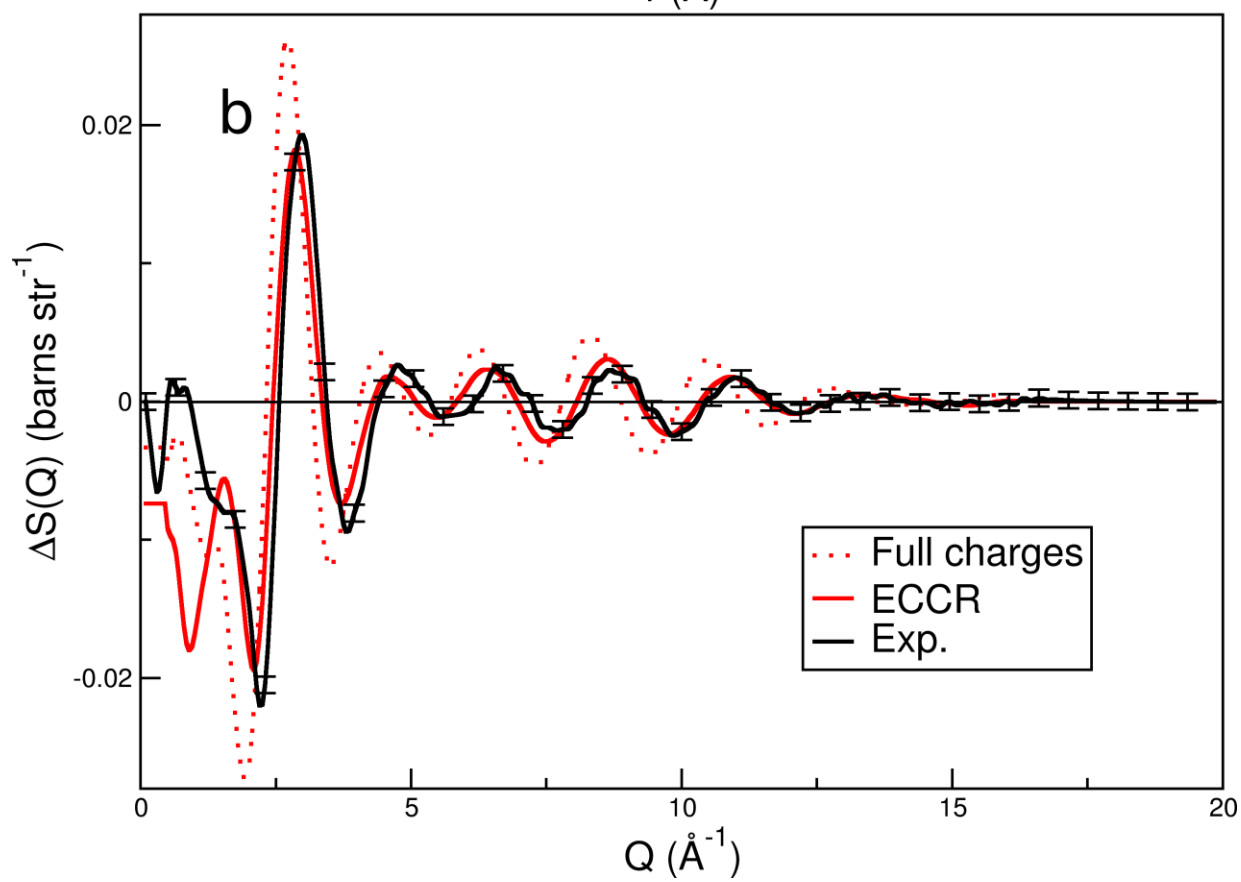
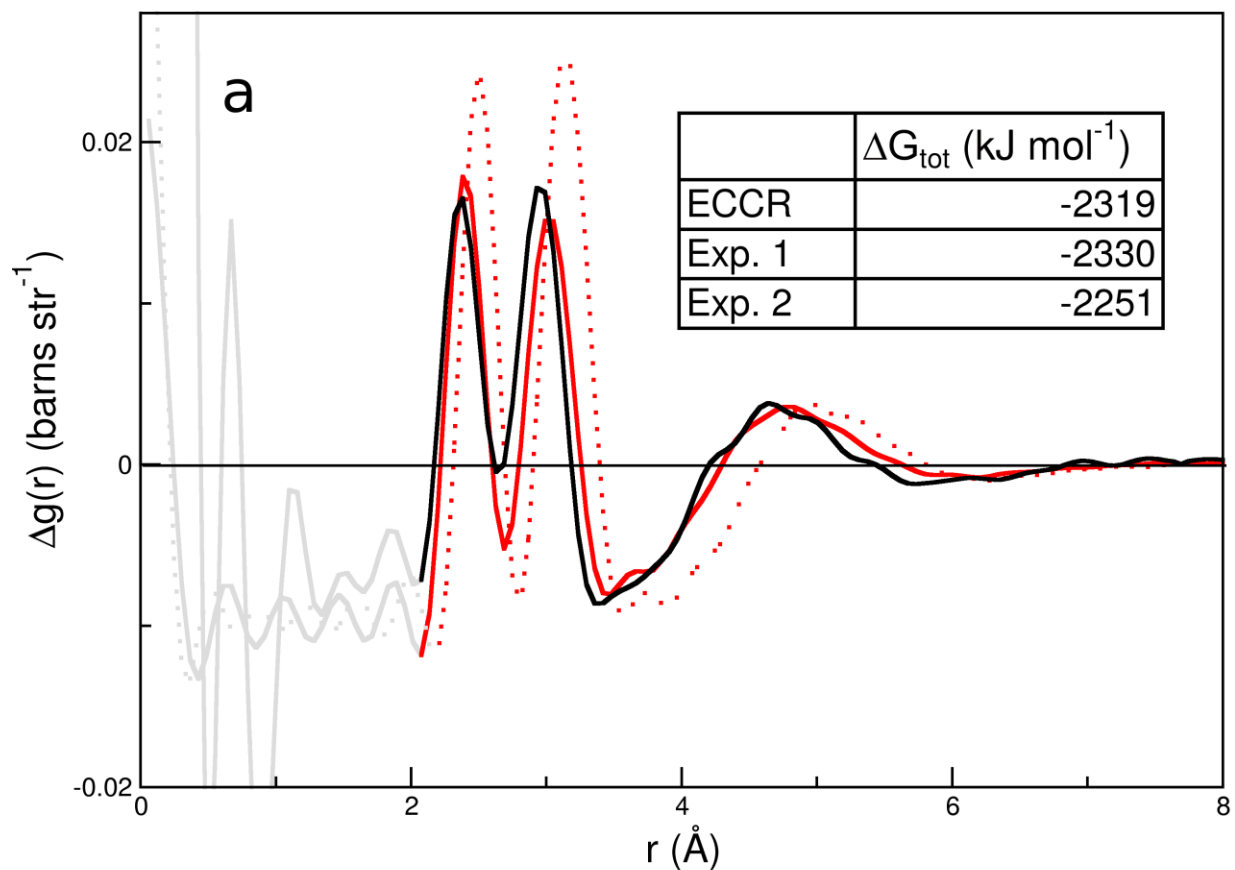
In this Perspective, we summarize recent progress in quantifying ion pairing in aqueous solutions with special focus on our advances in three areas: i) construction of ionic force fields implicitly including electronic polarization,<sup>15-17</sup> ii) development of techniques for calculating free energy profiles for ion-ion interactions in water by ab initio molecular dynamics,<sup>18-21</sup> and iii) establishment of experimental benchmarks for molecular structures of salt solutions using neutron diffraction with isotopic substitution.<sup>22-25</sup>

