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What Some Anions do to H_2O? : Towards Understanding the Hofmeister Series[1)]

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The effects of selected anions on the molecular organization of H_2O are elucidated by the 1-propanol (1P) probing methodology developed by us. We determine the third derivative quantity, the 1P-1P enthalpic interaction, $H^E_{1P\text{-}1P}$, in ternary system of 1P - sample salt (S) - H_2O. In the presence of S, the $H^E_{1P\text{-}1P}$ pattern changes while keeping the basic peak anomaly. These induced changes are used to elucidate the effect of S on H_2O. We found within the sodium salts of SO_4^{2-}, F^-, Cl^-, Br^-, I^- and ClO_4^- that the first three act basically as hydration centers. The hydration numbers are 17, 14 and 2.3, respectively. The remaining Br^-, I^- and ClO_4^- are basically hydrophiles. They form hydrogen bonds directly to the existing hydrogen bond network of H_2O and thus work as impurity centers in the existing network, retarding the degree of fluctuation inherent in H_2O. Their effects are stronger in the order of $Br^- < I^- < ClO_4^-$. Thus in terms of the Hofmeister effects, the kosmotropes (salting out agents) are hydration centers and the more the hydration number, the stronger the salting out effect. The chaotropes (salting in reagents), Br^-, I^- and ClO_4^-, are hydrophiles, and the stronger the hydrophilicity the stronger the salting-in power.

1. Introduction - Hofmeister series

In 1985, Collins and Washabaugh[2)] reviewed a large number of papers regarding the Hofmeister effect[3)] and ranked some anions in the order,

$$SO_4^{2-} > F^- > Cl^- > Br^- > I^- \approx ClO_4^- > SCN^- \qquad (1)$$

Originally, this shows the ranking of the effect of an ion to induce precipitation of a protein from its aqueous solution. Namely, the left end of eq.(1) is a more salting out agent, while the right end a more salting in reagent. The former salting-out ion requires a smaller amount to precipitate the protein. This means that the excess chemical potential of the protein becomes more positive when a unit amount of a salting-out agent is added to the aqueous solution of the test protein, while with the same amount of a salting-in agent the excess chemical potential of protein will be less positive. Another point to stress is that this ranking is almost ubiquitous; a large number of examples of functions and structures of biopolymers follow the almost same ranking in their ion effects. In order to cover the generality of the ion effects, Collins and Washabaugh thus coined the words, "Kosmotrope/Chaotrope" for the ions in place of the salting out/in naming of the above ranking.[2)]

We earlier developed a novel differential approach in solution thermodynamics, and applied it to aqueous solutions.[4-7)] We determine the excess partial molar quantities of the i-th component, F_i^E, ($F = G$, H, S, or V), by perturbing the system with the amount of the i-th component only in the mixture, and by measuring the reaction of the entire system in terms of the excess thermodynamic quantity, F^E. Thus,

$$F_i^E = \left(\frac{\partial F^E}{\partial n_i}\right)_{n_{j \neq i}, p, T} \qquad (2)$$

This signifies the actual thermodynamic situation of the i-th component in the mixture. We note that for

$F=G$, F_i^{E} is written as μ_i^{E} instead from the historical reason, and is the first derivative of G. For $F=H$, S, or V, the resulting F_i^{E} is the second derivative of G. We then take one more derivative with respect to the amount of the j-th component as,

$$F_{i-j}^{\mathrm{E}}=N\left(\frac{\partial F_i^{\mathrm{E}}}{\partial n_i}\right)_{n_{k\neq j},p,T} \tag{3}$$

where N is the total amount of the system, i.e. $N=\sum_i n_i$. The quantity defined by eq. (3) gives the effect of the j-th component on the excess partial molar quantity of the i-th component, or the effect of j on the actual thermodynamic situation of i. Hence it signifies the i-j interaction in terms of the thermodynamic quantity, F. Thus, the order of derivative for F_{i-j}^{E} increases by one step.

Using these terminologies, what the ranking, eq. (1), tells us for the original protein precipitation case is that the interaction between the protein (i) and the ion (j) in terms of chemical potential, μ_{i-j}^{E} , (where F in eq.(3) is Gibbs function, G), is more positive for a kosmotropic ion than that for a chaotrope. Furthermore, if the ranking is indeed ubiquitous, and is also applicable to 1-propanol (abbreviated as 1P) in a ternary system 1P - a sample salt (S) - H_2O, then the excess chemical potential of 1-propanol, $\mu_{\mathrm{1P}}^{\mathrm{E}}$, must become more positive in the unit addition of a kosmotropic S than that for a chaotrope in 1P-S-H_2O system. Namely, the 1P-S interaction in terms of the excess chemical potential, $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$, should be more positive for a kosmotrope. Indeed, we determined approximate values of $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ at 25℃ for S$=Na_2SO_4$, NaF, NaCl, NaI, and $NaClO_4$ at the infinite dilution, $x_{\mathrm{1P}}\rightarrow 0$, and $x_{\mathrm{S}}^0\rightarrow 0$.[8] x_{1P} and x_{S}^0 are the mole fraction of 1P and the initial mole fraction of S in 1P-S-H_2O. The results are shown in **Fig.1**. Since the counter cation is fixed with Na^+ in the series of salts studied, the effect of S could be considered as that of anion. Thus, on the abscissa S is placed in the order of the Hofmeister ranking, eq.(1), and the values of $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ at the infinite dilution are plotted on the ordinate. As is evident from the figure, $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ decreases exactly in the order of eq.(1), including the approximate equality $I^- \approx ClO_4^-$. This assures that the Hofmeister ranking is operative for 1P also. We earlier determined the excess partial molar enthalpy of 1P, $H_{\mathrm{1P}}^{\mathrm{E}}$ ($F=H$ in eq.(2)), in ternary 1P-S-H_2O at 25℃ varying x_{1P} and x_{S}^0.[9,10] Thus, the values of $H_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ ($F=H$ in eq.(3)) are available at the infinite dilution for each S. It follows then that the values of $TS_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ are readily calculated at the infinite dilution. The results are also plotted in **Fig.1**. It is striking that the net $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ ($=H_{\mathrm{1P\text{-}S}}^{\mathrm{E}}-TS_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$) decreases smoothly following the order of the Hofmeister ranking, but each term $H_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ and $TS_{\mathrm{1P\text{-}S}}^{\mathrm{E}}$ show a sharp break in the middle at about Cl^-. While this is another example of the enthalpy-entropy compensation prevalent in aqueous solutions,[11,12] the important point to note is that the mechanisms operating for the net Hofmeister effect are different between kosmotropes and chaotropes and the null point is at about Cl^-. How different are they? To this end, we study the mechanism how each ion modifies the molecular organization of H_2O by using the 1P-probing methodology developed by us.[1,6]

