ELSEVIER

Biophysical Chemistry 51 (1994) 279–289

Biophysical Chemistry

Role of hydrogen bonds in hydrophobicity: the free energy of cavity formation in water models with and without the hydrogen bonds

B. Madan [1], B. Lee *

Laboratory of Molecular Biology, Division of Cancer Biology, Diagnosis and Centers, National Cancer Institute, National Institutes of Health, Bldg. 37, Rm. 4B15, Bethesda, MD 20892, USA

Received 7 November 1993; accepted 5 March 1994

Abstract

The free energies of cavity formation in water with and without hydrogen bonding potential were computed from the results of a set of Monte Carlo simulation calculations on pure liquid TIP4P water model and on the same model but with the electrostatic charges turned off (Lennard-Jones liquid). The free energies of cavity formation in the Lennard-Jones liquids are higher than or approximately equal to those in TIP4P water, depending, respectively, on whether the Lennard-Jones size parameter σ is set equal to 3.15 Å, which is the value of σ for TIP4P water, or to 2.8 Å, which is the commonly assumed value for the oxygen–oxygen distance between two hydrogen-bonded water molecules. This result indicates that changes in the hydrogen-bonded structure of water and/or in the orientational degree of freedom of water are not essential features in the production of the large free energy change upon cavity formation.

Key words: Hydrophobicity; Water structure; Cavity free energy; Simulation

1. Introduction

Non-polar molecules dissolve freely in non-polar liquids but their solubility in water is exceedingly small. Surprisingly, the molecular mechanism for this well-known phenomenon, called hydrophobicity in this article, is still a matter of much debate. Since Frank and Evans [1], the hydrophobicity has been attributed to the formation of low entropy structures, originally called the 'iceberg', around the solute molecules in water. However, many computer simulation studies performed in the past decade [2–8] indicate that an ice-like structure formation does not occur in water at room temperature. Also, the integral equation theory of Pratt and Chandler [9], which is highly successful in reproducing the experimental thermodynamic quantities of hydration, works without assuming any particular struc-

---

* Corresponding author.

[1] Present address: Department of Biochemistry and Biophysics, University of Pennsylvania, Philadelphia, PA 19104, USA.

SSDI 0301-4622(94)00049-P

ture formation around the added solute molecule beyond the structure that is already present in pure water. The orientational degree of freedom of water molecules immediately surrounding the solute molecule is obviously different from that of those in the bulk. This difference has only recently been quantitatively, albeit approximately, translated into the entropy change [10,11]. However, it has been suggested that such an entropy change is always accompanied by a compensating enthalpy change so that its effect on the free energy change, and therefore on the solubility, is small [12–16].

Whereas the 'iceberg' hypothesis attempts to explain hydrophobicity in terms of the hydrogen bonding property of the water molecules, an entirely different argument is possible based on the small size of water molecules. It has been known for a long time [17,18] that the essence of hydrophobicity is in the difference in the work of cavity formation in water and in non-aqueous solvents, probably because the van der Waals interaction between an apolar solute and the solvent is approximately the same in all liquids at ordinary temperature and pressure [19]. The work of cavity formation depends on the size of the solvent molecules; if the volume density is the same, it is more difficult to produce a cavity of given size in the liquid of small molecules than in that of larger molecules. This is because the empty space in the liquid of smaller molecules is expected to be broken into correspondingly smaller pieces than that in the liquid of larger molecules. Thus it is more difficult to find a piece of empty space large enough to accommodate a given size cavity if the molecules of the solvent are small than if they are large. Another way of looking at the same effect is to recognize that the work of cavity formation in a liquid must depend on the ratio between the size of the cavity to that of the solvent particles, since the latter defines the length scale of the system. Therefore a cavity of given size becomes effectively bigger when the size of the solvent molecules is reduced. It has been argued [20–22] that this effect is large enough to account for essentially all of the free energy difference observed in the phenomenon of hydrophobicity.

In an effort to assess the importance of the latter effect, Pohorille and Pratt [23] computed and compared the free energies of cavity formation in water with those in various organic solvents such as *n*-hexane, *n*-dodecane, *n*-undecyl alcohol, chloroform, and carbon tetrachloride. They concluded that, even though the fraction of free volume is larger in water than in the organic solvents, the free volume in water is indeed distributed in smaller packets, consistent with what is expected from the small size of water molecules. Therefore, it is harder to find large cavities in water than in the organic liquids. Using scaled particle theory, they estimated that this effect amounts to 80% of the total free energy of cavity formation of 2.0 Å size. Guillot et al. [24] also calculated the free energy of cavity formation in water by simulation and similarly concluded that it is only about 16% higher than the scaled particle theory result for the Ar- and methane-sized cavities.

