5 COSOLVENT EFFECTS ON NEUTRAL HYDROLYSIS

M.F. Lensink, T. Rispens, J.B.F.N. Engberts, H.J.C. Berendsen,
Cosolvent effects on neutral hydrolysis, to be submitted.

We have studied, by means of Molecular Dynamics (MD) simulations, the effect of adding hydrophobic cosolutes, tert-butanol (t-BuOH) and ethanol (EtOH), on the neutral hydrolysis of p-methoxyphenyl dichloroacetate (MPDA) in water. Determined were shifts in a defined pre-equilibrium. The pre-equilibrium was defined as a spatial distribution of reacting molecules satisfying specific geometric constraints. The constraints applied represent the configurations from which the reaction can take place (Reactive Conformation, RC). The results are not sensitive to the specific values of these constraints. The experimentally found rate retardation was reproduced; a linear correlation between the amount of added cosolute and the logarithm of the reaction rate was found. The percentage of RC's in pure water was found to be 14.6%, complementing earlier work performed in this group (M.F. Lensink, J. Mavri, H.J.C. Berendsen, J. Comp. Chem. 20 [1999], 886), leading to a calculated rate constant of $k_{\text{calc}} = 3.9 \times 10^{-3} \text{ s}^{-1}$ (exp: $k_{\text{obs}} = 3.03 \times 10^{-3} \text{ s}^{-1}$). The molecular structure of the RC was looked at more extensively, investigating correlation between the position of participating molecules.
Chemical reactivity in the liquid phase is a subject that continues to offer many challenges to both experimental and theoretical chemists and physicists. Because of its complexity, a general comprehensive theory is lacking. Nevertheless, a variety of theories do exist, the most useful of them still being Transition State Theory (TST)\textsuperscript{41-43}, despite its long existence. The main advantage of TST is the direct link with thermodynamics, making it highly useful for studies of chemical reactivity in diverse media. The differences in reaction rates found for a reaction performed in different solvents can be straightforwardly linked to the differences in chemical potentials of the reactants (Initial State, IS) and an activated complex in these solvents\textsuperscript{273-275}. This activated complex (Transition State, TS) is the structure corresponding to the highest Gibbs energy along the reaction path. Within TST the problem of chemical reactivity therefore reduces to a thermodynamic problem.

For chemical reactions, the solvent is of prime importance. Besides different pure solvents, mixtures of solvents may be used as reaction medium. In this way information about both the reaction and the medium itself can be obtained. The most interesting mixtures are mixtures where one of the solvents is water. Water is undoubtedly one of the most studied molecules. It has many peculiar properties, one of them being its behavior as a solvent for organic compounds: they are often not very soluble in water, giving rise to the term “hydrophobic”. Nevertheless, life itself is based upon organic chemistry in water. Therefore there is a lot of interest in the study of intermolecular interactions and reactivity of organic compounds in water and aqueous mixtures. Many a study has focused in this respect on so called hydrophobic interactions\textsuperscript{276-280}, where added hydrophobic cosolute to a reaction occurring in water interacts favorably with specific parts of the reacting solute, (de)-stabilizing e.g. the IS with respect to the TS. For these processes, it is still unclear as to what occurs at the atomic level.

In this paper we will focus on solvent effects on reaction rates where the solvent consists of water and a low percentage of a simple, hydrophobic, alcohol. For these low concentrations the properties of the mixtures closely resemble those of pure water, perturbed, often in a systematic way. We will therefore in fact use the term “cosolute” for the disturbing molecule(s), “solvent” for the water, and “solute” for the reacting molecules.

In the thermodynamic description, the properties of another species dissolved in the mixture, i.e. a reactant, can, due to its low concentration, be treated likewise: the rate at different concentrations of alcohol, compared to the rate at zero concentration, and the change in rate may be explained as the difference in chemical potentials of IS and TS as caused by interactions between IS and TS with alcohol\textsuperscript{281, 282}. Typically, in cases where the solvent is actively participating in the reaction, this can be translated into a changed chemical activity. Alternatively, in the molecular or atomic description one can focus on the local effects that arise upon adding cosolute, e.g. a changed local water density around the reactive center, versus a changed macroscopic water activity as a lowered volume fraction of water. For this purpose, one can use Molecular Dynamics (MD) simulations, where thermodynamic properties follow from computer simulations, based upon a simple atom-atom pair-interaction model. The evolution of atomic coordinates forms trajectories that can give insight into the molecular dynamics of the system at the atomic level. Their elegance and ease-of-use make MD a valuable tool in the study of molecular behavior of organic compounds in aqueous mixtures.