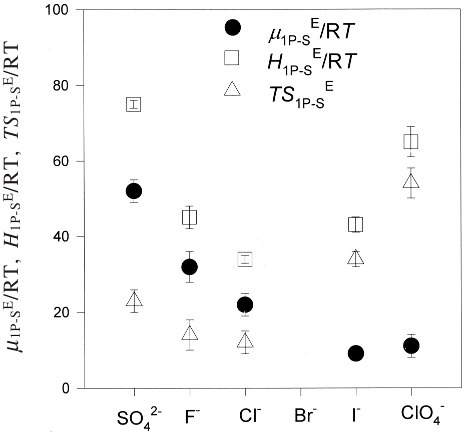

Fig.1 1-Propanol (1P)-sample salt (S) interactions in 1P-S-H_2O at 25℃. The filled circles show the 1P-S interaction in chemical potential, $\mu_{\mathrm{1P\text{-}S}}^{\mathrm{E}}/RT$, the hollow squares $H_{\mathrm{1P\text{-}S}}^{\mathrm{E}}/RT$, and the hollow triangles $TS_{\mathrm{1P\text{-}S}}^{\mathrm{E}}/RT$. The ions of sodium salt are displayed on the abscissa in the Hofmeister order from the kosmotrope to the chaotrope.

2. 1-Propanol (1P) probing methodology

Using the higher order derivative quantities, eq.(2) and eq.(3), we studied the series of mono-ols, varying the size of alkyl group and keeping - OH fixed in order to exemplify the nature of the hydrophobic moiety in aqueous solutions.[4-7] We learned that in the H_2O - rich region, a hydrophobic solute enhances the hydrogen bond network of H_2O in its immediate vicinity (corresponding to the classical "iceberg formation") with the concomitant

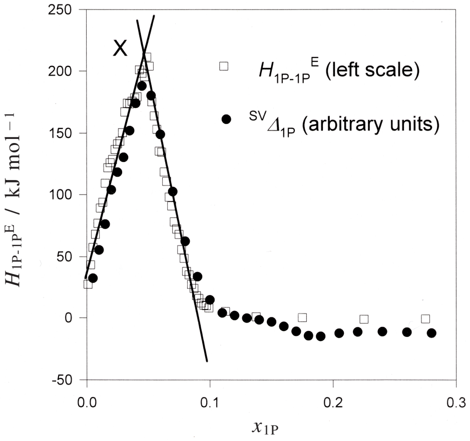

Fig.2 The 1P-1P interaction function, $H^{E}_{1P\text{-}1P}$, in binary 1P-H_2O and the partial molar entropy-volume cross normalized fluctuation of 1P, $^{SV}\Delta_{1P}$, as a function of x_{1P}. The ordinate scale is for $H^{E}_{1P\text{-}1P}$, and in arbitrary units for $^{SV}\Delta_{1P}$.

reduction of the hydrogen bond probability of H_2O in the bulk away from the solute molecules. We call this mode of mixing Mixing SchemeI. The solute(i)-solute(i) enthalpic interaction, H^{E}_{i-i}, shows a sharp peak anomaly as shown in **Fig.2**. Point X in the figure corresponds to the onset of transition from Mixing Scheme I to II. The latter mixing scheme is pertinent to the intermediate composition. Also shown in **Fig.2** is "the partial molar normalized *S*-*V* cross fluctuation of 1P", $^{SV}\Delta_{1P}$, devised by us earlier.[13-15] Briefly, the normalized *S*-*V* cross fluctuation, $^{SV}\Delta$, indicates qualitatively the intensity as well as the extensity of the entropy-volume cross fluctuation. If fluctuation is regarded as wave, the intensity is the amplitude and the extensity corresponds to the wavelength. Unlike normal liquid, H_2O is unique in that $^{SV}\Delta$ contains a negative contribution due to putative formation/destruction of ice-like patches in the bulk H_2O. This negative part therefore is the signature of liquid H_2O. Its partial molar quantity of 1P, $^{SV}\Delta_{1P}$, shows the effect of 1P on the negative contribution, the H_2O-likeness, of the system. What is striking in **Fig.2** is that the x_{1P}-dependence of $H^{E}_{1P\text{-}1P}$ and that of $^{SV}\Delta_{1P}$ matches exactly on scaling the ordinate by a single factor! This indicates that $H^{E}_{1P\text{-}1P}$ and $^{SV}\Delta_{1P}$ share the same cause. We suggest that the enthalpic 1P-1P interaction is operative via the bulk H_2O away from the iceberg-clad solutes, where the H_2O-likeness is gradually changing due to the presence of 1P. The process of this modification manifests itself in the increase in $^{SV}\Delta_{1P}$ and $H^{E}_{1P\text{-}1P}$ from the start, $x_{1P}=0$, to point X shown in the figure. The initial increase in $^{SV}\Delta_{1P}$ is caused by the decrease in the negative contribution in the bulk H_2O away from "icebergs". In addition, we suggested that "icebergs" themselves do not participate in fluctuation, i.e. they are rigid units in the time scale of fluctuation, by the x_{1P}-dependences of other fluctuation functions; the mean square entropy or volume fluctuations.[13-16]