Another way to shed light on the relative importance between above two competing possibilities is to compute and compare the free energies of cavity formation in a model water and in a reference liquid, which is just like the model water but with the hydrogen bonding potential turned off. If the hydrogen-bonded structure formation and/or the loss of orientational degree of freedom of water molecules were the important factor in determining the hydrophobicity, the free energy of cavity formation would be much lower in the liquid with the hydrogen bonds turned off. On the other hand, if the size were the controlling factor, the cavity free energies would be similar in the two liquids. Since the cavity free energy is simply related to the number of natural cavities that occur in the pure liquid [14], such a study can be made from pure liquid simulations alone without explicit introduction of a hard sphere [23].

Tanaka [25] studied such pure systems. Although he did not report the values of the free energy of cavity formation itself, it can be seen from his cavity distribution data that the free energy of cavity formation is *higher* in the *reference* liquid. This strongly indicates that solvent molecular size is the overriding feature for the hydrophobic effect. More recently, Pratt and Pohorille [26] studied similar systems, concentrating mainly on the contact correlation function $G(\lambda)$, which is basically the rate of change of the free energy with respect to the cavity size. They found differences in the values of this function for the model water and the reference liquid. However, the differences appear to be small compared to the size of the hydrophobic effect. These results of Tanaka [25] and Pratt and Pohorille [26] will be discussed further when they are compared with the results of this paper.

In the present study, we computed the free energies of cavity formation in a model water liquid, TIP4P [27], and in a reference liquid, which is the same TIP4P model but with the electrostatic interaction turned off. Consistent with Tanaka's [25] data, we find that the free energy of cavity formation is higher in this reference liquid than in water. We trace the reason for this behavior to the increase in the effective size of the solvent molecules when the hydrogen bonding potential is turned off. When the size of the solvent molecules is adjusted so that it is the same as that of the effective size of hydrogen-bonded water molecules, the cavity free energies become comparable for the two liquids.

2. Method

Only the pure liquid systems were simulated. The Helmholtz free energy, ΔA, of cavity formation was calculated from the ensemble of pure liquids using the following formula [14]:

$$\Delta A = -kT \ln\langle\zeta\rangle, \qquad (1)$$

where ζ is the ratio of the cavity volume to the total volume of the system, the angled brackets indicate the ensemble average, and other symbols have their usual meanings. The volume ratio ζ was calculated numerically by counting the number of cubic grid points that fall within a cavity of given size over those in the entire system [23,26]. The grid interval used for this calculation was 0.55 Å. A smaller interval of 0.2 Å was tried with TIP4P water and found to yield indistinguishable results.

Two related, but different, measures of cavity size are used in this article. One is the center-to-center distance R. This is defined so that the *centers* of the solvent molecules are excluded from a spherical region of radius R [28]. The second measure is the cavity radius, r_c, which is defined as the radius of the spherical region from which any *part* of any solvent molecules is excluded. If the solvent molecules are hard spheres of radius r_1, the two definitions of cavity size are related by $R = r_c + r_1$. The advantage of using the center-to-center distance, R, is that it is defined independent of the radius of the solvent molecules, which is often assumed to have different values by different authors. On the other hand, the size of a cavity is more naturally defined by r_c. For instance, when the free energies of cavity formation in solvents of different sizes are compared, the comparison should be made at the same value of r_c rather than of R. (We are indebted to Dr. Alex Rashin for this point.) In this work, we used R for all calculations, but the results are reported using both R and r_c. The value of the latter was calculated assuming that the effective hard core diameter of the solvent molecules is given by the Lennard-Jones σ parameter of the liquid potential for all liquids, including the TIP4P model water. The calculations were done for a set of R values in increments of 0.45 Å.

The liquid water model used was TIP4P of Jorgensen et al. [27]. The first reference liquid without the hydrogen bonding potential was identical to this water model except that all charges were set to zero. We shall refer to this liquid model, which consists of the Lennard-Jones potential alone, as LJ3.15. (The number 3.15 is the value of the σ parameter in the potential function.) Later, we also made calculations on a second reference liquid, which is identical to the first except that the value of the σ parameter was changed to 2.80 Å. This liquid will be referred to as LJ2.8. The simulations were done at 25°C and under the same constant volume for all three liquids. The same volume, rather than the same pressure, was chosen because the two reference liquids will have the density of a gas and behave like a gas if kept under the same pressure as the

TIP4P water at the same temperature. The ϵ parameter of the Lennard-Jones potential was kept the same as that for the TIP4P water. This is in contrast with Pratt and Pohorille [26] who used a reference liquid with altered ϵ parameter. The relation between this work and the work of Pratt and Pohorille will be discussed in section 4.

The computer program used to generate the liquid ensembles was the Monte Carlo simulation program BOSS, version 3.3 [29]. Initially a 750-molecule system of TIP4P water, which is supplied as a part of the BOSS program package, was equilibrated for 1.2×10^6 Monte Carlo steps at 25°C and 1 atm. One configuration towards the end of this constant pressure equilibration run was chosen as the starting structure for the constant volume runs for water and for the two different Lennard-Jones liquid models. This configuration had a density of 0.997 g cm^{-3}. Each system was again equilibrated at 25°C under the constant volume for 2.25×10^6 steps. The cavity calculations were made from ten consecutive runs that were then made on each system. Each run consisted of 3×10^6 Monte Carlo steps, from which 3000 evenly spaced configurations were selected for calculation. The precision of the calculation was estimated by observing the variation in the ten runs.

