Structure and interactions in simple solutions

D. T. Bowron

ISIS Facility, CCLRC Rutherford Appleton Laboratory, Chilton, Didcot OX11 0QX, UK (*d.t.bowron@rl.ac.uk*)

Neutron scattering with hydrogen/deuterium isotopic substitution techniques has been used to investigate the full range of structural interactions in a dilute 0.02 mol fraction solution of tertiary butanol in water, both in the absence and in the presence of a small amount of sodium chloride. Emphasis is given to the detailed pictures of the intermolecular interactions that have been derived using the empirical potential structure refinement technique. Analysis has been performed to the level of the spatial density distribution functions that illustrate the orientational dependence of the intermolecular interactions between all combinations of molecular and ionic components. The results show the key structural motifs involved in the interactions between the various components in a complex aqueous system. They underline the structural versatility of the water molecule in accommodating a range of different kinds of interactions while retaining its characteristic first-neighbour interaction geometry. Within this framework, the results highlight the complex interplay between the polar, non-polar and charged molecular interactions that exist in the system.

Keywords: water and aqueous solutions; hydration structure; neutron diffraction

1. INTRODUCTION

At the most fundamental level, the interactions that occur between atoms, molecules and ionic species underpin the entirety of the chemical and biochemical world, and throughout these fields the liquid state plays a major role as a very common environment in which the interactions occur. Owing to the intrinsic structural disorder of this state, it has proved a considerable experimental challenge to probe the character and mechanisms of the atomic and molecular interactions that are found in even simple solutions. This information is, however, becoming increasingly important as we strive to improve our understanding and control of many critical chemical and biochemical reactions (Plumridge & Waigh 2002). The timely advance of both experimental and computational methodology has now brought us to the point where it is a realistic possibility to specifically probe the geometries of selected in-solution atomic and molecular interactions. Chemically specific techniques such as neutron diffraction with isotopic substitution now allow us to focus our viewpoint on key atomic and molecular sites and ‘see’ the local environment in which they sit. Modern computer technology then assists us to interpret and visualize the statistical descriptors required for the representation of structurally disordered media.

The natural test bed for the development of these methods has been, and remains, the aqueous solution. Many questions still revolve around how the small and versatile water molecule interacts with a wide range of ionic, non-polar and amphiphilic solute species. Recently, a great deal of progress has been made in these areas, and

we will aim to give an overview of results relating to the structural aspects of the hydration of polar and non-polar moieties, the interaction between non-polar groups in the aqueous environment, and the interaction of simple ions with water and with amphiphiles. To illustrate the majority of these interactions we will present results from two model systems. First, we will investigate a dilute (0.02 mol fraction) solution of tertiary butanol in water, and second, this same binary mixture but with added sodium chloride (at a concentration of 0.01 mol fraction). The binary solution itself presents us with the opportunity to study a diverse range of intermolecular interactions, namely: (i) the interaction of water with the non-polar methyl groups on the alcohol molecule and with the polar alcohol hydroxyl group; (ii) the interaction of water molecules with other water molecules in the presence of these other groups; and (iii) the interactions that occur between alcohol molecules themselves when in aqueous solution. The salt perturbed system will then allow us to investigate the effect that ionic co-solutes can have on the structure of the binary mixture, and also allow us to study the typical interactions that simple anions and cations have with both water and alcohol molecules.

2. METHODS

A vital step along the path to improving our structural understanding of simple molecular liquids has been the development of methods to probe beyond the pair-distribution function, $g(r)$. This measure is the traditional structure descriptor of disordered atomic networks and is experimentally accessible via radiation scattering techniques. However, for molecular systems, a more useful function to study is the *orientational*-correlation function, $g(r, \omega_1, \omega_2)$ (Finney & Bowron 2002). This function not only contains information about molecular separations (r), but also describes relative molecular orientations of molecule 1 (ω_1) and

One contribution of 16 to a Discussion Meeting Issue ‘The molecular basis of life: is life possible without water?’.

molecule 2 (ω_2). This additional information is crucial if progress is to be made in understanding the subtle details of the intermolecular interactions that drive chemical and biochemical processes. Although the orientational-correlation function is computationally accessible from theoretically generated molecular ensembles (Gray & Gubbins 1984), there are as yet no experimental techniques, to our knowledge, that allow us to measure it directly. This lack of experimental data has thus hampered our understanding of 'real' molecular systems, and advances have been limited by the rate of the concomitant development of advanced structural modelling procedures. A particularly successful approach to the orientational-correlation problem, largely developed over the past decade, has been the integration of the neutron scattering with isotopic substitution method with what are generically termed reverse modelling methods. I will present a particular variant based on neutron scattering with hydrogen/deuterium isotopic substitution and the EPSR procedure (Soper 1996, 1998, 2001).

(a) Neutron scattering and isotopic substitution

The result of a single neutron scattering experiment performed on a multi-component structurally disordered atomic system, is a function called the total structure factor, $F(Q)$. This can be written as

$$F(Q) = \sum_{\alpha\beta} c_\alpha c_\beta b_\alpha b_\beta (S_{\alpha\beta}(Q) - 1), \quad (2.1)$$

where c_α and c_β are the concentration of the atomic components of species α and β , respectively, and b_α and b_β are their associated neutron scattering lengths. $S_{\alpha\beta}(Q)$ are the partial structure factors that correspond to structural correlations between pairs of atoms of species α and β , and the sum is made over all possible pairs of atom types in the mixture. For example, if the system consists of three types of atom, A, B and C, then there are six partial structure factors, symmetric between atom pairs, that will contribute to the total, namely $S_{AA}(Q)$, $S_{AB}(Q)$, $S_{AC}(Q)$, $S_{BB}(Q)$, $S_{BC}(Q)$ and $S_{CC}(Q)$. Q is the magnitude of the momentum transfer vector of the scattered neutrons and is defined as $(4\pi\sin\theta)/\lambda$, where 2θ is the scattering angle and λ is the wavelength of the incident neutron.

The total structure factor is related to the total pair-distribution function by a Fourier transform relationship, and the same also holds true for the partial structure factors and what are termed the partial pair-distribution functions $g_{\alpha\beta}(r)$:

$$S_{\alpha\beta}(Q) - 1 = 4\pi\rho \int r^2 [g_{\alpha\beta}(r) - 1] \frac{\sin(Qr)}{Qr} dr, \quad (2.2)$$

where ρ is the average atomic number density of the sample. These partial pair-distribution functions tell us about the radial distribution (number and distance) of atoms of type β around α and vice versa. Just as the total structure factor is a weighted sum of the partial structure factors, so the total pair-distribution function (accessible from a single diffraction measurement) is a weighted sum of the partial pair-distribution functions.

The composite nature of the total pair-distribution function is one of the key challenges that has to be faced when trying to interpret the structural information it contains. In multi-component systems it is often not a trivial matter to separate the relative contributions of the various atom pairs. Each distinct pair combination often makes a contribution to the structure functions on similar length scales to another pair, and as the aim of the experiment is to determine this structure, knowledge of the relative concentration and scattering power of each atom type

proves an insufficient constraint to unfold the form of each partial pair-distribution function.