### **Force Field Molecular Dynamics Accounting for Polarization Effects**

Direct classical molecular dynamics simulations have been used extensively for modeling ion pairing in water and aqueous solutions (see, e.g., Refs. 26-29 and references therein). Several recent studies employing free energy techniques investigated systematically pairing of common monovalent salt ions (typically alkali cations and halide anions) using non-polarizable force fields, yielding for each pair the potential of mean force, i.e., the free energy profile as a function of the ion-ion distance.<sup>13, 30-31</sup> The results, in particular the positions and depths of the contact and solvent-shared (or solvent-separated) free energy minima turn out to depend sensitively on the force field used. This reflects, among others, the neglect of electronic polarization effects which are only rarely included in ion pairing studies<sup>32</sup> despite their importance, particularly for pairing of high charge density ions.<sup>16</sup>

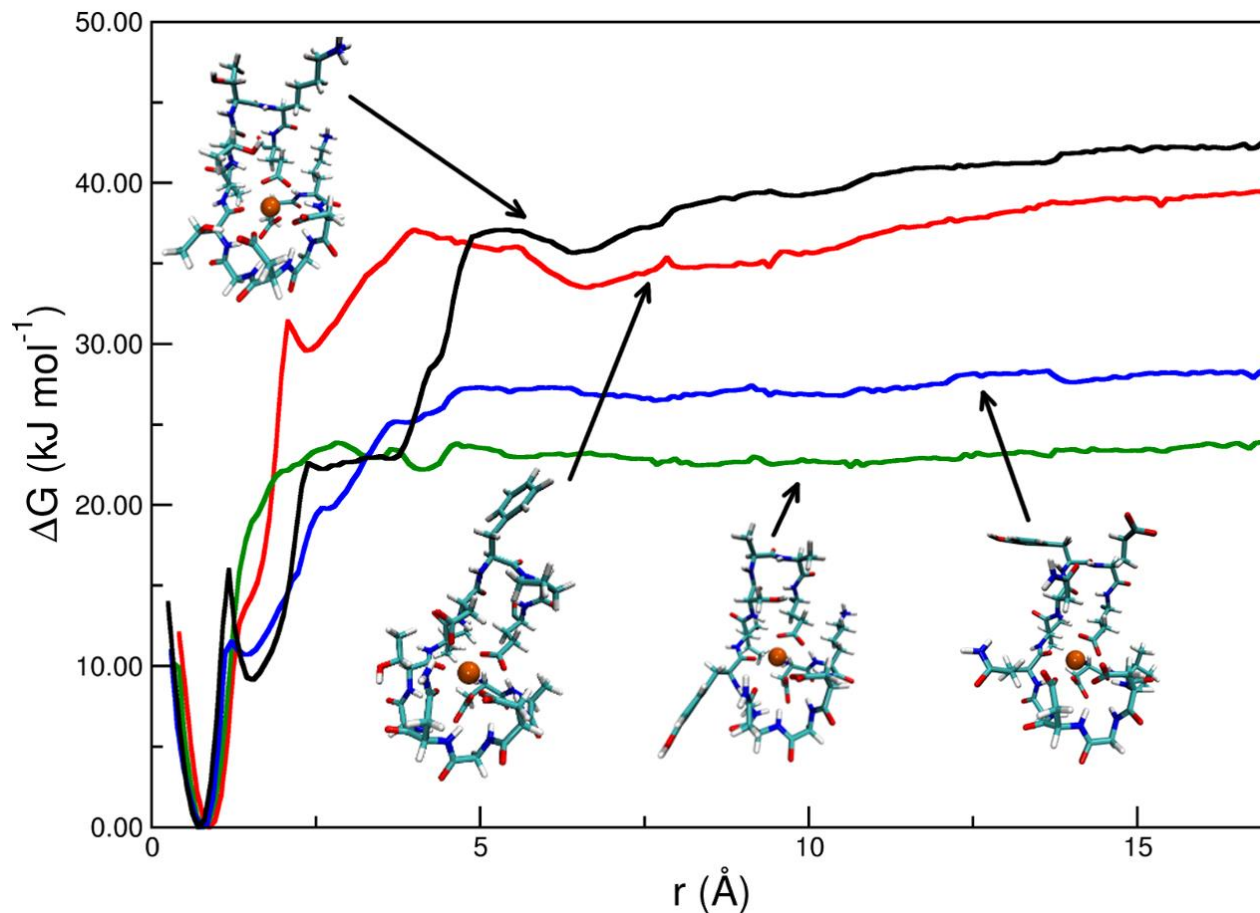
A simple and efficient, but at the same time rigorous way for including electronic polarization effects in a mean-field way is via rescaling ionic charges by the inverse square root of the