The computed results were compared with each other and with the results of the scaled particle theory calculations. The scaled particle theory was slightly modified as follows. In the original expression for the free energy of cavity formation [28], the third-order term in R was equated with the pressure–volume work in producing the cavity. In our constant volume system, however, the pressure–volume work is zero. We therefore modified the free energy expression to retain only up to the second-order term in R and redetermined the three coefficients by requiring that the free energy and its first two derivatives, computed separately for the two domains, $R \leqslant r_1$ and $R > r_1$, agree at $R = r_1$, where r_1 is the radius of the solvent molecule. The new expression turns out to be identical to that of ordinary scaled particle theory, but with the pressure terms deleted. All scaled particle theory calculations were done using the same number density and the two different solvent radii used in the Lennard-Jones liquid simulations.

3. Results

The results of the free energy calculation are shown in Table 1 and Fig. 1. The free energy could not be computed for values much higher than 40 kJ/mol because the free energy formula of Eq. (1) depends on observing naturally occurring cavities in the pure liquid and, when ΔA is 40 kJ/mol or more, the probability of observing a cavity at room temperature becomes 1.0×10^{-7} or less.

Comparing first the TIP4P water with the reference liquid of the same σ parameter, LJ3.15, it can be seen from Table 1 and Fig. 1 that, for R values greater than 1.35 Å, the free energy of cavity formation is always significantly *larger* in the *reference* liquid with no hydrogen bonds than in water. This phenomenon is almost certainly related to the effect of the hydrogen bonds on the distance of closest approach between molecules. The value of the Lennard-Jones size parameter, σ, used was 3.15 Å for both liquids, which is the value for the TIP4P water (27). However, because of the hydrogen bonds, the molecules in TIP4P water can come closer to one another than those in the Lennard-Jones liquid. This can be seen

Table 1
The free energy of cavity formation, ΔA, in kJ/mol as a function of the center-to-center distance, R, in three different model liquids. The numbers in parentheses are the standard deviations, given in units of the last digit of each given mean

R (Å)	0.45	0.90	1.35	1.80	2.25	2.70	3.15	3.60
TIP4P	0.032(0)	0.266(0)	1.04(0)	3.08(0)	7.07(3)	13.6(2)	23.2(9)	37(3)
LJ2.8	0.032(0)	0.266(0)	1.04(0)	3.19(2)	7.57(1)	14.8(3)	25.5(3)	41(3)
LJ3.15	0.032(0)	0.266(0)	1.04(0)	3.51(0)	9.72(1)	21.4(1)	40(2)	-

clearly from the radial distribution function, $g(r)$, for the two liquids shown in Fig. 2: the curve for the LJ3.15 (broken line) peaks at approximately 3.1 Å, near the value of the σ parameter, but the curve for the TIP4P water (solid line) peaks at approximately 2.8 Å. This difference in the distance of closest approach will affect the free energy of cavity formation because, if two systems, A and B, have equal volume and equal number of molecules of equal size, but molecules can come closer to each other in A than in B, then there will be more empty space in the former and hence less work will be required to form a cavity in A.

Since the distance of closest approach between water molecules is 2.8 Å, it may be argued that a more relevant Lennard-Jones system to use for comparison is one with its σ parameter reduced to 2.8 Å. We therefore ran identical Monte Carlo simulations using the LJ2.8 liquid, which is identical to LJ3.15 except that its σ parameter is reduced to 2.80 Å. The distance of closest approach in this liquid is similar to that in TIP4P water as indicated by the first peak of its $g(r)$, shown in Fig. 2 as the dotted line. The free energies of cavity formation for this liquid are included in Table 1 and in Fig. 1. The work of cavity formation is now quite comparable to that in the TIP4P water. The scaled particle theory calculations were also made using the same number density and the two different solvent radii. The results of these calculations are included in Fig. 1.

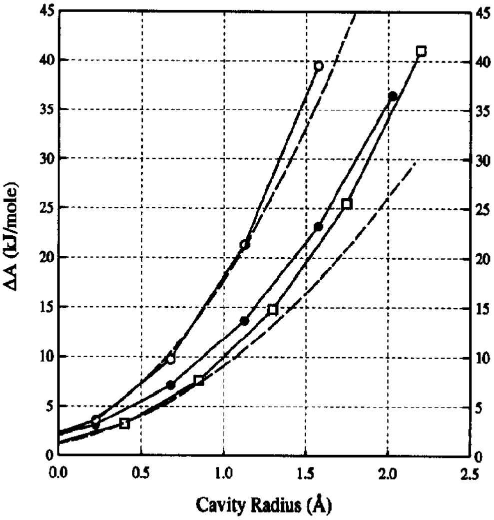

Fig. 1. The Helmholtz free energy of cavity formation, ΔA, as a function of the cavity radius, r_c, as calculated from the simulation using TIP4P water (solid line with solid circles), Lennard-Jones reference liquid with $\sigma = 3.15$ Å (solid line with open circles), and Lennard-Jones reference liquid with $\sigma = 2.8$ Å (solid line with squares) and as calculated by the scaled particle theory with $\sigma = 3.15$ Å (upper dashed line) and with $\sigma = 2.8$ Å (lower dashed line).