The challenge of trying to interpret the total pair-distribution function motivated the development of isotopic substitution techniques in neutron scattering (Enderby & Neilson 1979). These methods make use of a particularly beneficial property of the interaction of a neutron with an atomic nucleus, namely that the scattering properties of an element can vary considerably from isotope to isotope, as characterized by its coherent scattering length. A good example is the scattering length of hydrogen and deuterium. These are -3.74 and 6.67 fm, respectively. By combining neutron scattering measurements from isotopically distinct *but otherwise identical* samples, it is possible to extract structural information centred on the isotopically substituted atomic sites (Finney & Soper 1994). This can be illustrated as follows. First, note that for an H/D substitution experiment we can write equation (2.1) in the form

$$F(Q) = c_X^2 b_X^2 [S_{XX}(Q) - 1] + 2c_X c_H b_X b_H [S_{XH}(Q) - 1] + c_H^2 b_H^2 [S_{HH}(Q) - 1], \quad (2.3)$$

where c_H is the concentration of hydrogen sites that we intend to substitute in the system, b_H is the scattering length associated with those hydrogen sites and b_X and c_X are defined as the weighted average of the scattering lengths and concentration of the atomic sites in the system that will not be substituted, i.e.

$$b_X = \sum_{\alpha \neq H} \frac{c_\alpha b_\alpha}{c_X} \quad \text{and} \quad c_X = \sum_{\alpha \neq H} c_\alpha. \quad (2.4)$$

$S_{XX}(Q)$ and $S_{XH}(Q)$ are the *composite* partial structure factors corresponding to correlations between non-substituted atomic sites alone, and the correlations between non-substituted atomic sites and the hydrogen sites that will be substituted. $S_{HH}(Q)$ is a pure partial structure factor involving only correlations between the substituted hydrogen sites. It is then a trivial matter to illustrate how isotopic substitution techniques work. If we perform three experiments on a system where, for example, we label the hydrogen sites of a constituent methyl group (M), with hydrogen, with deuterium and then with a known mixture of hydrogen and deuterium we can write the three accessible total structure factors, one for each experiment, as

$$F_H(Q) = c_X^2 b_X^2 [S_{XX}(Q) - 1] + 2c_X b_X c_M b_H [S_{XM}(Q) - 1] + c_M^2 b_H^2 [S_{MM}(Q) - 1],$$

$$F_D(Q) = c_X^2 b_X^2 [S_{XX}(Q) - 1] + 2c_X b_X c_M b_D [S_{XM}(Q) - 1] + c_M^2 b_D^2 [S_{MM}(Q) - 1],$$

$$F_{HD}(Q) = c_X^2 b_X^2 [S_{XX}(Q) - 1] + 2c_X b_X c_M b_{HD} [S_{XM}(Q) - 1] + c_M^2 b_{HD}^2 [S_{MM}(Q) - 1]. \quad (2.5)$$

This is then nothing other than a simple system of simultaneous equations. We have three unknown quantities, the structure factors $S_{XX}(Q)$, $S_{XM}(Q)$ and $S_{MM}(Q)$, and we have the required three equations to solve them that differ by the known values of the scattering length imposed on the methyl hydrogen site by our selected substitutions.

These equations are the basis of both the 1st-order and 2nd-order isotopic difference techniques that in favourable cases allow us to label specific atomic sites (and molecular functional groups) to then obtain a very detailed insight into the intermolecular interactions in which they are involved (Finney & Soper 1994). In the 1st-order difference experiment, only two

experiments are performed and one can extract the sum of $S_{\text{XH}}(Q)$ and $S_{\text{HH}}(Q)$. In the 2nd-order difference technique, all three experiments are performed and the full separation of the two composite and one pure partial structure factor is performed. Each of these partial structure factors is considerably simpler to interpret than a total structure factor and carries us one step closer to the goal of the structural description of a multi-component molecular system.

In the case of a binary molecular system where it is possible to perform independent isotopic substitutions on molecular groups of each of the two molecular constituents, it is possible to use this method to extract, via seven isotopic substitution experiments, three specific partial pair-distribution functions that characterize the key intermolecular interactions—solute–solute, solvent–solvent and solute–solvent (Finney & Soper 1994). In addition to the three pure partials, this combination of experiments also provides us with six other composite partial structure factors that each give different viewpoints on the structure that exists in the liquid.

(b) Empirical potential structure refinement

Despite the structural insight into the pair-distribution functions provided by neutron scattering methods, more information is often required to understand even simple molecular systems. These typically contain upwards of five atomic components distributed across two or more molecular species. In a system containing N structurally distinct atomic sites, $N(N+1)/2$ partial structure factors are required to fully describe all the possible pair correlations. Even if every element had suitable isotopes to enable neutron scattering difference techniques to be applied, and many key species such as oxygen do not, it would be prohibitively time-consuming and expensive to undertake the number of experiments required for a full extraction. If we are to make progress in this area we are thus led to investigate how computer-based techniques can assist us to maximize our access to the information content of our data and help us build chemically sensible structural models that are consistent with all the information we have available.

The advent of classical atomistic simulation techniques such as Monte Carlo and molecular dynamics methods have provided us with a readily accessible means to model and visualize in great detail the structure of liquids and disordered materials (Allen & Tildesley 1987). These techniques allow us to produce models of atomic and molecular configurations from which it is a trivial procedure to calculate any structural quantity of interest, for example, any desired partial pair-distribution function. However, as with all simulation methods, it is important to bear in mind that their reliability is ultimately governed by the quality of the inter-atomic potentials used to describe the interactions of the atoms and molecules (Stone 1997). Usually, when deriving appropriate potentials to describe a system, many trial simulations are required, each using slightly different parameters. Each trial is tested against how well a particular quantity of interest is reproduced, for example, internal energy or pressure. Once this is reproduced to a satisfactory level, the potential may then be further tested against its suitability for modelling an appropriate solid-state crystalline structure and its associated lattice properties (Allen & Tildesley 1987).

Classical simulation methods have now been extensively applied to studies of molecular liquid systems and by and large have been very successful in capturing the global characteristics and reproducing the thermodynamic properties against which their governing potentials were parameterized. However, when

detailed structural analysis of many of these models is performed, it is frequently found that considerable differences exist when they are compared with experimentally derived measures such as pair-distribution functions (Bowron *et al* 1998a,b).

The observation that simulations are successful at capturing global characteristics against which they are parameterized, while missing out the detailed structure, suggests a natural line of development for the method. Instead of refining the parameters of the inter-atomic potentials against a global entity, we could refine them against experimentally available structural data. This is one of the key underlying principles of the EPSR technique (Soper 1996, 1998, 2001), which aims to produce structural models that are consistent with diffraction data.

The starting point of the EPSR method is a standard Monte Carlo simulation. First, a box of atoms and molecules representative of the molecular liquid system of interest is prepared, storing the atomic coordinates and ensuring that fundamental physico-chemical constraints are obeyed—in particular the atomic density of the system and the shape of the molecular units. At this initial stage of the refinement process, the atoms and their interactions are usually described by standard Lennard–Jones parameters plus charges, all taken from the simulation literature. Once the box of atoms has been produced, the Monte Carlo procedure is started, and the structure of the system allowed to equilibrate. As in a standard simulation, the internal energy and pressure of the system are monitored to ensure that the parameters of the initial seed potentials are reasonable.