Many simulations of pure water and pure alcohol systems have been performed\textsuperscript{283-288}. In spite of the inherent simplicity of the pair-interaction potentials that are commonly used, and the fact that these were fitted to the properties of the pure components, the use of traditional combination rules for pair-interaction parameters yields an overall good agreement with experiment\textsuperscript{286-288}.

Most simulation studies of water/alcohol mixtures focus on water/methanol, or other systems...
where the balance between hydrophobic hydration of the methyl group and hydrogen bonding of the hydroxyl group lies such, that the solute cannot be considered purely hydrophobic286–292. Studies of larger alcohols, or other purely hydrophobic solutes, in water reveal that these solutes will show a certain amount of aggregation. This is an entropic effect to compensate for the more ordered water structure around the hydrophobic substance. Water molecules orient themselves tangentially with respect to an apolar surface to keep the number of disrupted hydrogen bonds as low as possible286. In addition, both by simulation291–295 and experiment296–299 a slower translational motion of water is found. Recently, simulation studies of liquid t-BuOH298, as well as t-BuOH in the aqueous phase299 have been performed. Although these studies find a significant enhancement in water structure upon increasing the t-BuOH concentration, they find little aggregation in the dilute phase. For an excellent review on hydrophobic effects we refer to Ref. 280.

The reaction we have studied is the neutral hydrolysis of p-methoxyphenyl dichloroacetate (MPDA) in aqueous solution247,249,250, for different concentrations of added cosolutes tert-butanol (t-BuOH) and ethanol (EtOH). In the water-catalyzed reaction — the neutral hydrolysis — the reactive complex (see Fig. 5.1) contains two water molecules, with a proton transfer between these two water molecules being the rate limiting step. Concerted with this a covalent bond is formed between the nucleophilic water oxygen and the ester carbonyl carbon. The observed pseudo first-order rate constant in pure water is $k_{\text{obs}} = 3.03 \times 10^{-3} \text{ s}^{-1}$ at 298 K. The effect of alcohols on the rate of this hydrolysis reaction has been determined301,302. For low concentrations of solute and cosolute the assumption can be made that the (co)solute molecules interact with each other independently and a linear dependence of the observed rate constant on the alcohol percentage is expected281,282,303,304.

**Thermodynamic Description**

Consider Fig. 5.2. We assume a pre-equilibrium to exist within the Initial State (IS), with the reaction taking place through a so called Activated State (AS), where two water molecules are
electrostatically bound to MPDA. The IS then consists of a Relaxed State (RS) in equilibrium with the AS. The AS is defined as having two water molecules within a certain distance and angle of the carbonyl functionality of the solute (see also Fig. 5.3 on the facing page). Obviously the AS represents a percentage of the entire ensemble comprising the IS, we will call this percentage \( x_{\text{reactive}} \). In this picture the reaction rate constant is given by

\[
k = \frac{k_B T}{h} x_{\text{reactive}} e^{-\Delta G_{\text{AS}} / RT} = x_{\text{reactive}} \times k_{\text{AS}}. \tag{5.1}
\]

Upon adding cosolute (indicated in Fig. 5.2 and subsequently in the text by the prime), the chemical potentials of the states are changed, resulting in a shift in the Gibbs energy. In this example the cosolute interacts favorably with the RS and slightly favorably with the AS, resulting in a positive \( \Delta \Delta G \) and an effective decrease in reaction rate constant \( k = x_{\text{reactive}} \times k'_{\text{AS}} \). For this work we will assume that the effect of cosolute on AS or TS is equal, i.e. \( \Delta \Delta G_{\text{AS}} = \Delta \Delta G_{\text{TS}} \). This is e.g. the case for systems where cosolvent effects are entirely governed by diffusion. This assumption enables us to use the calculated value\(^{174}\) of \( k_{\text{AS}} = 2.7 \times 10^{-3} \ \text{s}^{-1} \) and limit our survey to changes in \( x'_{\text{reactive}} \).