In ternary 1P-S-H_2O systems, then the changes induced in the $H^{E}_{1P\text{-}1P}$ ($^{SV}\Delta_{1P}$) pattern in the presence of S will be the reflection of the state of the bulk H_2O modified by S. As long as the basic $H^{E}_{1P\text{-}1P}$ pattern displays a peak anomaly, the integrity of H_2O remains intact, i. e. the system is still within Mixing Scheme I. This led us to an idea of the 1-propanol (1P) probing methodology of studying the effect of a sample solute (S) on the molecular organization of H_2O in the ternary 1P-S-H_2O system. We determine the excess partial molar enthalpy of 1P in 1P-S-H_2O, H^{E}_{1P}, by perturbing the amount of 1P only. We then graphically differentiate H^{E}_{1P} data to obtain $H^{E}_{1P\text{-}1P}$, without resorting to any model system. We then examine the induced changes in the x_{1P}-dependence of the $H^{E}_{1P\text{-}1P}$ pattern. The induced changes are used in turn to learn about the effect of S on H_2O.

3. The effects of hydrophobes, hydrphiles and amphiphiles on H_2O by the $H^{E}_{1P\text{-}1P}$ pattern changes

We first summarize the results of the 1P-probing methodology for S = non-electrolytes in 1P-S-H_2O.[17] For a hydrophobic solute, its effect on the $H^{E}_{1P\text{-}1P}$ pattern could be sketched in **Fig.3**. The original pattern for the binary 1P-H_2O [O] in the figure shifts parallel to the left, in the presence of an equally hydrophobic S (case [A] in the figure). Thus, the value of $H^{E}_{1P\text{-}1P}$ at point X remains the same, its x_{1P} locus decreases, and the value of $H^{E}_{1P\text{-}1P}$ at the start, $x_{1P}=0$, increases steadily as x^{0}_{S} increases. For a weaker (stronger) hydrophobe, in addition to the shift to the left, the value of $H^{E}_{1P\text{-}1P}$ decreases (increases) in the entire range of x_{1P} within Mixing Scheme I. As a result, the value of $H^{E}_{1P\text{-}1P}$ at point X decreases (increases), the rate of increase of $H^{E}_{1P\text{-}1P}$ at $x_{1P}=0$ decelerates (accelerates). The mechanism for these effects on the $H^{E}_{1P\text{-}1P}$ pattern comes from the way a

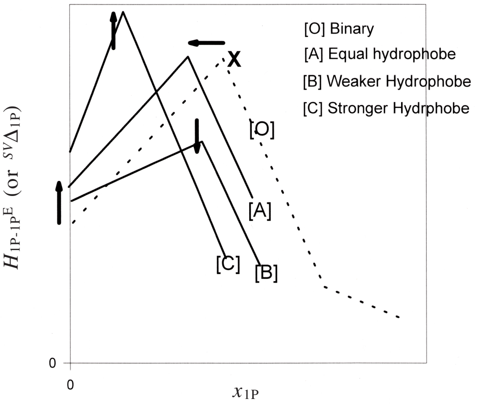

Fig.3 The induced changes in the $H^{E}_{1P\text{-}1P}$ pattern by the presence of a hydrophobic S, in 1P-S-H_2O. [O] is the $H^{E}_{1P\text{-}1P}$ pattern for the binary 1P-H_2O; [A] shows that with an equally hydrophobic S as the probing 1P; [B] is that with a weaker (methanol), and [C] that with a stronger hydrophobe (tert-butanol).

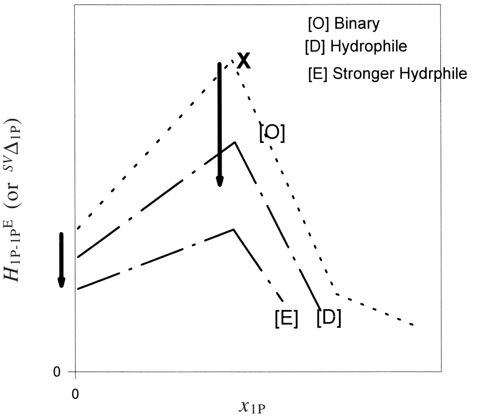

Fig.4 The induced changes in the $H^{E}_{1P\text{-}1P}$ pattern by the presence of a hydrophilic S, urea for example, in 1P-S-H_2O.

hydrophobic S affects the molecular organization of H_2O within Mixing Scheme I. Namely, S enhances the hydrogen bond network of H_2O in its immediate vicinity ("iceberg" formation) and concomitantly reduces the hydrogen bond probability of bulk H_2O away from S. This in turn reduces the chance of putative formation of ice-like patches. As a result, the negative contribution decreases in the *S-V* cross fluctuation. The effect of 1P on the rate of this decrease increases as x_{1P} increases, resulting in the increase in $^{SV}\Delta_{1P}$ and hence in $H^{E}_{1P\text{-}1P}$.