As the simulation will ultimately be used to model experimentally derived structure data, it is important to ensure that the molecules represented in the model box are not rigid, as real molecular liquids are not made of inflexible molecules that are instantaneously structurally identical. In the EPSR procedure each molecule is given sufficient flexibility in both bond angles and bond lengths to adopt all the expected conformations that exist within a real system, yet each is sufficiently constrained so as to avoid dissociation or the adoption of chemically unreasonable forms.

Once a satisfactory starting configuration of molecules has been produced under the standard Monte Carlo procedure, the link between the atomic potentials and experimentally measured structure is established through a potential of mean force $\psi_{\alpha\beta}(r) = -k_{\text{B}}T \ln[g_{\alpha\beta}(r)]$. There are several methods for doing this and here we outline the simplest, operating directly on the pair-distribution functions $g_{\alpha\beta}(r)$. The more recent implementations of the EPSR method operate directly on the experimentally measured total or partial structure factors $S(Q)$ (Soper 2000), to reduce the number of potential sources of uncertainty that can result from imperfect Fourier transform methods. However, tests have shown that regardless of the method selected—real-space or Q -space refinement—the final structural results are indistinguishable from each other, for example compare Bowron *et al.* (1998c, 2001).

The starting box of molecules is created using the standard starting potentials $U_{\alpha\beta}^{\text{O}}(r)$ between sites α and β . From this box, it is now possible to calculate a pair-distribution function $g_{\alpha\beta}(r)$. This in turn is used to generate a perturbed potential energy function $U_{\alpha\beta}^{\text{N}}(r)$ by the simple relationship

$$U_{\alpha\beta}^{\text{N}}(r) = U_{\alpha\beta}^{\text{O}}(r) + (\psi_{\alpha\beta}^{\text{D}}(r) - \psi_{\alpha\beta}(r)), \quad (2.6)$$

$$U_{\alpha\beta}^{\text{N}}(r) = U_{\alpha\beta}^{\text{O}}(r) + k_{\text{B}}T(\ln(g_{\alpha\beta}(r)/g_{\alpha\beta}^{\text{D}}(r))). \quad (2.7)$$

The superscript labels N and O, for new and old, illustrate how a new potential energy perturbation function is derived at each subsequent iteration of the Monte Carlo cycle, based on the structural configuration arrived at in the previous iteration. The Monte Carlo simulation then continues to run under this new potential, which equation (2.7) shows is linked to the structure derived from experimental data, $g_{\alpha\beta}^D(r)$. A repetitive procedure of evaluating a new potential and allowing the simulation to develop for a period of time is then established and continued until

$$U_{\alpha\beta}^O \approx U_{\alpha\beta}^N \quad (2.8)$$

and hence

$$g_{\alpha\beta}(r) \approx g_{\alpha\beta}^D(r). \quad (2.9)$$

At this point, the model is a reasonable representation of the data, and structural information can be obtained from the molecular configurations generated. This is carried out by continuing the Monte Carlo procedure under the final potential while ensemble average functions are calculated.

The empirical potentials generated by this method are more complex than specific site-site functions; they relate to all the types of atom pairs that contribute to the pair-distribution functions from which they are derived (Soper 1996, 1998, 2001). The relative strength of interaction for a given atomic pair that is subject to this composite potential is determined by atomic concentration and neutron scattering weight, in a manner analogous to the magnitude of the pair's contribution to the composite partial pair-distribution function. The calculated empirical potentials themselves are, in essence, perturbation functions that are applied to the initial seed potentials. The functional forms of the empirical potential include the effects of many-body interactions via the impact they have on the experimentally derived pair-distribution functions, and as such we note that the finally produced potentials (seed + empirical potential) are not easily parameterized in terms of the variables used to define the initial seed potentials based on Lennard-Jones forms and charges (Soper 2001).

In principle, it is possible to perform the EPSR procedure on a system where we have performed only one diffraction experiment and thus have access to only a single total pair-distribution function (Soper 1998). The net result would then be a structure that is consistent with our initially imposed physico-chemical constraints of density and molecular shape, but with the reliability of the structural configurations subsequently evaluated from the model, determined largely by the relative strengths that relevant atomic pair correlations made to the input experimental total pair-distribution function. The true strength of the method is really shown when the refinement procedure is applied to a series of measurements, such as an isotopic substitution set, that are made on the system. The method provides a systematic framework in which a range of complementary structural information is brought together to generate a model consistent with all the applied constraints. In general, the more complementary data that are available to the model, the more reliable the final model is likely to be. In particular, the isotopic methods allow us to weight more heavily, within the model generation process, those structural motifs associated with the local environment of specific substituted atom sites and molecular units. The combination of this very specific information with the imposed constraints on the shapes and sizes of the molecules enables us to probe beyond the directly measured correlations and investigate

the likely configurations that would occur between non-isotopically labelled groups and atoms in the system.

As the EPSR procedure has allowed us to produce snapshots of molecular ensembles and as all the atomic coordinates of the model are known, it is a trivial matter to calculate any specific site-site pair-distribution function we desire. Furthermore it requires little extra effort to calculate the orientational-correlation functions, $g(r, \omega_1, \omega_2)$, that are crucial for correctly characterizing molecular interactions (Soper 1998; Finney & Bowron 2002).

3. EXPERIMENT

The example selected to illustrate the potential of combined neutron diffraction and empirical potential-structure refinement is the structure of a dilute aqueous solution of tertiary butanol. The intermolecular interactions in such alcohol-water solutions are of fundamental interest as both an archetypal water-amphiphile system subject to significant hydrophobic effects (Bowron *et al.* 1998c, 2001), and as a widely used reaction supporting solvent medium (Bowron & Díaz Moreno 2002). Both the alcohol and water molecules have atomic sites that are suitable for isotopic substitution with hydrogen and deuterium, which makes the system ideal from the neutron scattering viewpoint.

The system investigated here is a 0.02 mol fraction solution of tertiary butanol in water. At this concentration, the current capabilities of available neutron scattering instruments are sufficient to allow us to use H/D isotopic substitution techniques to specifically probe the 'solvent-solvent' interactions from the viewpoint of the water hydrogen sites, and the 'solute + solvent-solute + solvent' interactions from the viewpoints of the water hydrogen sites, the alcohol methyl hydrogen sites and the alcohol hydroxyl group hydrogen sites. The structural correlations between non-substituted atom pairs are accessible through the composite partial structure factors to which the isotopic substitution method gives us access.

The five experiments that were performed were made on the following isotopic mixtures:

- (i) $(\text{CD}_3)_3\text{COH}$ in H_2O ,
- (ii) 1 : 1 mixture of $(\text{CD}_3)_3\text{COH}$: $(\text{CD}_3)_3\text{COD}$ in a 1 : 1 mixture of H_2O : D_2O ,
- (iii) $(\text{CD}_3)_3\text{COD}$ in D_2O ,
- (iv) 1 : 1 mixture of $(\text{CH}_3)_3\text{COH}$: $(\text{CD}_3)_3\text{COD}$ in a 1 : 1 mixture of H_2O : D_2O ,
- (v) $(\text{CH}_3)_3\text{COH}$ in H_2O .

In experiments (i)–(iii), the isotopic substitution was effectively performed on the water hydrogen sites, though also, owing to inevitable in-solution isotopic exchange, on the alcohol hydroxyl hydrogen sites. In experiments (iii)–(v), the isotopic exchange was performed on all hydrogen sites in the solution. These five experiments thus provide the functions that have subsequently been used to guide the empirical potential refinement of the solution structure.