electronic part of the dielectric constant of the solvent.<sup>15</sup> This follows directly from the Coulomb expression for electrostatic interactions in an electronic dielectric continuum and for ion pairing in water it simply means rescaling the ionic charges by a factor of about 0.75 within a non-polarizable force field setup.<sup>33</sup> Applying this approach to aqueous lithium salts provides a grossly improved structural description of the solution; in particular it removes the artificial excessive ion pairing and clustering observed upon using standard non-polarizable force fields.<sup>25</sup> The improvement becomes particularly significant for aqueous solutions of divalent ions, such as calcium or sulfate, which are notoriously difficult to describe within non-polarizable models.<sup>16-17</sup> Comparison to ab initio molecular dynamics and neutron scattering shows that with a slight readjustment of the ionic radii the structures of the solutions, in particular the amount and geometry of ion pairing, can be described quantitatively using the charge scaling approach (denoted as electronic continuum correction with rescaling – ECCR).<sup>16-17</sup> For a concentrated  $\text{CaCl}_2$  aqueous solution this is demonstrated in Figure 1, which shows the dramatic improvement of the total structure factor with respect to neutron scattering experiment (*vide infra*) in both real (Figure 1a) and reciprocal (Figure 1b) space upon applying this approach. Moreover, charge scaling brings dynamical properties like viscosity significantly closer to experiment.<sup>17</sup> Finally, the hydration free energy of  $\text{CaCl}_2$ , i.e. a single calcium ion and two chlorides, is also put in accord with experiment<sup>34-35</sup> in this way (see inset of Figure 1a).



**Figure 1:** *First-order difference function in a)  $r$ -space and b)  $Q$ -space for the ECCR (full red line) vs. full charges (dotted red line) simulations and the experimentally measured curve (black) for a 4 m aqueous solution of calcium chloride.<sup>17</sup> The table in the inset in Figure 1a shows the presently calculated vs. experimental<sup>34-35</sup> hydration free energies of  $\text{CaCl}_2$ .*

Encouraged by these results, we have recently employed the same charge scaling model to investigate calcium pairing in a biological context, namely its interactions with a calcium-binding protein calmodulin in water. The most common calcium-binding motif in proteins is the so called EF-hand loop containing negatively charged side chain carboxylic groups of aspartates and glutamates, as well as backbone carbonyls.<sup>36</sup> Aqueous calmodulin possesses four such loops and we have quantified using umbrella sampling simulations calcium binding to each of them, as demonstrated on the four free energy curves presented in Figure 2.<sup>37</sup> The principal result is that the experimental binding free energies and the corresponding structures of the four calcium-containing loops, which differ quantitatively from each other, are much better reproduced within the charge scaling approach than upon employing standard non-polarizable force fields (for a detailed comparison between calculations based on scaled vs. unscaled charges see Ref. 37). An open question remains, how to perform best the additional rescaling of the charged amino acid side chain groups<sup>15</sup> without upsetting the overall balance of a given protein force field (note that the partial charges on hydrogens and oxygens of water molecules have been effectively scaled already during the development of the water force field based on experimental data, therefore, they need not be further rescaled).<sup>33, 38</sup>



**Figure 2:** Calcium binding free energy profiles (loop I – black, loop II – red, loop III – green, and loop IV – blue) calculated using the ECCR approach and the corresponding structures of the four EF-hand loops of aqueous calmodulin with bound  $\text{Ca}^{2+}$  (orange balls).<sup>37</sup>

### Ab Initio Molecular Dynamics

Arguably the most rigorous approaches to ion pairing in water should be based on electronic structure methods rather than on empirical force fields. The necessary minimal size of the system encompassing tens to hundreds of water molecules effectively narrows the choice to computationally “cheap” density functional theory (DFT) methods. Even for them, the propagation times necessary to obtain statistically significant methods have been prohibitive