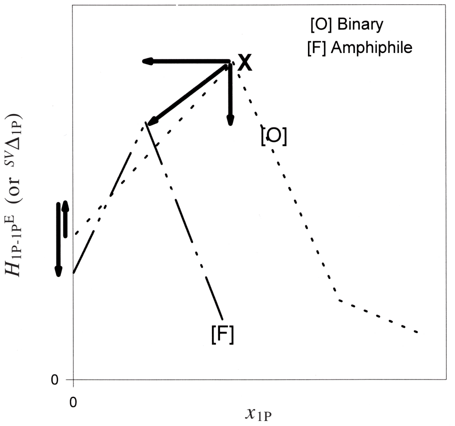

Fig.5 The induced changes in the $H^{E}_{1P\text{-}1P}$ pattern by the presence of an amphiphile S, 1,2-propanediol for example, in 1P-S-H_2O. The hydrophobic part shifts point X to the left and raise the value of $H^{E}_{1P\text{-}1P}$ at the start, $x_{1P} = 0$. The hydrophilic part, on the other hand, lower the value of $H^{E}_{1P\text{-}1P}$ in the entire range including at point X. The net result is a combination of these two effects.

On the other hand, the probing 1P-1P interaction occurs via the bulk H_2O away from S, which is already modified by S and is influenced by the mixed ways how S and 1P modify the bulk H_2O. Hence, if S is a weaker(stronger) hydrophobe than 1P, the value of $H^{E}_{1P\text{-}1P}$ is suppressed (enhanced).

Fig.4 shows the sketch of the effect of a hydrophile on the $H^{E}_{1P\text{-}1P}$ pattern. The x_{1P} locus remains unchanged but the value of $H^{E}_{1P\text{-}1P}$ decreases by the presence of a hydrophile. A hydrophile forms hydrogen bonds to the existing hydrogen bond network of H_2O, and thus the connectivity is retained, but the H donor/acceptor symmetry of H_2O is broken, not only in the geometry but also in the hydrogen bond strength. As a result, the hydrogen bond network becomes more rigid and loses the degree of fluctuation inherent in liquid H_2O. Thus, the regular positive part of *S-V* cross fluctuation decreases in the presence of a hydrophilic S. Urea is a typical example for this behaviour.[18)] A simulation study on urea-H_2O concluded that the presence of urea rigidifies the dynamics of both H_2O and urea.[19)]

For an amphiphile, its effect on the $H^{E}_{1P\text{-}1P}$ pattern is a combination of the above two cases, depicted in **Fig.3** and **Fig.4**. **Fig.5** sketches the resulting change in

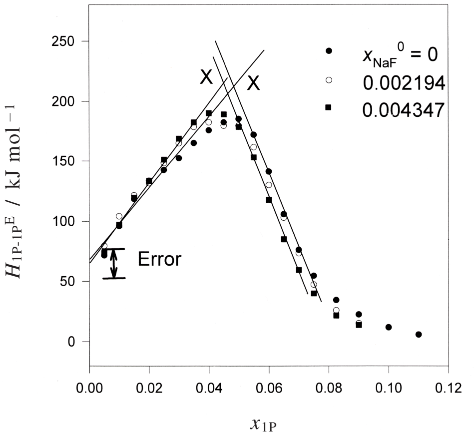

Fig.6 The enthalpic 1-propanol (1P)-1P interaction, $H^{E}_{1P\text{-}1P}$, in 1P-NaF-H_2O at 25℃.

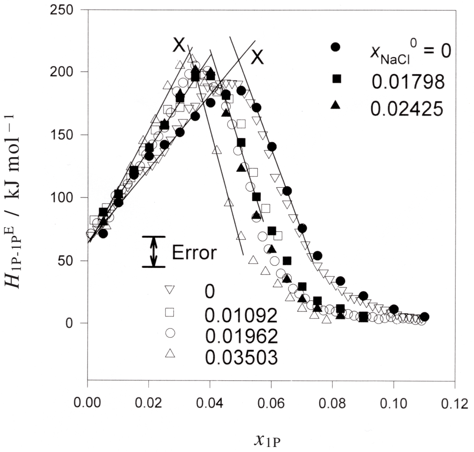

Fig.7 The enthalpic 1-propanol (1P)-1P interaction, $H^{E}_{1P\text{-}1P}$, in 1P-NaCl-H_2O at 25℃.

$H^{E}_{1P\text{-}1P}$ by an amphiphile. A typical example is 1,2-propanediol.[20)]

4. The effects of NaF and NaCl on H_2O as probed by the $H^{E}_{1P\text{-}1P}$ pattern change

We now apply the same methodology for Na-salts of selected anions. **Fig.6** and **Fig.7** show the $H^{E}_{1P\text{-}1P}$ pattern in 1P-S-H_2O at 25℃, for S = NaF and NaCl respectively.[9)] As is evident in the figures, for x_S^0 (S = NaF) < 0.0043 and x_S^0 (S = NaCl) < 0.035, the x_{1P}-dependence of $H^{E}_{1P\text{-}1P}$ retains the peak anomaly. Thus, within these concentration ranges, the aqueous solutions are in the Mixing Scheme I region. In **Fig.7**, our earlier data are also included.[21)]

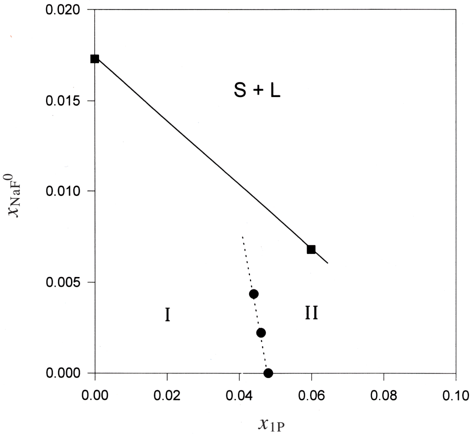

Fig.8 The mixing scheme and phase diagram of 1-propanol (1P)-NaF-H_2O at 25℃. The solid line is the phase boundary, and the dotted line is the mixing scheme boundary, indicating the onset of the transition from Mixing Scheme I to II.