**Figure 5.2**  Schematic representation of the relative dispositions of the various states involved in the neutral hydrolysis reaction, with or without cosolute present. The Gibbs energy for the reaction is the weighted average over the two states comprising the IS: \( \Delta \Delta G_{1}^{0} = \frac{1}{2}(G_{\text{TS}} - G_{\text{RS}}) + \frac{1}{2}(G_{\text{TS}} - G_{\text{AS}}) \), with \( \frac{1}{2} \) corresponding to \( x_{\text{reactive}} \). Note that the proton transfer between the two water molecules and the nucleophilic attack of the water on MPDA are concerted.
Figure 5.3  The frame of axes used for defining the reactive conformations according to the constraint parameters described in the text

Computational Details

All MD simulations were performed with the GROMACS 1.6 package\textsuperscript{15}, using the GROMOS-87 force field\textsuperscript{12} and the SPC water model\textsuperscript{183}. Simulations lengths were typically between 1.0 and 2.0 ns. The systems consisted of a periodic box with, besides the solute and an amount of cosolute, approximately 700 water molecules, and were coupled to a temperature bath at 298 K and a pressure bath at 1 bar with coupling constants of 0.1 ps and 1 ps respectively\textsuperscript{264}. A spherical cut-off of 1 nm was employed. Bond lengths were constrained using the LinCS algorithm\textsuperscript{205}. Equations of motion were integrated by the Verlet leap-frog integration scheme with a time step of 2 fs. Partial atomic charges for the solute were determined by fitting them to the electrostatic potential in a solvent reaction field (AM1-SM1\textsuperscript{206}) and to the calculated dipole moment\textsuperscript{267}. Molecular geometry for t-BuOH was taken from the OPLS force field\textsuperscript{265}.

A reactive conformation (RC) was defined as having the nucleophilic water molecule within a certain distance and angle from the carbonyl functionality (based upon possible orbital overlap during the activation process) and a second water molecule, that is acting as a general base, within a certain distance and angle from the first one (based upon conventional hydrogen bond criteria), as illustrated in Fig. 5.3. The carbonyl planar group is situated in the xy-plane. Furthermore the constraint $r_3 > r_1$ was applied, i.e. that the distance carbonyl oxygen - water hydrogen be greater than the distance carbonyl carbon - water oxygen.
Chapter 5

Results

<table>
<thead>
<tr>
<th>mole-% t-BuOH:</th>
<th>r_&lt;sup&gt;n&lt;/sup&gt;</th>
<th>θ_&lt;sup&gt;n&lt;/sup&gt;</th>
<th>x</th>
<th>x'</th>
<th>x'/x</th>
<th>x'/x'</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 %</td>
<td>0.36</td>
<td>0.31</td>
<td>26</td>
<td>0.146</td>
<td>0.77</td>
<td>0.65</td>
</tr>
<tr>
<td>0.5 %</td>
<td>0.35</td>
<td>0.31</td>
<td>26</td>
<td>0.101</td>
<td>0.78</td>
<td>0.67</td>
</tr>
<tr>
<td>1.0 %</td>
<td>0.34</td>
<td>0.31</td>
<td>26</td>
<td>0.0661</td>
<td>0.78</td>
<td>0.70</td>
</tr>
<tr>
<td>1.5 %</td>
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<td>0.31</td>
<td>26</td>
<td>0.122</td>
<td>0.79</td>
<td>0.65</td>
</tr>
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<td>26</td>
<td>0.137</td>
<td>0.78</td>
<td>0.64</td>
</tr>
<tr>
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<td>0.29</td>
<td>26</td>
<td>0.119</td>
<td>0.79</td>
<td>0.66</td>
</tr>
<tr>
<td>3.0 %</td>
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<td>0.31</td>
<td>30</td>
<td>0.149</td>
<td>0.78</td>
<td>0.65</td>
</tr>
<tr>
<td>3.5 %</td>
<td>0.36</td>
<td>0.31</td>
<td>20</td>
<td>0.138</td>
<td>0.77</td>
<td>0.63</td>
</tr>
</tbody>
</table>

* without constraint r<sub>3</sub> > r<sub>1</sub>.