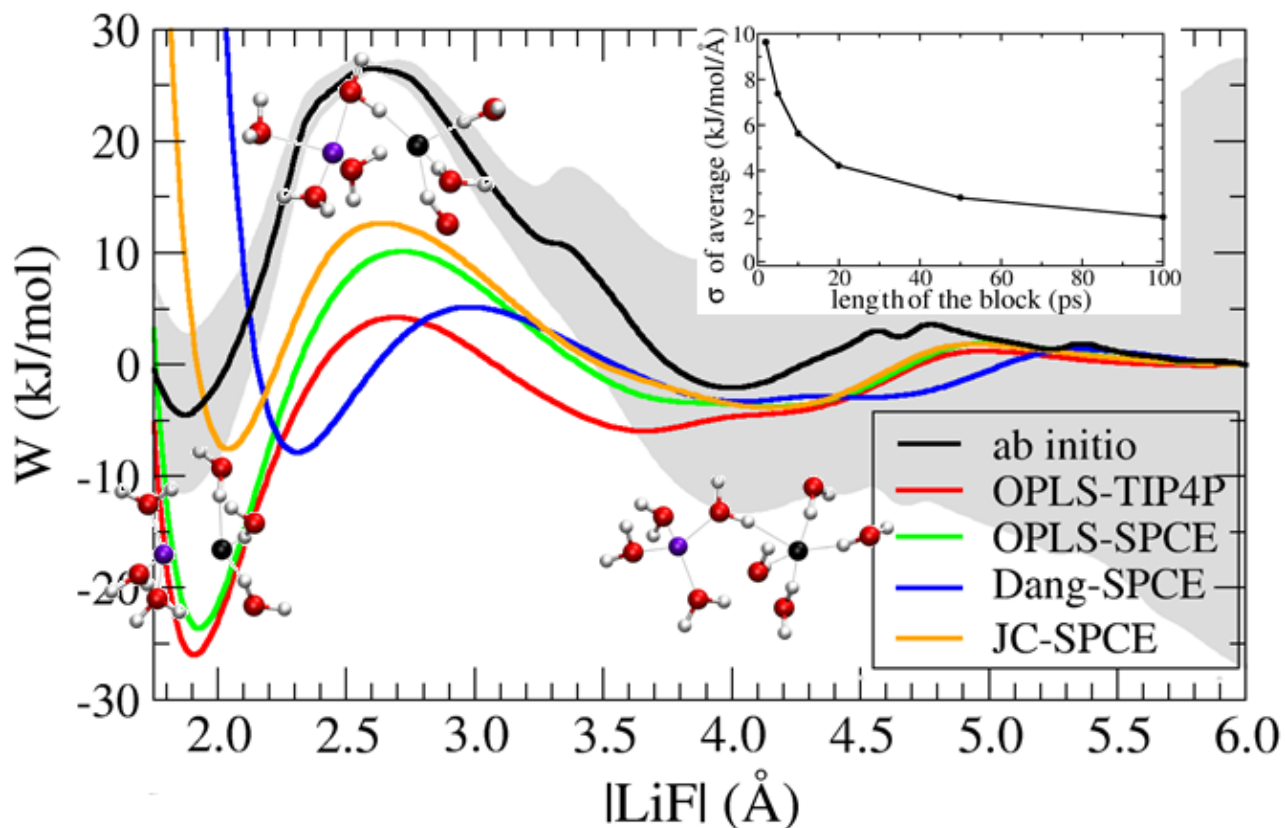
until recently. For this reason, first attempts to map ion-ion interactions by ab initio molecular dynamics<sup>18-19</sup> based on short (tens of picoseconds) propagation times can be viewed as semi-quantitative at best.