What is striking in **Fig.6** and **Fig.7** is that on addition of S (S = NaF or NaCl), the starting point at $x_{1P} = 0$ remains fixed while the locus of point X shifts to the left, to smaller values of x_{1P}, without changing the height of the peak, the value of $H^{E}_{1P\text{-}1P}$, and hence that of ${}^{SV}\Delta_{1P}$ at point X. This immediately suggests that the availability of H_2O for 1P to interact is reduced as x_S^0 increases. Hence, it requires a lesser x_{1P} to drive the system to point X. We suggest that this is due to hydration of the salt. From the observation that the values of $H^{E}_{1P\text{-}1P}$ remains the same at the start as well as at point X, we conclude that the molecular organization of bulk H_2O away from the hydration shell remains the same as that in pure H_2O.

Fig.8 and **Fig.9** show the loci of point X, which is the onset of the transition from Mixing Scheme I to II, together with the phase boundary of 1P-S-H_2O (S = NaF and NaCl).[9)] The figures indicate that the loci of point X seem to form a straight line. Namely, the shift of point X to the left is linear to x_S^0. If this left shift is due to hydration, as we suggest, its slope provides an estimate of the hydration number. Alternatively, the x_S^0 intercept of the straight-line Mixing Scheme boundary gives the mole fraction of S such that all H_2O molecules would be used up for hydration. Thus, the x_S^0 intercept is equal to $1/(1 + n_H)$, where n_H is the hydration number within the context of the present 1P-probing methodology.

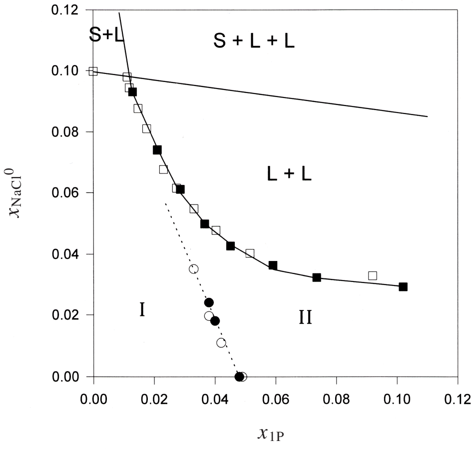

Fig.9 The mixing scheme and phase diagram of 1-propanol(1P)-NaCl-H_2O at 25 ℃. The solid lines are the phase boundaries and the dotted line is the mixing scheme boundary indicating the onset of the transition from Mixing Scheme I to II.

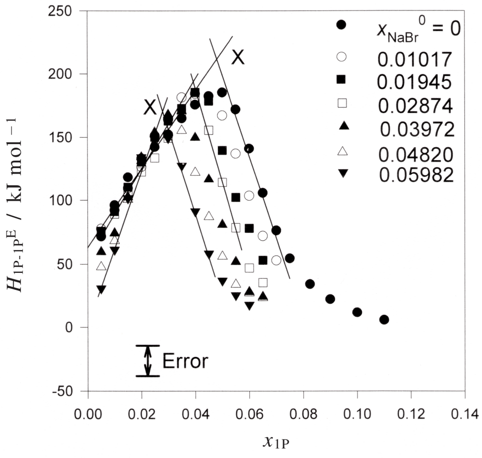

Fig.10 The enthalpic interaction, $H^E_{1P\text{-}1P}$, in 1-propanol (1P)-NaBr-H_2O at 25 ℃.

Thus, the effects of NaF and NaCl on H_2O are that they hydrate 19 ± 1.5 and 7.5 ± 0.6 of H_2O respectively, and that their presence does not affect the properties of the bulk H_2O away from the hydration shells.

5. The effects of NaBr and NaI to H_2O as probed by the $H^E_{1P\text{-}1P}$ pattern change.

The $H^E_{1P\text{-}1P}$ plots are shown in **Fig.10** and **Fig.11** for 1P-S-H_2O at 25 ℃ with S = NaBr and NaI.9) While point X shifts to the left as x^0_S (S = NaBr or NaI) increases, the values of $H^E_{1P\text{-}1P}$ at point X and at the start, $x_{1P} = 0$ decrease. These induced changes in the $H^E_{1P\text{-}1P}$ pattern, particularly for S = NaBr, **Fig.9**, resembles the effect of an amphiphile shown in **Fig.5**. Thus, the effects of NaBr and NaI on H_2O have the hydrophilic propensity. For the present cases of S = NaBr and NaI, are the shifts of point X to the left due to the hydration to Na^+? **Fig.12** and **Fig.13** show the plots of the x_{1P}-loci of point X vs. x^0_S, representing the shift to the left by whatever a mechanism. They appear to form straight lines, and their intercepts provide $n_H = 5.2 \pm 0.6$ for NaBr, and 5.6 ± 0.6 for NaI. This suggests that Na^+ hydrates about 5 H_2O, and therefore Br^- and I^- work as hydrophiles, as in **Fig.4**. White et al. used the first-principles molecular dynamics simulations that avoid empirically determined parameters or a restriction to pair-wise interaction potentials and studied the Na^+-H_2O system.22) The results indicated that a Na^+ hydrates 5.2 molecules of H_2O and leaves the orientation dynamics of the bulk H_2O away from the hydration unperturbed.22) More recently, Varma & Rempe used also an *ab initio* molecular dynamics simulations and suggested that the hydration number of Na^+ is 5.23) Experimentally, another recent study by the 'difference method' of neutron diffraction with isotopic substitutions and the anomalous x-ray diffraction concluded that the hydration number of Na^+ is also 5.24) Thus it appears to be safe to use the hydration number of Na^+ to be 5.2 form our NaBr result, and evaluate that for counter anion accordingly.