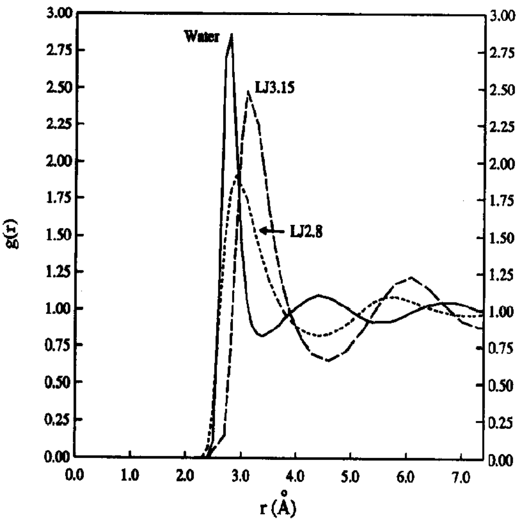

Fig. 2. The radial distribution function, $g(r)$, for TIP4P water (solid curve) and for the two Lennard-Jones liquids (broken line for $\sigma = 3.15$ Å and dotted line for $\sigma = 2.8$ Å).

4. Discussion

4.1. Two effective sizes of a water molecule

Results shown in Fig. 1 clearly indicate that the calculated free energy of cavity formation depends sensitively on the size of the solvent molecules. That the results of the scaled particle theory calculation is sensitive to the solvent radius used has been pointed out by many authors [23,30]. Therefore, when selecting the reference liquid for comparison with water, it is important that the size of the solvent molecules be properly chosen. This task is, however, complicated because of the fact that there are two sizes associated with water.

A common diameter used for water is 2.75 Å [17]. This value is near the first peak of the radial distribution function of water (Fig. 2) and represents the distance of closest approach between two water molecules, which are presumably hydrogen bonded. On the other hand, the Lennard-Jones size parameter, σ, ranges from 2.96 Å for the Bernal-Fowler model to 3.15 Å for the TIP4P model used here and 3.24 Å for the TIPS2 model [27]. These significantly larger values are presumably the size applicable when the water molecule interacts with other molecules through the van der Waals interaction without the hydrogen bond. When the cavity radius, r_c, is to be obtained from the center-to-center distance, R, it is this size, and not the distance of closest approach, that must be subtracted, since we are interested in the cavity as a substitute for a non-polar solute, which cannot hydrogen-bond with water.

4.2. *Turning off the hydrogen bonds does not reduce the cavity free energy*

Given this dual effective sizes of water, no Lennard-Jones liquid of one size will serve as a completely satisfactory reference liquid: The molecules in LJ3.15 are effectively too large and those in LJ2.8 too small. Therefore, if the size were the controlling factor, one expects that the cavity free energy in water will fall in between those for these two liquids. If, on the other hand, the hydrogen bond rearrangement and/or the restriction in the orientational degree of freedom were the essential features of hydrophobicity, the cavity free energies should be significantly smaller in both reference liquids than in water. The actual result, shown in Fig. 1, is that the free energy of cavity formation in water is significantly smaller than in LJ3.15 and is essentially the same as that in LJ2.8. Thus the calculation unambiguously favors the former over the latter as the mechanism of hydrophobicity.

This result is not restricted to the TIP4P model since Tanaka [25] obtains similar results using the TIPS model. For example, when his reported values of $\langle \zeta \rangle$ are converted to the free energies according to Eq. (1), the free energies of cavity formation in the TIPS water and in the 'simple liquid', which is TIPS model with the electrostatics turned off, are 17.9 and 20.1 kJ/mol, respectively, at the R value of 2.8 Å and 26.7 and 28.3 kJ/mol, respectively, at the R value of 3.1 Å. Again it can be seen that the free energy of cavity formation slightly *increases* upon turning off the electrostatic interaction in this water model. (Parenthetically we note that the free energy of cavity formation in the TIPS model is somewhat higher than in the TIP4P model at the same center-to-center distance, R (Table 1). However, if one assumes that the size of the solvent molecules in the TIPS model is given by the σ parameter for the oxygen atom, 2.82 Å, it is 0.33 Å smaller than that in the TIP4P model. Thus, if the free energies are compared at the same r_c value, they are less in the TIPS than in the TIP4P model.)