In the second system of interest, the 0.02 mol fraction tertiary butanol-water solution was perturbed by the addition of a 0.01 mol fraction sodium chloride. The same series of isotopic substitutions was used, thus providing a set of directly comparable constraints for the independent

structure-refinement procedure of the salt-containing system.

The EPSR procedure was performed on molecular ensembles containing 300 water molecules and 6 alcohol molecules in a cubic box of side length *ca.* 21.5 Å. In the salt-containing system three Na⁺ and three Cl[−] ions were included. The full details of both of the experiments and the corresponding structure refinements can be found elsewhere, along with the associated neutron scattering weights governing the partial structure factors and structural weights of the resultant models (Bowron & Finney 2002, 2003).

Throughout the subsequent discussion of the resulting structural models, the following atom-site-labelling convention has been used. The central carbon of the alcohol molecule is assigned the label CC, the methyl group carbon sites C, the methyl group hydrogen sites M, the alcohol hydroxyl group oxygen atom O, and the alcohol hydroxyl group hydrogen H. The water hydrogen sites are labelled HW, and the water oxygen sites OW. In the NaCl-perturbed system, the Na⁺ cation is labelled Na, and the Cl[−] anion is labelled Cl.

4. RESULTS

(a) *The pure alcohol–water solution*

Figure 1 shows the partial pair-distribution functions generated by the EPSR procedure that most closely approximate to the molecular-centres correlations between the two in-solution species. The alcohol–alcohol correlation function $g_{CC-CC}(r)$ tells us that in this system there is a preferred separation distance of *ca.* 6.2 Å, which when integrated between 4.3 and 7.5 Å corresponds to each alcohol molecule having, on average, 1.5 ± 0.5 other alcohol molecule neighbours—a result consistent with the formation of dimeric and trimeric alcohol species in solution. The water–water correlation function $g_{OW-OW}(r)$ is very reminiscent of that found in pure water. The integral of the first peak in this function, between 2.5 and 3.4 Å, corresponds to each water molecule having, on average, 4.6 ± 0.2 neighbours, i.e. slightly larger than the 4.0 that would be expected for a perfect tetrahedral network. As this is exactly what is found from the measured values obtained for the pure liquid, the first-neighbour water geometry as shown by this function seems to be similar to that found in pure water. The same does not appear to be the case for the second peak from 3.4 to 5.5 Å, which describes the arrangement of the second neighbours: the single broad peak observed in pure water (Soper 2000) is replaced by one showing two sub-peaks. In this solution, therefore, the second neighbourhood appears to be more structured than in the bulk. The third centres' function $g_{CC-OW}(r)$ has a form between 3 and 6.5 Å that is consistent with the alcohol's having two hydration zones, a short-range zone centred at *ca.* 3.7 Å and a slightly longer-range region centred at *ca.* 5.0 Å. The integrated coordination numbers for these two features correspond to *ca.* 2.3 ± 0.6 water molecules in the region from 3.2 to 4.1 Å from the central carbon atom of the alcohol molecule, and 25.1 ± 2.0 water molecules in the region from 4.1 to 6.5 Å from the alcohol centre.

Although providing us with very useful quantitative information about the interaction between the two

Figure 1. Molecular centres pair-distribution functions for (a) alcohol–alcohol (CC–CC), (b) water–water (OW–OW) and (c) alcohol–water (CC–OW) pair correlations in a 0.02 mol fraction solution of tertiary butanol in water determined by the EPSR procedure.

molecular species, it remains very difficult to directly interpret the simple pair-distribution functions in terms of the favoured types of molecular interaction. To improve on this situation we now calculate a function called the spatial-density function $g(r, \Omega)$ (Svishchev & Kusalik 1993). Instead of the simple radial average represented by the pair-distribution function, this is a simplified form of a full orientational-correlation function. It represents a three-dimensional map of the probability density for finding a selected molecular species around another, as a function of both radial distance from (r), and orientation ($\Omega = \theta, \phi$) relative to, the selected central molecule. In its simplest form, the density function is calculated as an

Figure 2. (a) The spatial-density function showing the 10% isosurface for finding a central carbon of a tertiary butanol molecule (CC) in the distance range 5–10 Å about another tertiary butanol molecule in a 0.02 mol fraction tertiary butanol–water solution. (b) The spatial-density function showing the 40% isosurface for finding a water molecule oxygen atom (OW) in the distance range 2.3–3.1 Å about another water molecule in a 0.02 mol fraction tertiary butanol–water solution. The isosurface is defined so as to display the regions that are most likely to be filled based on the specified fraction of the total number of neighbouring molecules that constitute an entire radial shell.

average over all possible orientations of the neighbouring molecule. For identical pairs, the functions are fully symmetric and thus even with full orientational averaging it is still usually possible to infer information about the nature of molecular interactions.

In figure 2a we show the spatial-density functions corresponding to the probability of finding an alcohol centre (CC) around any arbitrarily selected alcohol molecule. The isosurface is set to show the most probable 10% of such interactions in the distance range from 5 to 10 Å. Noting the symmetry of the function we can infer that tertiary butanol molecules in aqueous solution have a most preferential molecular configuration that is consistent with direct non-polar group to non-polar group interactions. This is a classic example of direct hydrophobic association, i.e. reducing the exposure of the non-polar methyl groups to the polar solvent water environment (Kauzmann 1959). Figure 2b shows the distribution of water molecules around another water molecule in the range 2.3–3.1 Å. This illustrates the classic near-tetrahedral coordination of water, the two lobes aligned with the OW–HW directions defined by the central molecule corresponding to neighbouring water molecules accepting hydrogen bonds from the central one (Soper 1994). The two lobes below the central molecule correspond to the water molecules donating hydrogen bonds to the central molecule's electron lone-pair regions. Figure 2 thus illustrates how, by calculating the spatial-distribution function, we can greatly enhance the quantitative information regarding distance and coordination number accessible from the single first peaks in the pair-distribution functions. With respect to the nature of the water–water interaction, the water–water spatial distribution function confirms what was implied by the first peak in figure 1b: its first-neighbour geometry is essentially the same as that found in pure water. Furthermore, the nature of the second-neighbour perturbation mentioned previously can be further examined (Bowron *et al.* 2001).

To use spatial-density functions to assist us to interpret the more complex unlike-molecule-pair interactions such

as alcohol–water and water–alcohol correlations, we can calculate corresponding functions where we place each molecular species at the origin of the figure in turn. For example, figure 3a shows the most probable location of water molecule centres around a tertiary butanol molecule in the distance range 3–4.2 Å, while figure 3c shows the most probable location of tertiary butanol molecular centres around a water molecule in the same distance range. Figure 3a thus allows us to assign the first peak in figure 1c to water molecules hydrating the alcohol molecule's hydroxyl group, while figure 3c tells us that the most favourable interactions with that group in the selected distance range are hydrogen-bonding configurations. The intensity parallel to the HW–OW–HW plane that is just below the equatorial plane of the central water oxygen is consistent with the water molecule's accepting a hydrogen bond from the alcohol's OH group, while the lobes in the OW–HW direction correlate with the solvent molecule's donating a hydrogen bond to the alcohol's OH group.