Table 5.1 Results for MPDA in water/t-BuOH, r<sub>3</sub> > r<sub>1</sub>

Table 5.1 shows the dependence of the percentage of RC's on the constraint parameters. x stands for the percentage RC's in the pure water solution (x<sub>reactive</sub> in the Section on page 75), while x' stands for the same quantity in the water/t-BuOH mixture. The table shows that although the percentage RC's is rather significantly dependent on the constraint parameters, the ratio is not. Changes in the angle constraints led to a variation typically within 6%, while changes in the distance constraints led to a scattering within 10%.

The last row in Table 5.1 shows that the constraint r<sub>3</sub> > r<sub>1</sub> rules out a large number of RC's, while not changing the ratio x'/x. This is, however, a rather important constraint, since without it, the nucleophilic water molecule can have a hydrogen pointing towards the carbonyl group and the reaction would not take place. We will therefore in the text henceforth assume that this constraint is applied, unless mentioned otherwise. This constraint was also applied in Ref. 174 for our definition of an AS.

<table>
<thead>
<tr>
<th>mole-% alcohol</th>
<th>x'</th>
<th>x'/x&lt;sub&gt;1&lt;/sub&gt;</th>
<th>x'/x&lt;sub&gt;1&lt;/sub&gt;</th>
<th>ln(x'/x)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5 %</td>
<td>0.112</td>
<td>0.126</td>
<td>0.89</td>
<td>0.768</td>
</tr>
<tr>
<td>1.0 %</td>
<td>0.095</td>
<td>0.101</td>
<td>0.88</td>
<td>0.652</td>
</tr>
<tr>
<td>1.5 %</td>
<td>0.072</td>
<td>0.079</td>
<td>0.91</td>
<td>0.491</td>
</tr>
<tr>
<td>2.0 %</td>
<td>0.051</td>
<td>0.059</td>
<td>0.87</td>
<td>0.352</td>
</tr>
<tr>
<td>2.5 %</td>
<td>0.123</td>
<td>0.139</td>
<td>0.88</td>
<td>0.829</td>
</tr>
<tr>
<td>3.0 %</td>
<td>0.097</td>
<td>0.110</td>
<td>0.88</td>
<td>0.662</td>
</tr>
<tr>
<td>3.5 %</td>
<td>0.074</td>
<td>0.084</td>
<td>0.88</td>
<td>0.507</td>
</tr>
</tbody>
</table>

Table 5.2 Results of water/alcohol mixtures for the constraints r_<sup>n</sup> = 0.36 nm, θ_<sup>n</sup> = 45 deg, r_<sup>α</sup> = 0.31 nm, θ_<sup>α</sup> = 26 deg, r<sub>3</sub> > r<sub>1</sub>.

The results for a particular choice of constraint parameters are summarized in Table 5.2. The values of x' (fraction RC's in water/alcohol mixture) show that the presence of solute sig-
significantly decreases the number of RC's. $x'_{i1}$ stands for the fraction of RC's in the water/alcohol mixture where the constraint for the presence of a second water molecule is left out, i.e. it is assumed to be always present; the column $x'/x'_{i1}$ shows that in close to 90% of the cases when the nucleophilic water molecule is present at a reactive position there is in fact also a second water molecule present, that can pick up the proton. Since this percentage seems not to depend on the concentration of alcohol, one can conclude that the reaction retardation primarily stems from the effect the cosolute has on the nucleophilic waters and that there is hardly any effect on the waters acting as a general base.

In order to obtain insight into the steric effect the phenyl moiety may have on the occurrence of RC's, a correlation was defined between the position of the phenyl ring (indicated by the first arrow between the brackets), given the position of the RC (defined by the position of the nucleophilic water molecule and indicated by the second arrow), both relative to the MPDA carbonyl plane:

$$r_{corr} = -\frac{1}{2} \left( \frac{p(\uparrow | \downarrow) - p(\downarrow | \uparrow)}{p(\uparrow | \uparrow) + p(\downarrow | \downarrow)} \right),$$

e.g. $p(\uparrow | \downarrow)$ means the probability that the phenyl ring is located above the carbonyl plane ($\uparrow$), given a RC below that plane ($\downarrow$). No dependence on the presence of cosolute could be detected. Averaged over all the simulations an anti-correlation of 42.5% was found ($r_{corr} = -0.42(5)$).