Numerous authors [17,19-21,30,31] made free energy calculations using the scaled particle theory and noted that it gives results that agree well with the experimental and simulation results on aqueous solutions. Scaled particle theory considers all molecules as spheres and contains no notion of the strongly directional hydrogen bonds. Therefore, it is expected to work on the Lennard-Jones liquids but its success on the aqueous system was surprising. On the other hand, Pratt and Pohorille [23,26] reported that the scaled particle theory underestimates the free energy when compared to that from the simulation results. Our results, shown in Fig. 1, are similar; the scaled particle theory tends to give smaller free energy values than the simulation results for the Lennard-Jones liquids of the same molecular size, particularly at large cavity sizes. Thus, a judicious choice of the solvent molecular size is needed for the success of the scaled particle theory. However, the fact that success can be obtained at all appears to be due to the same reason that the Lennard-Jones liquid gives a similar cavity free energy as the real water.

4.3. *High pressure versus stronger van der Waals interaction versus hydrogen bonds*

It should be noted that the Lennard-Jones reference liquids used here are under high pressures: According to the data of Nicolas et al. [32] on Lennard-Jones liquids, the pressures are estimated to be approximately 7000 and 10000 atm for LJ2.8 and LJ3.15, respectively. Unlike large hydrocarbon molecules, which form liquids at room temperature and atmospheric pressure, water molecules are even smaller than a methane molecule and do not exist in the liquid form without the hydrogen bonds. The high pressures are therefore necessary in order to preserve the number density after the hydrogen bonds are turned off.

If the cavity is formed under the constant *pressure*, the work of cavity formation, ΔG, is directly affected by the high pressure since it includes the $p\Delta V$ term. However, the work of cavity formation under the constant *volume*, ΔA, is not an explicit function of pressure. This can be seen from the following general expression for the free energy of cavity formation in any liquid [28]:

$$\Delta A(R) = kT\rho \int 4\pi\lambda^2 G(\lambda)\, d\lambda, \tag{2}$$

where ρ is the number density of the liquid, the integral is from $\lambda = 0$ to $\lambda = R$, R is the center-to-center distance measure of the size of the cavity, and $G(\lambda)$, not to be confused with the Gibbs free energy ΔG, is the cavity correlation function defined as the ratio of the number density of solvent molecules in contact with a cavity of size λ to that in the bulk. Thus, the constant volume work depends only on the temperature and the number density of the solvent at the surface of the cavity during its growth. Pressure affects the work only indirectly through this number density.

Pratt and Pohorille [26], who also compared the properties of cavity formation in water with that in a Lennard-Jones reference liquid, kept the pressure of the liquid comparable to that of ordinary water at room temperature by increasing the ϵ parameter of the Lennard-Jones potential by approximately 3.5-fold, from 0.155 kcal/mol in TIP4P to 0.541 kcal/mol in the reference liquid. Thus, this reference liquid has a considerably stronger van der Waals interaction than water. The σ parameter was also changed to 2.67 Å in order to preserve the distance of closest approach. They concentrated on the contact correlation function, $G(\lambda)$ and found definite differences in this function between this reference liquid and the TIP4P model water. However, according to their Fig. 4, the difference appears to be small relative to the total free energy of cavity formation.

Thus, we have three different liquids in which the free energy of cavity formation is comparable; water with its hydrogen bonds, the reference liquid of Pratt and Pohorille [26] with its strong van der Waals attraction, and the reference liquid of this study which is under a high pressure. The common feature among these three liquids is the similar bulk density and molecular size. Evidently, as long as the bulk density and molecular size are kept the same, the presence of hydrogen bonds is not required for the production of the high free energy of cavity formation.

4.4. Hydrogen bonds 'bunch up' the molecules

It is remarkable that the cavity free energy *increases* upon simply turning off the hydrogen bonds under constant volume. This means that, as long as the density is kept constant, water with no hydrogen bonds is even more hydrophobic than real water and that the presence of the hydrogen bonds actually *reduces* the hydrophobicity. It is clear from the results of this study that this happens because hydrogen bonds enable water molecules to come closer together than if they were not present. The effect of this 'bunching up' is that it leaves more empty space in the system into which a cavity can be inserted. In the context of the scaled particle theory, this effect appears as the reduced effective size of water molecules, which results in the lower volume packing density, which in turn reduces the free energy of cavity formation [22]. The possibility that the hydrogen bonds actually help the dissolution process has been suggested earlier [33,34] from an entirely different, thermodynamic perspective.

4.5. Summary

In summary, we found that it is possible to obtain a large solvophobic behavior from liquids which have the same van der Waals attraction as water but which has no hydrogen bonds. Whatever the mechanism is that produces the solvophobic behavior in these liquids, there appears to be no reason why the same mechanism would not operate in the real water. Since the magnitude of the free energy change produced in these liquids is similar to or greater than that produced in the real water model, there is little room for a contribution to the free energy by the 'structure formation', loss of orientational degree of freedom, or any other change due explicitly to the presence of hydrogen bonds in water. This result is consistent with the suggestion [20–22] that the hydrophobicity arises from the small size of water molecules.