In our attempt to get the statistical uncertainties under control we relied on the assumption that the statistical convergence of the ion-ion potentials of mean force is comparable for DFT and force field based method.<sup>21</sup> This allowed us to establish the necessary length of a single sampling window to reach a given statistical accuracy in a potential of mean force or umbrella sampling simulation (see inset of Figure 3). Figure 3 shows the potential of mean force for the lithium-fluoride ion pair in water established using the BLYP functional with the empirical dispersion correction.<sup>39</sup> with 50 ps per window (total ~2 ns of simulation time), compared to four common non-polarizable force fields.<sup>21</sup>  $\text{Li}^+$  and  $\text{F}^-$  represent high charge density ions with non-negligible charge transfer to the surrounding water molecules. As a result, they are difficult to describe using empirical force fields and, consequently, the resulting potentials of mean force deviate significantly from the DFT result which shows an almost isoergic contact and solvent separated ion pairs separated by a rather high barrier (Figure 3).<sup>21</sup> Note that the gray shading in Figure 3 represents an upper bound to the statistical error of the DFT potential of mean force.<sup>21</sup> This strongly indicates that the deviations of the free energy curves from standard non-polarizable force fields are well beyond the statistical error of the DFT results.

If DFT should serve as a “policeman” of the empirical force fields, then who checks the policeman? One way we benchmarked our DFT approach was by checking it against more accurate electronic structure methods (such as the second order Møller-Plesset perturbation theory or Coupled Clusters) for small ion-water clusters, while the other option was to compare directly to experiment (X-ray absorption in the present case).<sup>21</sup> Encouraged by a satisfactory



performance in both respects, we have most recently employed ab initio molecular dynamics to quantify interaction of sodium and calcium ions with the aqueous protein backbone modeled by a N-methylacetamide molecule.<sup>40</sup> Our calculations not only correctly reproduced the relative strengths of interactions of  $\text{Ca}^{2+}$  vs.  $\text{Na}^+$  with the peptide bond, but also rationalized the corresponding fingerprints in the amide I infrared band in terms of calcium replacing on average two water molecules (while sodium only one) upon pairing with the carbonyl group of the amide.<sup>40</sup>



**Figure 3:** Potential of mean force along the ion-ion coordinate for aqueous lithium fluoride calculated employing ab initio molecular dynamics (black), with an upper bound of the statistical uncertainty obtained as accumulated error during the integration procedure (grey area), as well as using four empirical force fields (colored lines).<sup>21</sup> The investigated system

*contained a single LiF ion pair and 64 water molecules in a 12.6 Å cubic box. The inset shows the standard deviation of the mean force acting along the ion-ion distance for a lithium-fluoride separation corresponding to the contact ion pair, calculated employing the OPLS-TIP4P empirical force field as a function of the duration of the simulation. Snapshots show ions and water molecules in their first solvation shell for contact ion pair, transition state and solvent shared ion pair.*

### **Neutron Scattering with Isotopic Substitution**

Scattering methods, such as neutron or X-ray diffraction, represent direct experimental tools for examining the structure of aqueous salt solutions. In this respect they may have a distinct edge over spectroscopic methods, which report directly on energetics rather than structure. Each of the two scattering techniques has its advantages and disadvantages. The principal advantage of neutron scattering, on which we focus here, is that neutrons scatter off the nuclei via the strong force, while X-rays scatter off the electrons via the electromagnetic force. In practice this means that the scattering centers for the purposes of neutrons scattering are effectively point sources, while in the case of X-ray scattering, they are more “fuzzy blobs”.<sup>41</sup> This makes the subsequent analysis of neutron scattering data more direct than that of the X-ray data. Another important advantage of neutron scattering is that, unlike in X-rays, both H and D are as visible as atoms with heavier nuclei, which is of a key importance in aqueous solutions.<sup>41</sup> At the same time, neutron scattering has the obvious disadvantage that a monochromatic, collimated neutron beam is considerably more difficult and expensive to construct than a comparable X-ray source.