Note that $r_{corr}$ ranges from -1 to 1, where a value of 0 indicates no correlation. One can conclude that the steric hindrance by the phenyl ring negatively affects the formation of any RC's on that side of the carbonyl plane.

To investigate the effect of the cosolute on the occurrence of any RC, or rather, on the presence of the nucleophilic water molecule, radial distribution functions (RDF's) were calculated for the t-BuOH and EtOH carbon atoms around the MPDA carbonyl carbon. The RDF's are calculated over all simulations. The RDF's show a tendency for the cosolute molecules to reside in the vicinity of the carbonyl group, with peaks at 0.5 nm [t-BuOH] and 0.45 nm [EtOH].

We also consider the “correlated” and “cross-correlated” “conditional” RDF's$^{(a)}$. Here “conditional” means the RDF given a RC. For the “correlated” conditional RDF the RDF is calculated for cosolute molecules residing on the same side of the ester carbonyl plane as the RC, while the “cross-correlated” conditional RDF shows the same for cosolute molecules given a RC on the other side of the carbonyl plane. In the conditional RDF's, the peaks are shifted by +0.05 nm, as well as lowered significantly. For the correlated conditional RDF, this can easily be explained since the required presence of a reactive water molecule can be seen to push the cosolute molecule sideways and thus slightly farther away, while in the cross-correlated conditional RDF the phenyl ring, which was found to be anti-correlated with the occurrence of a RC, would be responsible for this task. Interestingly, the effect of lowered density is more prominently present for the cross-correlation conditional RDF for t-BuOH. This can be explained by the smaller volume of EtOH cosolute molecules, resulting in them not being hindered by the phenyl moiety.

Fig. 5.4 shows the position of the nucleophilic water molecule oxygens, projected in the $x'y$-plane, for the pure water solution and two concentrations of added cosolute t-BuOH. No correlation was found between the position of a reactive water molecule relative to the ester molecule: within the zone of the applied constraints the RC's are distributed evenly. This is the case for both the pure water simulation, as well as in the presence of cosolute, indicating that the cosolute has no preference to bind to a specific region of the ester molecule.

In Fig. 5.5 the experimental values of $\ln(k/k_0)$ and calculated values of $\ln(x'/x)$ are plotted versus the molality of alcohol. Clearly a linear trend is found, as was predicted$^{281,282}$ and

$^{(a)}$ A better indication would be to calculate the RDF for water (satisfying the RC constraints) around the ester carbonyl carbon, since the presence of cosolute would show a decreased RDF first peak — a direct correlation (t-BuOH pushes the water away), while the investigation of t-BuOH RDF gives an indirect correlation.
Figure 5.4 Scatterplot showing the RC nucleophilic water oxygens in the xy-plane over a 1.6 ns MD simulation of a MPDA/water/t-BuOH system. The slightly empty region in the northwest corner corresponds to the carbonyl oxygen.

experimentally found. The magnitude of the rate retardation, indicated by the higher slope, is in both cases overestimated. This may be due to inaccuracies in the force field parameters for the alcohols or water-alcohol, causing an overestimated clustering, but it is more likely due to cosolvent effects on the TS.

Neutral Hydrolysis in Pure Water

We have simulated the neutral hydrolysis reaction in pure water. We assumed a pre-equilibrium, from which we then calculated the reaction rate by combining a Gibbs (free) energy calculation with a quantum dynamical simulation. The assumed pre-equilibrium contains two

Figure 5.5 Plots of \( \ln(k/k_0) \) (experiment) and \( \ln(x'/x) \) (calculated) versus alcohol molality. Linear fits were forced to pass through (0,0).
water molecules bound at an ionic distance (see also Fig. 5.1 on page 75). Ignoring the quantum dynamics (it is not relevant for this chapter), we can summarize that we calculated the reaction rate by calculating the Gibbs energy difference between IS and TS, applying Eyring theory to the obtained value, yielding a reaction rate of \( k_{\text{AE}} = 2.7 \times 10^{-2} \text{ s}^{-1} \). The constraint implied by our pre-equilibrium represents an additional Gibbs energy term, governed by diffusion. A simple MD simulation of MPDA in water, and counting the number of configurations over the ensemble that satisfy the applied constraints — we called this fraction \( x_{\text{reactive}} \), on page 75 — is enough for the correction. If we apply this correction, using the constraint parameter values from Table 5.2 on page 78 and the first row in Table 5.1, we substitute \( x_{\text{reactive}} = 0.146 \) in Eq. 5.1 on page 76 to get an overall rate constant of \( k_{\text{calc}} = 3.9 \times 10^{-3} \text{ s}^{-1} \), which is in excellent agreement with the experimentally observed value of \( k_{\text{obs}} = 3.03 \times 10^{-3} \text{ s}^{-1} \).