Acknowledgement

We thank Mr. Joseph J. Cammisa for his dedicated help on keeping the computer systems up and running. We also thank Dr. Norma Allewell, Dr. Robert L. Baldwin, Dr. Jean Durup, Dr. John T. Edsall, Dr. William L. Jorgensen, Dr. Eaton E. Lattman, Dr. Norbert Muller, Dr. Andrew Pohorille, Dr. Lawrence R. Pratt, Dr. Alex Rashin, Dr. Frederic M. Richards, Dr. Peter J. Rossky, Dr. David Thirumalai, and Dr. Bruno H. Zimm for reading an earlier draft of the manuscript and for making comments that were invaluable in shaping the final style of the presentation.

References

[1] H.S. Frank and M.W. Evans, J. Chem. Phys. 13 (1945) 507.
[2] A. Geiger, A. Rahman and F.H. Stillinger, J. Chem. Phys. 70 (1979) 263.
[3] P.J. Rossky and M.J. Karplus, J. Am. Chem. Soc. 101 (1979) 1913.
[4] S. Okazaki, K. Nakanishi, H. Touhara, N. Watanabe and Y. Adachi, J. Chem. Phys. 74 (1981) 5863.
[5] M. Mezei and D.L. Beveridge, J. Chem. Phys. 74 (1981) 622.
[6] D.C. Rapaport and H.A. Scheraga, J. Phys. Chem. 86 (1982) 873.
[7] W.L. Jorgensen, J. Gao and C. Ravimohan, J. Phys. Chem. 89 (1985) 3470.
[8] D. van Belle and S.J. Wodak, J. Am. Chem. Soc. 115 (1993) 647.
[9] L.R. Pratt and D. Chandler, J. Chem. Phys. 67 (1977) 3638.
[10] A.A. Rashin and M.A. Bukatin, J. Phys. Chem. 95 (1991) 2941.
[11] T. Lazaridis and M.E. Paulaitis, J. Phys. Chem. 96 (1992) 3847.
[12] A. Ben-Naim, Biopolymers 14 (1975) 1337.
[13] R. Lumry, E. Battistel and C. Jolicoeur, Faraday Symp. Chem. Soc. 17 (1982) 93.
[14] B. Lee, J. Chem. Phys. 83 (1985) 2421.
[15] E. Grunwald, J. Am. Chem. Soc. 108 (1986) 5726.
[16] H.-A. Yu and M.J. Karplus, J. Chem. Phys. 89 (1988) 2366.
[17] R.A. Pierotti, J. Phys. Chem. 69 (1965) 281.
[18] A. Ben-Naim, Water and aqueous solutions (Plenum Press, New York, 1974).
[19] B. Lee, Biopolymers 31 (1991) 993.
[20] M. Lucas, J. Phys. Chem. 80 (1976) 359.
[21] B. Lee, in: Mathematics and computers in biomedical applications, eds. J. Eisenfeld and C. Delisi (North-Holland, Amsterdam, 1985) p. 3.
[22] B. Lee, Biopolymers 24 (1985) 813.
[23] A. Pohorille and L.R. Pratt, J. Am. Chem. Soc. 112 (1990) 5066.
[24] B. Guillot, Y. Guissani and S. Bratos, J. Chem. Phys. 95 (1991) 3643.
[25] H. Tanaka, J. Chem. Phys. 86 (1987) 1512.
[26] L.R. Pratt and A. Pohorille, Proc. Natl. Acad. Sci. US 89 (1992) 2995.
[27] W.L. Jorgensen, J. Chandrasekhar, J.D. Madura, R.W. Impey and M.L. Klein, J. Chem. Phys. 79 (1983) 926.
[28] H. Reiss, Advan. Chem. Phys. 9 (1966) 1.
[29] W.L. Jorgensen, BOSS, Version 3.3 (Yale University, New Haven, 1992).
[30] J.P. Postma, H.J.C. Berendsen and J.R. Haak, Faraday Symp. Chem. Soc. 17 (1982) 55.
[31] E. Wilhelm and R. Battino, J. Chem. Phys. 56 (1972) 563.
[32] J.J. Nicolas, K.E. Gubbins, W.B. Streett and D.J. Tildesley, Mol. Phys. 37 (1979) 1429.
[33] K. Shinoda and M. Fujihira, Bull. Chem. Soc. Japan. 41 (1968) 2612.
[34] A. Hvidt, Acta Chem. Scand. A 37 (1983) 99.