The hydration shell of the remaining exposed non-polar cap of the molecule is specifically shown in figure 3b and can clearly be associated with the second peak in the $g_{\text{CC-OW}}(r)$ pair-distribution function (figure 1c). Figure 3d shows that the prominent density lobes tend to be parallel to the HW–OW–HW plane. This indicates that the dipole moment of the water molecules in the non-polar hydration region of the alcohol head group tends to be tangential to a sphere centred on the central carbon of the alcohol, and highlights the preference for tangential water geometries in this hydrophobic hydration shell (Soper & Finney 1993; De Jong *et al.* 1997). However, the fact that this density is continuous around the water molecule shows that there is still considerable structural disorder within the shell (Soper & Finney 1993).

(b) *The salt-perturbed alcohol–water solution*

Figure 4 shows the alcohol and water molecule centres pair-distribution functions refined from the data collected on the system perturbed by sodium chloride. These can

Figure 3. Spatial-density functions corresponding to the hydrophilic (*a,c*) and hydrophobic (*b,d*) hydration zones of the amphiphilic tertiary butanol molecule in a 0.02 mol fraction tertiary butanol–water solution. (*a*) The spatial-density function showing the 20% isosurface for finding a water molecule oxygen atom (OW) in the distance range 3–4.2 Å about a tertiary butanol molecule. (*b*) The spatial-density function showing the 20% isosurface for finding a water molecule oxygen atom (OW) in the distance range 4.2–6.0 Å about a tertiary butanol molecule. (*c*) The spatial-density function showing the 20% isosurface for finding a central carbon atom of a tertiary butanol molecule (CC) in the distance range 3–4.2 Å about a water molecule. (*d*) The spatial-density function showing the 20% isosurface for finding a central carbon atom of a tertiary butanol molecule (CC) in the distance range 4.2–6.0 Å about a water molecule.

be compared with those obtained on the salt-free system shown in figure 1. The alcohol–alcohol centres function shown in figure 4*a* is strikingly different from the no-added-salt case (figure 1*a*). Whereas in the simple binary system there appeared only one favoured inter-alcohol correlation distance, the addition of salt clearly separates this into two features: the first favoured correlation distance remains at *ca.* 6 Å, but a second develops, centred at *ca.* 8.5 Å. Integration of the two features, between 4.3 and 7.5 Å and between 7.5 and 10.0 Å, gives corresponding coordination numbers of 0.8 ± 0.5 and 1.8 ± 0.8 alcohol molecules.

Figure 4*b* shows the OW–OW pair distribution, which, as in the no-added-salt case, looks largely similar to that found for pure water. However, there again seems to be a degree of perturbation in the distance range 3.4–5.5 Å. The integral of the first peak is identical to the no-salt case, and similar observations apply in both cases. Figure 4*c* displays a very similar form to figure 1*c*, but some disruption to the first peak at 3.7 Å is apparent; in the salt-perturbed case the peak has become more of a shoulder-

like feature on the broad second peak corresponding to the hydrophobic hydration shell. The coordination numbers obtained on integrating this function are comparable to the no-salt case at 2.2 ± 0.3 and 25.8 ± 2.0 , respectively.

In figure 5 we turn our attention to the like-molecules spatial-density functions. Figure 5*a* corresponds to the alcohol–alcohol centres distribution. It shows that the addition of a small amount of sodium chloride has perturbed the nature of the amphiphilic molecular interactions in a major way. Instead of just the simple lobe above the methyl group region of the central alcohol molecule (shown in figure 2*a*), we now have that lobe (but reduced in size), plus a series of other lobes associated with the hydroxyl group of the alcohol molecule. These interactions are found to correspond to direct and bridged polar interactions between the alcohol molecules (Bowron & Finney 2002, 2003). The distance ranges of these sets of lobes correspond to the two peaks in the $g_{CC-CC}(r)$ molecular centres distribution function (figure 4*a*), and the associated coordination numbers allow us to infer that the probability of direct non-polar group to non-polar group

Figure 4. Molecular centres pair-distribution functions for (a) alcohol-alcohol (CC-CC), (b) water-water (OW-OW) and (c) alcohol-water (CC-OW) pair correlations in a 0.02 mol fraction solution of tertiary butanol in water perturbed by the addition of a 0.01 mol fraction of NaCl. The functions were determined by the EPSR procedure.

interactions has been significantly weakened. Figure 5b shows the water oxygen distribution around a water molecule. As in figure 2b this structure reflects the continued dominance of the expected tetrahedral bonding motif for a water network. This is shown to be unaffected by the addition of salt in the first-neighbour distribution, underlining once more the apparent robustness of this first-neighbour water geometry.

Figure 6 allows us to investigate the effect that the addition of salt has on the alcohol hydration shell around

both the polar hydroxyl group and in the hydrophobic region around the methyl groups. Figure 6a shows us that the previously ordered hydration of the hydroxyl group (figure 3a) is somewhat disrupted: the three individual lobes evident in the no-added-salt system become merged into a single trefoil-shaped lobe at the same isosurface level corresponding to the most favourable 20% of molecular interactions in the 3–4.2 Å range. Figure 6c, however, shows us that generally the nature of the water-hydroxyl group interaction is still the same (compare with figure 3c), though the magnitude of the water molecule H-bond-accepting configuration lobes appears weaker by virtue of the slightly smaller lobes evident at the same isosurface probability level as figure 3c. Figure 6b illustrates the increased spatial range accessible for water molecules to hydrate the non-polar methyl groups of the alcohol molecule, as the previous hole at the top of the no-salt distribution now shows considerable intensity at the same isosurface level as figure 3b. Figure 6d shows a remarkable increase in ordering in this hydrophobic hydration shell when compared with figure 3d, as the previously continuous band of intensity has now split clearly into two large ‘tangential’ hydration lobes and perpendicularly oriented configurations relative to the carbon at the centre of the alcohol molecule.

Figure 7 shows the pair-distribution functions corresponding to the interaction of the two molecular species with the ions. Figure 7a shows the average radial distribution of Na^+ around the central carbon of the alcohol molecule. Integration of the two peaks evident in this function tells us that between 3.9 and 5.8 Å one finds 0.2 ± 0.2 sodium cations, whereas between 5.8 and 9.5 Å there are 1.2 ± 0.4 . Figure 7b relates to the distribution of Cl^- . It is clear that the interaction of the two ionic species is markedly different and integration of $g_{CC-Cl}(r)$ between 3.3 and 7.0 Å tells us to expect 0.8 ± 0.4 chlorine anions in this range. Figure 7c,d shows the respective interactions of Na^+ and Cl^- with water. Integrating the functions gives the ion hydration numbers in this alcohol-water solution. The results are 4.2 ± 1.1 for Na-OW between 2.1 and 3.0 Å, and 4.9 ± 1.3 for Cl-OW between 2.8 and 3.6 Å.

The tertiary butanol-ion spatial-density functions illustrated in figure 8 highlight the very different natures of the anion and cation interactions with this amphiphile. The chlorine anions are strongly associated with the polar-hydroxyl-group region of the alcohol molecule while the sodium cations are found more distantly, beyond the hydrophobic hydration shell of the alcohol.

Figure 9 shows us classic illustrations of ion hydration by water (Neilson & Enderby 1982, 1983; Neilson *et al.* 1985). The negative charge of the chlorine anion leads to very strong interaction with the positively charged water hydrogen sites, while the positively charged sodium cation sits snugly in the region associated with the lone-pair electrons of the water molecule.