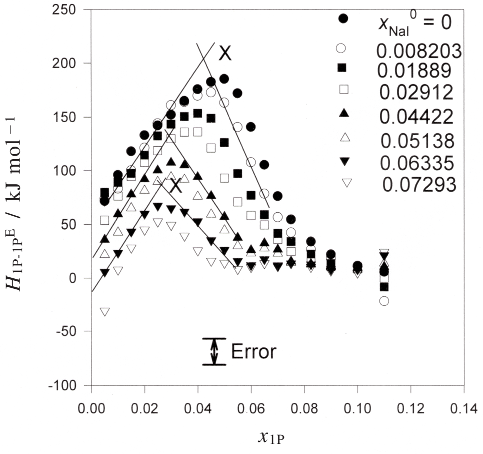

Fig.11 The enthalpic interaction, $H^E_{1P\text{-}1P}$, in 1-propanol (1P)-NaI-H_2O at 25 ℃.

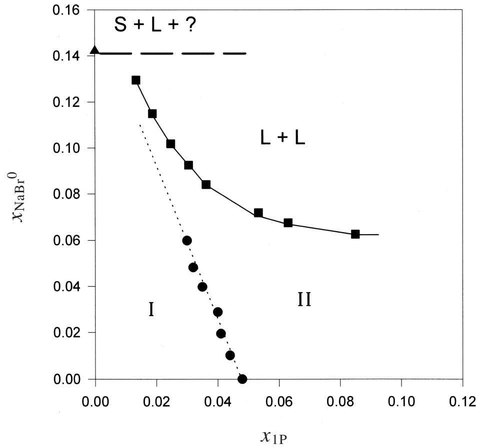

Fig.12 The mixing scheme and phase diagram for 1-propanol(1P)-NaBr-H_2O at 25℃. The solid line shows the phase boundary, the broken line a presumed phase boundary, and the dotted line shows the onset of transition from Mixing Scheme I to II.

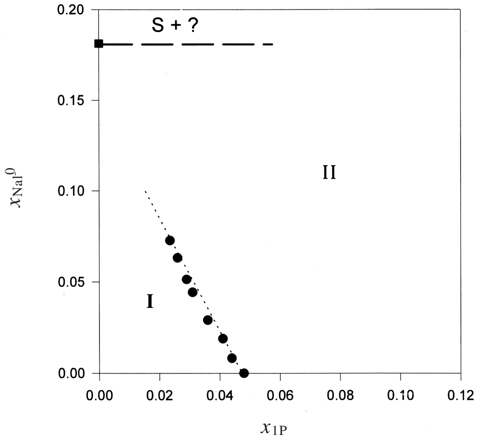

Fig.13 The mixing and phase diagram for 1-propanol(1P)-NaI-H_2O at 25℃. The broken line shows a presumed phase boundary and the dotted line the onset of the transition from Mixing Scheme I to II.

Thus, the hydration number, n_H, for F^- is 14±2 and 2.3±0.6 for Cl^-. These are listed in Table.

It is known[25-27] that halide ions are capable of forming hydrogen bonds with H_2O as proton acceptors, and their strength decease in the order of

$$F^- > Cl^- > Br^- > I^- \tag{4}$$

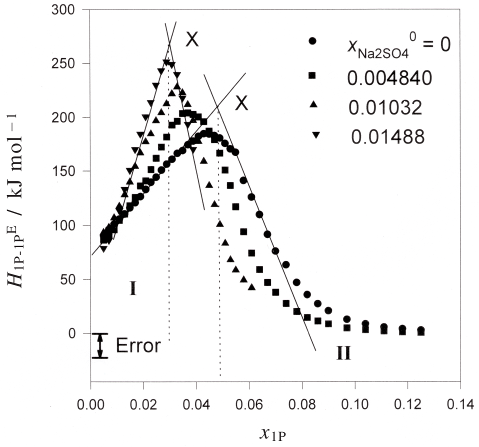

Fig.14 The 1-propanol(1P)-1P enthalpic interaction, $H_{1P\text{-}1P}^E$, in 1P-Na_2SO_4-H_2O at 25℃.

We suggest that F^- and Cl^-, being stronger proton acceptors, strip a number of H_2O molecules from the existing hydrogen bond network of H_2O and form the hydration shells, which do not extend their influences over to the bulk H_2O. The latter effect (non-effect) contrasts with what hydrophobes do to H_2O. As discussed above, hydrophobes form "icebergs" and influence concomitantly the bulk H_2O away from "icebergs" in that the hydrogen bond probability of the bulk H_2O is reduced progressively to the transition point to Mixing Scheme II. Clearly, F^- is a stronger hydration center than Cl^-, and n_H is 14±2 for F^- and 2.3±0.6 for Cl^-. Thus, in terms of the original Hofmeister's "H_2O withdrawing power",[3] the Hofmeister ranking $F^- > Cl^-$ is consistent.

Br^- and I^-, on the other hand, are weaker proton acceptors than F^- and Cl^-. As a result, they form hydrogen bonds to the existing network of H_2O, rather than stripping any H_2O molecules out of the network. As a result, the H_2O network now contains impurity centers that break the H donor/acceptor symmetry inherent in H_2O. Hence the hydrogen bond network of H_2O becomes more rigid and $H_{1P\text{-}1P}^E$ and ${}^{SV}\Delta_{1P}$ decreases. This effect is stronger for I^- than Br^-, in comparing their effects on the values of $H_{1P\text{-}1P}^E$ at point X. This comparison is shown below.