Discussion to the paper by B. Madan and B.K. Lee

Comments

By A. Rashin

The role of hydrogen bonds in the phenomenon of hydrophobicity that you adress is an old, important, and unresolved problem. The main point of your paper, as I understand it, is that hydrogen bonds contribute almost nothing to the free energy of transfer. Your simulations show that the free energy of cavity formation is the same in water and in a Lennard-Jones solvent. Because there is no orientational asymmetry for Lennard-Jones particles, you conclude that such asymmetry due to hydrogen bonds in water does not contribute to the free energy of transfer. There is quite a number of statements from work of others (including our own) which are in disagreement with your conclusion. Therefore I would like to formulate a few questions to clarify where the discrepancy may come from.

(1) Our calculations of hydration entropies (your ref. [10]) and those from Paulaitis' group (your ref. [11]) suggest that orientational entropy of water contributes significantly to the transfer entropy. Our calculations are, however, based on an intuitive model and may miss some factors (e.g., contributions from water–water orientational correlations in the bulk solvent). Paulaitis' calculations are also approximate (truncated at pair correlations) and may lead to their own artefacts. The results of these studies suggest that you may miss something but cannot be considered as a proof of it (others may disagree with my judgement here).

(2) A possible problem may concern your discussion of the role of high pressure. We argue in refs. [1,2] that free energies of transfer may differ significantly for transfer into weakly cohesive and strongly cohesive solvents. In PT transfer there is no compression entropy change but there is $P\Delta V$ work required to expand system after the transfer to maintain the constant pressure. This work is large for weakly cohesive solvents and negligible for strongly cohesive ones. In VT transfer there is no $P\Delta V$ work but there is an equal contribution due to the compression entropy. In weakly cohesive solvent there will be hardly any other contributions (in non-cohesive compressed ideal gas there are none). Thus your results and those from your ref. [26], where LJ solvent was made highly cohesive, may differ by $P\Delta V$ work or its compression entropy equivalent. This term is significant in your work and negligible in your ref. [26]. As ref. [26] finds the work of cavity creation in LJ liquid ("imposter 3") to be somewhat smaller ($\approx 20\%$) than in water, the $P\Delta V$ in your simulation will increase this work. 20% is not a negligible quantity, and may account for the orientational contribution. Thus, in your study of the system without orientational asymmetry the free energy may mimic that of water because of the compression contribution in your model system. Peter Rossky, as I recall, posed similar questions.

(3) The comment above is closely related to the discussion in this Issue of the controversial "volume terms" in the free energy of transfer. The controversy may stay with us for the time being, and so may the problem that you came up with. Your responses may help to resolve both.

[1] A.A. Rashin and M.A. Bukatin, J. Phys. Chem. 98 (1994) 386.
[2] A.A. Rashin and M.A. Bukatin, Biophys. Chem. 51 (1994) 167.

Responses by B.K. Lee to Comments.

To A. Rashin

(1) I think your work and that of Paulaitis's group on the direct calculation of entropy are very interesting. However, I do have some serious concerns with the methodology, some of which have been expressed in my comments to the paper by Paulaitis et al. in this Issue. Because of these concerns, I am not willing to accept all the results of these calculations at this time.

The results and the arguments presented in this paper are almost entirely at the level of the free energy change; no attempt has been made to delineate the possibly many enthalpic and entropic terms that contribute to the cavity formation process. It is possible, in fact highly likely, that the enthalpy and entropy changes upon cavity formation are different in water than in Lennard-Jones liquid. Since the total free energy changes are similar, the enthalpy and entropy changes that are different between these two liquids must be compensating. (See B. Lee, "Enthalpy-entropy compensation in the thermodynamics of hydrophobicity" in this Issue.) The significant orientational component of hydration entropy values that you and Paulaitis' group find, if confirmed, can be simply compensated by the correspondingly significant change in enthalpy due to a better (or worse, depending on the sign of the orientational entropy term) orientation.

(2) First of all, ref. [26] does not give numbers – we do not know if the free energy of cavity formation in their "imposter" liquid is 20% lower than in the water, although Fig. 4 of this reference does indicate that the free energy is lower for the "imposter" liquid than for the TIP4P water at large cavity sizes. We also do not know how sensitive this number is to the parameters of the Lennard-Jones liquid used nor how much leeway there is in choosing the particular values for this parameters. I hesitate to consider the free energies to be meaningfully different until these questions are cleared up. Certainly, the overall impression one gets from inspection of Fig. 4 is that the two are similar, one being higher than the other at one solute size and the other way around at a different size. Perhaps Dr. Pratt can comment on this.

It is true that the $p\Delta V$ term is significant for the Lennard-Jones liquid and negligible for water if the cavity formation is done under the constant pressure. This trivial difference between Lennard-Jones liquid and water is one reason why we do not consider the constant pressure process. The $p\Delta V$ term is of course zero for the constant volume process.