5. DISCUSSION

The results from these two systems allow us to establish the key structural motifs that illustrate the ways that water molecules interact in a range of atomic and molecular environments. Starting with the interaction of water molecules with themselves, we see that in both systems the

Figure 5. (a) The spatial-density function showing the 10% isosurface for finding a central carbon of a tertiary butanol molecule (CC) in the distance range 5–10 Å about another tertiary butanol molecule in a 0.02 mol fraction tertiary butanol–water solution with a 0.01 mol fraction of NaCl. (b) The spatial-density function showing the 40% isosurface for finding a water molecule oxygen atom (OW) in the distance range 2.3–3.1 Å about another water molecule in a 0.02 mol fraction tertiary butanol–water solution with a 0.01 mol fraction of NaCl.

Figure 6. Spatial-density functions corresponding to the hydrophilic (a,c) and hydrophobic (b,d) hydration zones of the amphiphilic tertiary butanol molecule in a 0.02 mol fraction tertiary butanol–water solution with a 0.01 mol fraction of NaCl. (a) The spatial-density function showing the 20% isosurface for finding a water molecule oxygen atom (OW) in the distance range 3–4.2 Å about a tertiary butanol molecule. (b) The spatial-density function showing the 20% isosurface for finding a water molecule oxygen atom (OW) in the distance range 4.2–6.0 Å about a tertiary butanol molecule. (c) The spatial-density function showing the 20% isosurface for finding a central carbon atom of a tertiary butanol molecule (CC) in the distance range 3–4.2 Å about a water molecule. (d) The spatial-density function showing the 20% isosurface for finding a central carbon atom of a tertiary butanol molecule (CC) in the distance range 4.2–6.0 Å about a water molecule.

Figure 7. Tertiary butanol–salt and water–salt pair-distribution functions. (a) Alcohol–cation (CC–Na), (b) alcohol–anion (CC–Cl), (c) water–cation (OW–Na) and (d) water–anion (OW–Cl) pair correlations in a 0.02 mol fraction solution of tertiary butanol in water perturbed by the addition of a 0.01 mol fraction of NaCl. The functions were determined by the EPSR procedure.

Figure 8. Ion–amphiphile interactions in a 0.02 mol fraction tertiary butanol–water solution with 0.01 mol fraction of NaCl. (a) The spatial-density function showing the 20% isosurface for finding a Cl^- anion (Cl) in the distance range 3–5 Å about a tertiary butanol molecule. (b) The spatial-density function showing the 20% isosurface for finding a Na^+ cation (Na) in the distance range 3–8 Å about a tertiary butanol molecule.

structural form of the first-neighbour environment of water molecules is, as expected, dominated by the classic close-to-tetrahedral hydrogen-bonding motif (figures 2b and 5b). In this motif, each water molecule participates, on average, in two H-donor and two H-acceptor interactions and the average separation of each water molecule

oxygen site is 2.80 ± 0.05 Å. The same is found to be the case for pure bulk water. The apparent robustness of this first-neighbour environment should be noted. It appears to remain unaffected by the presence of other polar groups, non-polar groups and ions. The same, however, cannot be said for the water–water second neighbourhood,

Figure 9. Ion–water interactions in a 0.02 mol fraction tertiary butanol–water solution with a 0.01 mol fraction of NaCl. (a) The spatial-density function showing the 20% isosurface for finding a Cl^- anion (Cl) in the distance range 2–5 Å about a water molecule. (b) The spatial-density function showing the 20% isosurface for finding a Na^+ cation (Na) in the distance range 2–5 Å about a water molecule.

where the addition of non-polar groups clearly does perturb the structure at longer distances.

In both the no-NaCl and NaCl-perturbed systems the interactions of the water molecules with the alcohol molecules display distinctive interaction motifs with the various functional groups of the amphiphile. Clearly, the solvent water molecules that coordinate the alcohol hydroxyl group do so by standard hydrogen-bonding interactions leading to a CC–OW separation distance between 3.0 and 4.2 Å (the shoulder in both figures 1c and 4c). The apparent distortion of the water molecule H-bond-accepting lobes of figures 3c and 6c when compared with the water–water interactions in figures 2b and 5b are simply a result of the relative position of the central carbon atom of the tertiary butanol molecules, whose probability densities are plotted in figures 3c and 6c. It is clear that for both the water–water interactions and the OH–water interactions, hydrogen-bonding configurations are highly directional. In contrast to this high directionality of the hydroxyl group solvation, the hydration of the alcohol molecule's methyl groups is far less orientationally defined. In the simple salt-unperturbed case, the solvent water molecules display a preference to be located in an equatorial girdle about the alcohol molecule at a centres separation distance between 4.2 and 6.0 Å. The form of the intermolecular interaction that corresponds to figure 3d has a strong component that relates to a solvation-shell water geometry where the dipole moment of the water molecule lies tangentially to the non-polar surface defined by the methyl groups of the alcohol molecule. This is shown by the significant lobes of intensity parallel to the HW–OW–HW plane of the central water molecule (figure 3d) and this geometry is consistent with standard models of hydrophobic hydration. However, the fact that there is continuous intensity around the water molecule shows us that there is also considerable orientational disorder in this hydrophobic hydration girdle. This is a finding that is consistent with the increasing body of experimental data and simulation results on the structure of hydrophobic hydration shells that cautions against the over-simplistic conventional, near crystalline, models that overemphasize the motif of tangential water molecule geometries

(Soper & Finney 1993; Bowron *et al.* 1998d; Ashbaugh *et al.* 2003; Finney *et al.* 2003).

The apparent hole above the hydrophobic hydration girdle of figure 3b is a direct consequence of the strong interaction between the alcohol molecules mediated by their non-polar methyl groups (figure 2a). In the salt-free system, these interactions appear to be a classic example of the hydrophobic interaction, the effective ‘force’ that is argued to drive the association of non-polar groups in aqueous solution to minimize their exposure to the solvent. If the alcohol molecules associate strongly in this way, they clearly reduce the probability for water molecules to hydrate the corresponding alcohol–alcohol contact region and the non-polar hydration shell is thus not closed. Figure 5a illustrates that the addition of sodium chloride to the system has a very marked effect on the nature of the alcohol–alcohol correlations. The salt induces a remarkable increase in alcohol–alcohol interactions mediated via their polar hydroxyl groups (Bowron & Finney 2002). Such interactions inevitably increase the exposure of the non-polar methyl groups to the solvent environment and correlate directly with increased water capping shown in figure 6b. Also very noticeable is the resulting change in the nature of the water–alcohol hydrophobic interactions shown in figure 6d. The addition of sodium chloride to the system appears to enhance the orientational order in the hydrophobic hydration shell. The ‘tangential’ hydration lobes parallel to the HW–OW–HW plane remain a marked feature of the correlations, but the in-plane interactions also increase their definition, becoming independent features at the same isosurface probability level as figure 3d. The reason for these interesting changes requires further investigation. One possible interpretation might be in terms of an enhanced hydrophobic effect in the system resulting from the decrease in disorder (entropy) of the hydration shell, which would correspond to a reduction in the compatibility of the alcohol molecules with the solvent water network. This would be consistent with the known salting out of aqueous tertiary butanol phases at higher concentrations.