The experimental scattering pattern, i.e., the total structure factor in the reciprocal space  $S(Q)$ , can be expressed in terms of pairwise distribution functions and neutron scattering prefactors.<sup>42</sup> For example, a mononuclear system consisting of atoms A contains only the pairwise distribution component  $S_{AA}(Q)$ . The function  $S_{AA}(Q)$  is related to the radial distribution function  $g_{AA}(r)$  via a Fourier transform. For water, in contrast, the total scattering pattern is constituted of three pairwise distribution functions ( $S_{HH}(Q)$ ,  $S_{HO}(Q)$  and  $S_{OO}(Q)$ ) and for salt solutions additional functions appear. E.g., for a simple sodium chloride solution containing four nuclei (O, H, Na, and Cl) there are in total ten components. The neutron scattering prefactors of these components are essentially products of the atomic concentration and the product of the neutron scattering lengths of the two nuclei. As all the neutron scattering lengths are roughly comparable in size (most are between 0-10 fm), in practice this means that the weighting prefactors generally follow the atomic concentrations of the nuclei in the solution. The small size of the crucial ion-ion term is thus mostly due to the low mole fraction of ions even in rather concentrated solutions (e.g., in a 3 m solution the relative weight of the ion-ion term is only about one percent). Clearly, neutron scattering signals are dominated by the most preponderant nuclei in aqueous solutions, i.e., hydrogen and oxygen. This is a problem when ions are the main subject of interest, since it can be almost impossible to deconvolute directly the minor scattering components due to ion-ion and ion-water interactions from the measurement.

This problem led to the development of a technique to render large portions of the experimental measurement invisible, and hence allow for a more direct examination of the ionic structure and ion-ion interactions in the solution. The basic trick is that two isotopes of a given ion nucleus yield practically identical solution structures, e.g., the radial distribution function  $g_{OO}(r)$  is the same for solutions of  $\text{Na}^{35}\text{Cl}$  and  $\text{Na}^{37}\text{Cl}$  of the same concentration. At the same time, different isotopes of

the same nucleus can have significantly different neutron scattering lengths. Therefore, if the neutron scattering patterns of the two chemically identical solutions are subtracted from each other, all of the components that have identical prefactor weightings and structure factors cancel out and only correlations from the isotopically substituted nucleus to the other nuclei in the system are non-zero. This constitutes the method of first order difference of neutron diffraction with isotopic substitution (NDIS).<sup>43</sup> For the above example of the NaCl solutions this approach reduces the original ten correlations to just four, all of which are related to the hydration of the chloride ion (i.e., Cl-H, Cl-O, Cl-Cl, and Cl-Na).

NDIS can be applied to any system where there is a high enough concentration of nuclei which can be substituted for an isotope with a sufficiently different scattering length. In combination with molecular dynamics simulations it has been used successfully to elucidate the hydration structure and ion pairing in aqueous solutions of salts of simple atomic ions (lithium, potassium, calcium, chloride, etc.), as well as more complex molecular ions like guanidinium, carbonate, or sulfate.<sup>22,</sup>  
<sup>44</sup> We close the circle of the narrative of this Perspective by stating that simulations play a twofold role here. First, force fields are carefully selected and refined such as to reproduce the overall experimental scattering patterns (see Figure 1). Second, these simulations are then used to interpret the experiment in terms of ion hydration and ion pairing structures with atomistic detail.

### **Summary and Future Outlook**

The aim of this Perspective has been to demonstrate that a detailed molecular description of ion-ion interactions in aqueous solutions can be obtained with the use of modern computational and experimental techniques. In order to be quantitative in terms of geometries and occurrences of different types of ion pairs in solution, key molecular interactions including polarization need

to be accounted for and the computational approaches must be benchmarked against structural experiments such as neutron scattering, as well as highly accurate electronic structure calculations for small model systems. At present, the situation in this field of research looks very promising in a twofold respect. On one hand, ab initio molecular dynamics techniques based on density functional theory are becoming applicable for accurate description of ion pairing in simple aqueous salt solutions. On the other hand, force field based simulations accounting implicitly for electronic polarization via rescaling of ionic charges represent a viable option for a faithful description of ionic interactions of complex aqueous systems involving salts, small molecular solutes, as well as large biomolecules.

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**Quotes:**

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