6. The effects of Na_2SO_4, and $NaClO_4$ to H_2O as probed by the $H_{1P\text{-}1P}^E$ pattern change

We examine the effects of the sodium salts of SO_4^{2-} and ClO_4^- by the same 1P-probing methodology. **Fig.14**

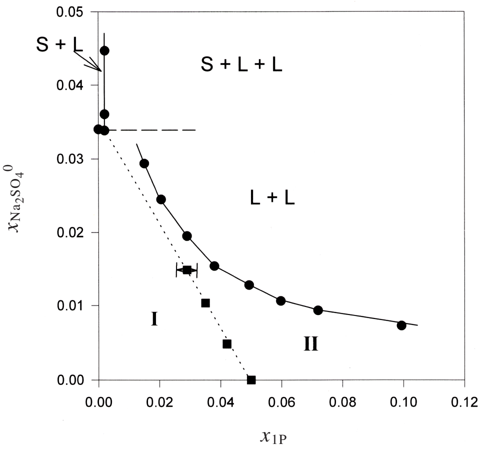

Fig.15 The mixing scheme and phase diagram for 1-propanol(1P -Na_2SO_4-H_2O at 25℃. The solid line is the phase boundary. The broken line is a presumed phase boundary. The dotted line is the mixing scheme boundary, the onset of the transition from Mixing Scheme I to II.

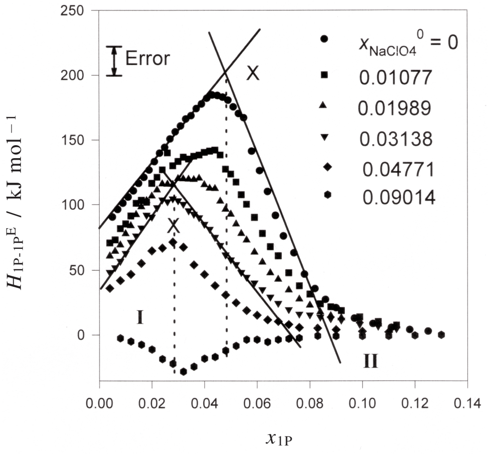

Fig.16 The 1-propanol (1P) - 1P enthalpic interaction, H^E_{1P-1P}, in 1P - $NaClO_4$ - H_2O at 25℃.

shows the H^E_{1P-1P} pattern in 1P-S-H_2O at 25℃ with S = Na_2SO_4. As is evident in the figure, Na_2SO_4 shows a hallmark of a hydration center in two respects. The H^E_{1P-1P} value at the start, $x_{1P} = 0$, remains constant and the x_{1P}-locus of point X shifts to the left, to smaller values of x_{1P}, as x_S^0 (S = Na_2SO_4) increases. Unlike NaF or NaCl, however, the value of H^E_{1P-1P} at point X increases. If the effects of cations and anions are additive, as we are taking as a premise, then that of Na^+ would not

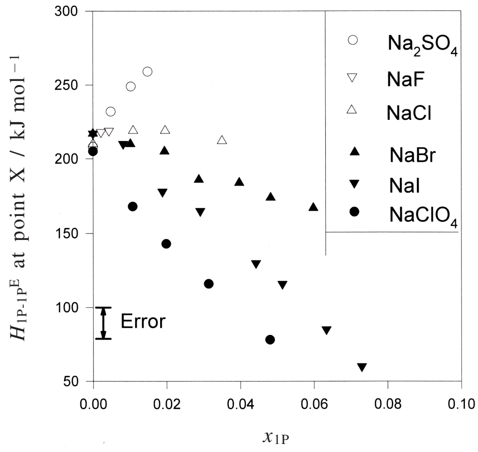

Fig.17 The values of the 1P - 1P interaction, H^E_{1P-1P}, at point X on addition of S in 1P-S-H_2O at 25℃. S = Na_2SO_4, NaF, NaCl, NaBr, NaI and $NaClO_4$. Na^+ is purely a hydration centre, with the hydration number, $n_H = 5.2$. The anions with hollow symbols are hydration centres. Others with filled symbols are hydrophiles. A stronger hydrophile lowers the H^E_{1P-1P} value more.

increase the value of H^E_{1P-1P} at point X. Thus, the observed increase in H^E_{1P-1P} at point X is due to $SO_4{}^{2-}$. We recall that a stronger hydrophobe than the probing 1P, makes the value of H^E_{1P-1P} increase both at the start and at point X, **Fig.3**. For the present case, the value of H^E_{1P-1P} at the start remains the same. Thus, the detail of the effect of $SO_4{}^{2-}$ is not clear at present. It could be that $SO_4{}^{2-}$ works as a hydration center in the absence of 1P in the system, resulting in the unchanged value of H^E_{1P-1P} at $x_{1P} = 0$. As x_{1P} increases, however, a direct interaction between $SO_4{}^{2-}$ and 1P might start to operate, in addition to the H_2O-mediated 1P-$SO_4{}^{2-}$ interaction. Nonetheless, from the fact that the H^E_{1P-1P} values at $x_{1P} = 0$ remains constant, we may classify $SO_4{}^{2-}$ as a hydration center. Thus, from the mixing scheme boundary, **Fig.15**, the hydration number, n_H, is estimated as 27 ± 3 for Na_2SO_4. It follows using the reference $n_H = 5.2$ for Na^+, n_H for $SO_4{}^{2-}$ is 17 ± 3.

Fig.16 shows that the effect of $NaClO_4$ on the H^E_{1P-1P} pattern in 1P-S-H_2O (S = $NaClO_4$) at 25℃. Clearly, its effect is similar to that of an amphiphile, as in **Fig.5**. Namely, there is a strong showing of hydrophilicity, i.e. the value of H^E_{1P-1P} decreases as x_S^0 increases. There is, however, some weak effect of the left shift. At least a part of the left shift could be due to Na^+. As discussed at some length, there seems to be a weak contribution of hydrophobicity apparent in ClO_4^-, although this requires a further clarification.[1,10]

Fig.17 is the summary of the effects of salts on the value of $H^{\mathrm{E}}_{\mathrm{1P\text{-}1P}}$ at point X including halide salts. Those showing the signature for a hydration center (including Na_2SO_4) are indicated by hollow symbols in the figure. The hydrophiles are shown by filled symbols. Within these hydrophiles, the relative effect of the anion is represented by the slope in the figure, since the counter cation is fixed at Na^+. Accordingly the slope for ClO_4^- is more negative than that of I^-, which in turn more negative than that of Br^-. This ranking is consistent with the chaotropic half of the Hofmeister series, eq.(1).