**Discussion**

Because water is a small molecule, certainly with respect to organic cosolvents like t-BuOH, one can easily make the misleading conclusion that cosolvent effects are large. However, were only volume exclusion the contributing factor, a rate retardation of only 15% of the experimentally found value would be found\(^{(b)}\). From this one can conclude that the rate retardation is not solely due to random encounters between the solute and cosolute molecules, but in fact there is a tendency for them to stick together, although no preference for a specific region of the solute molecule was found.

There is no effect on the water molecule to which the proton is transferred in the reaction, although the proton potential fluctuations are likely to be different for pure water and water-alcohol mixtures. These fluctuations are rate-determining for the proton transfer from the first water molecule to the second water molecule. At an atomic level one can visualize that there are not enough cosolute molecules to inhibit both sites of the nucleophilic water molecule. Higher concentrations of cosolute might start showing an effect, resulting in non-linearity of \( \ln(k/k_0) \) vs. mole-% cosolute. However, another effect then takes preference, namely that due to the relative size of t-BuOH vs. water, one can no longer talk about a diluted water phase, and self-association of t-BuOH will cause phase separation on a microscopic scale. One can also rationalize that there is no reason for a hydrophobic molecule like t-BuOH to stick to a water molecule. Rather, it will favor clustering with (the hydrophobic parts of) the solute and only an effect on the nucleophilic water molecule is found.

The change in reaction rate constant for the reaction studied is due to pairwise hydrophobic interactions, changing the Gibbs energy of the participating states. Apart from the changed chemical potential, also the changed — local — water activity (density) is responsible for the rate retardation. Within the present study only effects on the IS and the AS are taken into account. However, we expect the effect on TS to be small. The presence of alcohol might slightly favor the AS with respect to the TS; effects on the rate of proton transfer are small: the solvent deuteron isotope effect changes less than 10% upon adding 2.5 mole-% t-BuOH. In addition, a proton inventory study confirmed that no differences in the reaction pathway take place\(^{301, 302}\). Within the present approach a similar study of the effects of cosolute on the TS is difficult, considering the covalent nature of the bond that is in the TS formed between the solute and the nucleophilic water molecule, but mainly because of the nonexistence of force field parameters for the TS. Obviously, with a covalent bond formed, a t-BuOH-molecule cannot take the place of the nucleophilic water molecule. However, one can envisage that then any rate retardation effect

\(^{(b)}\) This is estimated by taking the log of the volume fraction of water in the mixture.
would manifest itself in the effect of cosolutes on the second water molecule, the one acting as a general base. As we concluded in the discussion of Table 5.2 on page 78, this is a small effect.

The proton potential energy curves and their evolution in time will change as a result of the added cosolutes. Pure water is more capable of stabilizing the participating states in the reaction, notably the more polar ones. Also the proton dynamics will change, resulting in a different rate constant. This affects the slopes of Fig. 5.5 on page 80, where we assumed zero effect on the TS. It is possible that this change is responsible for the different slope of these lines. The effect the cosolute has on the proton potentials and the quantum dynamical proton transfer and thus the transfer rate constant needs further investigation.

In the study of molecular behavior of organic compounds in aqueous mixtures, where chemical reactivity is not an issue, but thermodynamic properties and shifts in equilibria are, Molecular Dynamics (MD) simulations are a useful tool. Care should be taken since at the molecular level one is looking at tiny effects, which means that long simulations are required to obtain sufficient sampling of the quantities being studied. Moreover, the results are highly sensitive to the atomic topology used to describe the molecules. When force field parameters are slightly off, an overestimated clustering of alcohols might occur. This need not lead to other than linear effects, as long as the influence of each cosolute molecule is additive, and independent from other cosolute molecules, but a deviation from linear behavior would be expected to occur already at lower concentrations.