For the benefit of some readers, perhaps it is worth pointing out at the outset that the isothermal work of any process is equal to the change in the *Helmholtz* free energy whether the process is carried out under the constant volume or under the constant pressure. (See any standard thermodynamics textbook, e.g. ref. [1].) For the process of inserting a solute molecule (or a cavity) in a liquid, the work done under the constant pressure is given by $(\partial A/\partial n)_p$ and that under the constant volume by $(\partial A/\partial n)_V$. These two quantities are unequal and related by

$$\left(\frac{\partial A}{\partial n}\right)_p=\left(\frac{\partial A}{\partial n}\right)_V-p\left(\frac{\partial V}{\partial n}\right)_p, \qquad (1)$$

where the last term is equal to $p\Delta V$. This relation states that the work done under the constant pressure is equal to the non-$p\Delta V$ work, which is the same as the work done under the constant volume condition, minus the $p\Delta V$ work. The Gibbs free energy is defined so that change in the Gibbs free energy upon a constant pressure process is equal to the non-$p\Delta V$ work, which is, by definition, equal to the net total work done plus $p\Delta V$. When the $p\Delta V$ term is substantial, as in the case of our Lennard-Jones liquids, the work of cavity formation under constant pressure for the Lennard-Jones liquid is very different from that in water; the non-$p\Delta V$ work is the same but the total work includes the $p\Delta V$ term, which is large in case of the Lennard-Jones liquid and negligible in the case of water. The important points here are that physical chemists are usually interested in the non-$p\Delta V$ work and that this latter is equal to the constant volume work even when process is carried out under the constant pressure condition.

It seems that you would like to equate the $p\Delta V$ term of the constant pressure process with the "compression entropy" of the constant volume process. I suppose that the "compression entropy" is the entropy associated with the fact that the volume available to the solvent molecules is reduced when a solute molecule is inserted at a fixed point in the liquid. However, it is not easy to define this term quantitatively. The entropy of a collection of molecules is not determined by the volume of its system alone because entropy calculation involves intermolecular interaction, as you know very well. There is no simple way to relate the entropy change upon compression, except when the intermolecular interaction is simple as in the ideal gas system. Which interactions do you single out to include in the "compression entropy" term and which not? Whereas $p\Delta V$ has an unambiguous definition and is simple to calculate, the "compression entropy" term has an element of arbitrariness in its definition and, depending on how it is defined, is potentially difficult to calculate. I do not know how one can equate these two terms. The only possibility that comes to my mind is that one simply defines the "compression entropy" as the value of $-p\Delta V$ that would be observed if the process were carried out under the constant pressure. However, the "compression enfropy" defined this way will have no physical meaning for the constant volume process (except the imagined one derived solely from its name).

If we nonetheless follow this definition of the term "compression entropy", it will be negligible for TIP4P water (liquid #1) and for the "imposter" liquid of ref. [26] (liquid #2), whereas it will be substantial for our LJ liquid (liquid #3). You are then suggesting that the cavity free energy in liquid #1 is higher than that in liquid #2 because the former has the orientational entropy loss, whereas it is the same as that in liquid #3 because the latter has the "compression entropy" which would be comparable in magnitude to the orientational entropy.

My first argument against this suggestion is the following. As noted in point (1) above, the cavity free energy in liquid #2 is higher than that in liquid #1 for certain solute sizes. Straightforwardly interpreted in the context of this discussion, this means that the orientational entropy change in water is sometimes positive. This is certainly possible if the orientational degree of freedom of water molecules around a cavity is compared with that of water molecules around another, fixed water molecule. However, the "compression energy" change is always negative according to the definition given above. Therefore this suggestion does not explain the fact that the free energy changes in liquid #1 and liquid #3 are comparable at all solute sizes compared.

Secondly, as argued in the paper entitled "Enthalpy-entropy compensation..." in this Issue, most solvent reorganization processes are likely to be compensating. Thus, the onentational entropy change, if it is significant, can be expected to be largely compensated by a corresponding enthalpy change. The difference that remains after the compensation is not likely to be of the magnitude comparable to the sizable "compression entropy" term in liquid #3.

What we argue is that the net free energy change arises from the size effect, which is common to all three liquids. The entropy changes are different in these liquids, but they will be all compensated by the corresponding enthalpy changes, except that due to the size effect, which does not compensate. As stated above, I believe that the notion of "compression entropy" is hazy for real liquids. However, depending on how the "compression entropy" is defined, this term may contain the size effect of above description, in which case, the two notions are the same, despite the different language used. In this case, however, the notion of "compression entropy" should be defined to have at least the following two characteristics: (1) It should be substantial for all three liquids, including water, since there is compression even for water if the solute is inserted under constant volume. (2) It should depend on the intermolecular potential of the solvent molecules, as well as the volume of compression. In particular, it should depend on the size of the solvent molecules, since the entropy loss upon compression of a (compressed) ideal gas is different from that of a hard sphere liquid. Defined to have these characteristic, the "compression entropy" may in fact capture the notion of the size effect in this paper.

[1] G.N. Lewis, M. Randall, K.S. Pitzer and L. Brewer, Thermodynamics, 2nd Ed. (McGraw-Hill, New York, 1961).