To gain further insight into how the addition of sodium chloride to the tertiary butanol–water system so radically

changes the nature of the alcohol–alcohol correlations, it is necessary to consider the interaction of the Na^+ and Cl^- ions with the amphiphilic alcohol molecule. An examination of figure 8*a,b* clearly shows the very different nature of anion and cation interactions. The Cl^- anion closely associates with the alcohol hydroxyl group and adopts a position that is optimally located to form a direct bridge between two hydroxyl groups from two tertiary butanol molecules and effectively linking the groups via a O–H–Cl–H–O interaction (Bowron & Finney 2002, 2003). By contrast, the Na^+ cations seem to prefer a relatively distant interaction with the alcohol molecules, sitting largely outside the hydrophobic hydration shell in the equatorial plane of the alcohol molecule. These interactions serve to highlight the difficulties that have to be faced in any attempt to rationalize the complex nature of ion–amphiphile interactions. The fact that the alcohol hydroxyl group can formally accept two hydrogen bonds, yet only donate one, simplistically suggests that the small and positive Na^+ cation (ionic radius 0.95 Å) would be the preferred ion to associate closely with the alcohol hydroxyl group. However, it is the larger chloride (Cl^-) anion (ionic radius 1.8 Å) that dominates this local environment. The Na^+ ion appears to prefer tight hydration by the smaller solvent water molecules and consequently keeps its distance from the hydrated alcohol molecules. Its presence outside the non-polar hydration shell may be one reason for the enhanced tangential orientations of the water molecules on the addition of salt (figure 6*d*).

Finally, as demonstrated in § 4, the structural motifs of ion hydration in this system are exactly as would be expected (Neilson & Enderby 1982, 1983; Neilson *et al.* 1985; Yamaguchi & Soper 1999). The ions coordinate the water molecules in a manner consistent with the known charge distribution of the molecule; i.e. the slightly positive hydrogen sites are favoured by the anion and the slightly negative oxygen sites by the cation.

6. CONCLUSIONS

Collectively, these results demonstrate the high degree of structural insight into molecular liquids that can now be obtained by combining computer-modelling methods with experimentally obtained structural data. Here, the selected examples have allowed us to illustrate many key structural motifs that underlie the physical, chemical and biochemical properties of aqueous systems. In particular, they give a clear insight into how the water molecule interacts with other—polar, non-polar and ionic—groups, as well as with other water molecules in the presence of other solutes.

The main structural conclusions we can draw from these results are as follows.

- (i) The classic near-tetrahedral self-coordination of solvent water molecules in the bulk solvent is retained in these solutions. The strong directionality of the solvent water molecule's first-neighbour hydrogen-bonded interactions appears to be resilient and robust. The same cannot be said of the water molecule's second-neighbour structure, where the addition of non-polar groups results in a structural ordering of the second-neighbour shell. This

ordering is, however, fully consistent with the constraints imposed by the robust first-neighbour motif.

- (ii) A polar hydroxyl group is hydrated by highly oriented hydrogen-bonding interactions, illustrating how the larger alcohol molecule is orientationally accommodated relative to the solvent molecule.
- (iii) The water molecules in the hydrophobic hydration of methyl groups adopt a largely *tangential orientation* relative to the non-polar surface. Importantly, however, a considerable range of other *non-tangential* configurations is allowed. Again, these water arrangements appear to be accommodated within the first-neighbour average-tetrahedral structural motif.
- (iv) Non-polar groups placed in an aqueous environment associate through direct contact. The structural motifs observed are what would be expected in a system in which hydrophobic interactions are significant. The degree of solute clustering observed even at this low concentration is perhaps greater than would have been expected for a system that is generally thought to be well mixed.
- (v) Anion and cation interactions with amphiphilic alcohol molecules are complex, as is the resulting polar-group-mediated perturbation of hydrophobic amphiphile–amphiphile interactions.
- (vi) Ionic interactions with polar groups on an amphiphile can produce enhanced exposure of non-polar groups and associated increases in the extent of structural order in a hydrophobic hydration shell.
- (vii) The clear illustration has been given of how the hydration structure of ions is dominated by charge-mediated interactions with the polar water molecules.

The obvious complexity and range of interactions illustrated in even such simple liquid systems clearly illustrate the fact that for detailed progress to be made in our understanding of in-solution chemical and biochemical reactions, a comprehensive structural insight will be required. Though still restricted to case-by-case studies, the methods presented here at last provide us with a general and widely applicable means by which it is possible to investigate the subtle balance and interplay between polar/charged and non-polar atomic and molecular interactions. This information could ultimately allow us to both predict and control chemical and biochemical reactions in the liquid state.

REFERENCES

- Allen, M. P. & Tildesley, D. J. 1987 *Computer simulation of liquids*. Oxford: Clarendon.
- Ashbaugh, H. S., Asthagiri, D., Pratt, L. R. & Rempe, S. B. 2003 Hydration of krypton and consideration of clathrate models of hydrophobic effects from the perspective of quasi-chemical theory. *Biophys. Chem.* **105**, 323–338.
- Bowron, D. T. & Diaz Moreno, S. 2002 The structure of a concentrated aqueous solution of tertiary butanol: water pockets and resulting perturbations. *J. Chem. Phys.* **117**, 3753–3762.
- Bowron, D. T. & Finney, J. L. 2002 Anion bridges drive salting out of a simple amphiphile from aqueous solution. *Phys. Rev. Lett.* **89**, 215 508–1–215 508–4.

- Bowron, D. T. & Finney, J. L. 2003 Structure of a salt-amphiphile-water solution and the mechanism of salting out. *J. Chem. Phys.* **118**, 8357–8372.
- Bowron, D. T., Finney, J. L. & Soper, A. K. 1998a The structure of pure tertiary butanol. *Mol. Phys.* **93**, 531–543.
- Bowron, D. T., Finney, J. L. & Soper, A. K. 1998b ERRATUM The structure of pure tertiary butanol. *Mol. Phys.* **94**, 249–251.
- Bowron, D. T., Finney, J. L. & Soper, A. K. 1998c Structural investigation of solute-solute interactions in aqueous solutions of tertiary butanol. *J. Phys. Chem. B* **102**, 3551–3563.
- Bowron, D. T., Filipponi, A., Roberts, M. A. & Finney, J. L. 1998d Hydrophobic hydration and the formation of a clathrate hydrate. *Phys. Rev. Lett.* **81**, 4164–4167.
- Bowron, D. T., Finney, J. L. & Soper, A. K. 2001 Temperature dependence of the structure of a 0.06 mole fraction tertiary butanol-water solution. *J. Chem. Phys.* **114**, 6203–6219.
- De Jong, P. H. K., Wilson, J. E. & Neilson, G. W. 1997 Hydrophobic hydration of methane. *Mol. Phys.* **91**, 99–103.
- Enderby, J. E. & Neilson, G. W. 1979 X-ray and neutron scattering techniques applied to aqueous solutions. In *Water: a comprehensive treatise* (ed. F. Franks), pp. 1–46. New York: Plenum.
- Finney, J. L. & Bowron, D. T. 2002 Experimental determination of the structures of complex liquids: beyond the PDF. In *From semiconductors to proteins: beyond the average structure* (ed. S. J. L. Billinge & M. F. Thorpe), pp. 219–244. Dordrecht, The Netherlands: Kluwer.
- Finney, J. L. & Soper, A. K. 1994 Solvent structure and perturbations in solutions of chemical and biological importance. *Chem. Soc. Rev.* **23**, 1–10.
- Finney, J. L., Bowron, D. T., Daniel, R. M., Timmins, P. A. & Roberts, M. A. 2003 Molecular and mesoscale structures in hydrophobically driven aqueous solutions. *Biophys. Chem.* **105**, 391–409.
- Gray, C. G. & Gubbins, K. E. 1984 *Theory of molecular fluids, vol. 1: fundamentals*. Oxford: Clarendon.
- Kauzmann, W. 1959 Some factors in the interpretation of protein denaturations. *Adv. Protein Chem.* **14**, 1–63.
- Neilson, G. W. & Enderby, J. E. 1982 The structure around nitrate ions in concentrated aqueous solutions. *J. Phys. C* **15**, 2347–2352.
- Neilson, G. W. & Enderby, J. E. 1983 The structure of an aqueous-solution of nickel chloride. *Proc. R. Soc. Lond. A* **390**, 353–371.
- Neilson, G. W., Schiøberg, D. & Luck, W. A. P. 1985 The structure around the perchlorate ion in concentrated aqueous solutions. *Chem. Phys. Lett.* **122**, 475–479.
- Plumridge, T. H. & Waigh, R. D. 2002 Water structure theory and some implications for drug design. *J. Pharm. Pharmacol.* **54**, 1155–1179.
- Soper, A. K. 1994 Orientational correlation function for molecular liquids—the case of liquid water. *J. Chem. Phys.* **101**, 6888–6901.
- Soper, A. K. 1996 Empirical potential Monte Carlo simulation of fluid structure. *Chem. Phys.* **202**, 295–306.
- Soper, A. K. 1998 Recent advances in structure refinement for liquids and disordered materials. In *Local structure from diffraction* (ed. S. J. L. Billinge & M. F. Thorpe), pp. 59–83. New York: Plenum.
- Soper, A. K. 2000 The radial distribution functions of water and ice from 220 to 673 K and at pressures up to 400 Mpa. *Chem. Phys.* **258**, 121–137.
- Soper, A. K. 2001 Tests of the empirical potential structure refinement method and a new method of application to neutron diffraction data on water. *Mol. Phys.* **99**, 1503–1516.
- Soper, A. K. & Finney, J. L. 1993 Hydration of methanol in aqueous solution. *Phys. Rev. Lett.* **71**, 4346–4349.
- Stone, A. J. 1997 *The theory of intermolecular forces*. Oxford: Clarendon.
- Svishchev, I. M. & Kusalik, P. G. 1993 Structure in liquid water—a study of spatial distribution functions. *J. Chem. Phys.* **99**, 3049–3058.
- Yamaguchi, T. & Soper, A. K. 1999 Observation of chloride-ion hydration in high-temperature liquid and supercritical water by spherical harmonic expansion analysis. *J. Chem. Phys.* **110**, 3529–3535.

Discussion

J. R. Helliwell (*Department of Chemistry, University of Manchester, Manchester M13 9PL, UK*). Could you please clarify why you picked the mixtures you chose, especially with respect to their relevance to life and hydration? For example, are the concentrations (of NaCl and tertiary butyl alcohol) chosen good, i.e. relevant average values? In this meeting's context what next experiments are you planning?

D. T. Bowron. The mixtures were chosen to enable us to study the chemical interactions between our selected amphiphilic solute and ionic co-solute, under conditions of full hydration and below the concentration at which the addition of salt would cause phase separation of the alcohol and water components. The functional molecular groups of the model system are widely found in biomolecular systems and are subject to similar kinds of chemical interaction in both the model and *in vivo* environments. The reported results are the first of this kind obtained on such a complicated four-component liquid mixture, alcohol, water, cation and anion, and consequently have to serve as the baseline for future investigations. How relevant the selected concentration is, for understanding the interactions between similar groups in a biochemical context, is difficult to address. The baseline system is a homogeneous mixture at a fixed composition, yet in a biochemical system local fluctuations in water and salt concentration, at the chemical functional group length scale, are expected—other investigations over a broader range of concentration and thermal conditions are thus required and planned.

B. Halle (*Department of Biophysical Chemistry, Lund University, SE 22100 Lund, Sweden*). When you use the EPSR approach to construct orientational pair-correlation functions from neutron scattering data that only provide isotropic structure factors, how sensitive are the results to the details of the force field used in the simulation? Stated differently, would a simulation with the same force field, but without the constraints imposed by the neutron scattering data, yield similar orientational pair-correlation functions? If not, what do you think is the most important deficiency in the force field?

D. T. Bowron. The ability of the EPSR method to reconstruct orientational pair-correlation functions from experimental data has been tested a number of times and the results show that the pair- and higher-order correlation functions derived from experimental data are reasonably insensitive to the precise form of the seed potentials used to describe the atomic and molecular interactions (details can be found in Soper (2001)). If the model construction is performed without the constraints imposed by the experimental data, i.e. purely based on an assumed set of interatomic potentials, the results are generally found to favour a different balance between the charged and

non-polar interactions within the system. Charged interactions tend to dominate at a level beyond that supported by the experimental data. Within the current implementation of EPSR it is not possible to specifically isolate characteristics of the atomic potentials. The experimental data are inverted by a potential of mean force derived from a measured pair-distribution function (pdf). In consequence this empirical potential also contains information about the effect that many-body interactions have on the pairwise terms, and in most cases it is also the weighted sum of the atomic terms characteristic of the measured pdf. Methods for separating the specific contributions of particular atomic species to the pairwise interaction potentials are currently under investigation and development.

J. L. Finney (*Department of Physics and Astronomy, University College London, Gower Street, London WC1E 6BT, UK*). The data give hints about the molecular mechanisms relating to interesting processes such as the hydrophobic interaction and salting out. Do these help us in identifying other liquids in which these processes might also operate?

D. T. Bowron. The selected molecular mixtures were chosen as they illustrate many of the fundamental chemical interactions that are common to aqueous solvent systems. These are interactions that essentially induce or mediate key physico-chemical properties that are essential for biological action, such as solute-solvent hydrogen bonding and the hydrophobic interaction. The principal characteristics observed for charged, polar and non-polar group hydration are thought to be largely transferable to

other systems where water is the bulk solvent, and the observed interactions that occur between the non-polar and polar groups of the larger solute molecules are also thought to broadly capture the most preferable chemical contacts for such units. At the direct molecular contact level the transferability of such interactions to other liquids is related to the chemistry of the functional groups, i.e. it is most likely that hydroxyl groups will still be subject to hydrogen-bonding interactions and that methyl groups remain non-polar. However, care should be taken if the aim is to extrapolate the relevance of the observed interactions to larger solute and solvent systems. More complex molecules will be subject to greater steric effects such as molecular flexibility and the relative placement of neighbouring functional groups. These are very likely to play a significant role in the ultimately favoured molecular configurations. The observed tendency for anionic species to apparently drive solute reorientation and salting out is a very interesting observation, but at the current moment in time, based on this single study, it would be premature to claim that the effect is generic to the majority of salt-induced phenomena. So, the results do help us set a qualitative baseline for the types of interactions that can be expected in other liquids, but as yet there is no quantitative means, short of direct measurement, for evaluating different molecular mixtures.

GLOSSARY

EPSR: empirical potential structure refinement