7. Conclusion - Hofmeister Series

Table summarizes our findings from the 1P-probing methodology what each anion does to H_2O. From Table, the following conclusions could be drawn:-

(1) A kosmotropic ion within those listed here is a hydration center. The more kosmotropic the ion, the larger the hydration number. This is in the complete accord with the classical idea by Hofmeister, the ranking of "the H_2O withdrawing power."[3]

(2) The ions regarded as chaotropes are hydrophiles, since they reduce the value of $H^{\mathrm{E}}_{\mathrm{1P\text{-}1P}}$ within the 1P-probing methodology. This tendency of reduction is stronger for a more chaotropic ion. Thus, while the Hofmeister ranking seems continuous, the mechanism is clearly different between kosmotropes and chaotropes. We note that in terms of $\mu^{\mathrm{E}}_{\mathrm{1P\text{-}S}}$ the effects of I^- and ClO_4^- are almost the same. In terms of $H^{\mathrm{E}}_{\mathrm{1P\text{-}S}}$, however, they are clearly different. Thus, the third derivative quantity, $H^{\mathrm{E}}_{\mathrm{1P\text{-}S}}$, contains more detailed information than the second derivative, $\mu^{\mathrm{E}}_{\mathrm{1P\text{-}S}}$. This is consistent of what we advocate.[3-10] In this context, the present 1P-probing methodology uses the effect of x_{S}^0 on the $H^{\mathrm{E}}_{\mathrm{1P\text{-}1P}}$ pattern. Hence, this methodology is in effect using the fourth derivative of G. This is partly a reason for success in gaining deeper insights.

(3) The boundary between kosmotropes and chaotropes may be between Cl^- and Br^-.

We stress that results summarized above in Table are the effects of each ion on the molecular organization of H_2O probed by the x_{1P} dependence pattern of $H^{\mathrm{E}}_{\mathrm{1P\text{-}1P}}$ within Mixing Scheme I. While there is no doubt that the properties of the mixed solvent, the aqueous salt solution, play a major role in dictating the structure and the function of biopolymers, there must be a great many more issues to be sorted out before linking the above results to the actual Hofmeister effect for each testing system. Namely, there is another equally important question remaining as to how a given biopolymer behaves in a given aqueous salt solution.

There is yet another issue to be considered regarding the specificity of a biopolymer. Biopolymers are giant molecules whose surfaces are covered with hydrophobic and hydrophilic moieties in varying ratios. We used as a probe 1-propanol (1P) in which the balance of the polar to non-polar surfaces tips towards the more hydrophobic side. It seems therefore mandatory to study the complete replica of the present 1P probing study by using another probe that is more hydrophilic than 1P.

In closing, we comment on the recent findings on

Table 1 The effects of ions on the molecular organization of H_2O, as probed by the induced changes in the $H^{\mathrm{E}}_{\mathrm{1P\text{-}1P}}$ pattern in 1P-S-H_2O. Ions are listed in the order of the Hofmeister ranking, eq.(1), from top to bottom.

Ion (S)	$\mu^{\mathrm{E}}_{\mathrm{1P\text{-}S}}/RT$	$\mathrm{H}^{\mathrm{E}}_{\mathrm{1P\text{-}S}}/RT$	What the ion does to H_2O
SO_4^{2-}	52 ± 3	75 ± 1	Hydration Center, $n_{\mathrm{H}} = 17 \pm 3$, plus an extra hydrophobe-like effect. See text.
F^-	32 ± 4	45 ± 2	Hydration Center, $n_{\mathrm{H}} = 14 \pm 2$.
Cl^-	22 ± 3	34 ± 1	Hydration Center, $n_{\mathrm{H}} = 2.3 \pm 0.6$.
Br^-			Hydrophile.
I^-	9 ± 1	43 ± 2	Hydrophile, stronger than Br^-.
ClO_4^-	11 ± 3	65 ± 4	Overall hydrophile, stronger than I^-, but with some hydrophobic effect.[1,10]

aqueous solutions of salts by modern spectroscopy/diffraction/relaxation methods. The femto second two-color mid-infrared pump-probe spectroscopy can distinguish the dynamics of H_2O in the hydration shell from that in the bulk H_2O outside of the hydration shells. By this fine technique, the rotational dynamics of H_2O in the hydration shell was found much slower than in the bulk H_2O for Cl^-, Br^- and I^- [28,29] and for SO_4^{2-} and ClO_4^-.[30] Furthermore, the dynamics of the bulk H_2O away from the hydration shell were found to remain the same as that in pure H_2O. More recently, Soper & Weckstrom drew similar conclusion by neutron diffraction studies.[31] While these two sets of conclusions for F^- and Cl^- are consistent with our findings summarized in Table, those for SO_4^{2-}, Br^-, I^-, and ClO_4^- are not. The information gained by this spectroscopic technique is also insensitive to the change of the counter cation from Na^+ to Mg^{2+}.[30] The dielectric relaxation studies has drawn a similar conclusion.[32] Namely, the impacts of anions spanning from a kosmotropic SO_4^{2-} to a chaotropic ClO_4^- on the H_2O dynamics have almost no effect on the relaxation time of the bulk H_2O network, although there are very subtle differences in the concentration dependence of the relaxation time. Why these very fine, microscopic techniques are insensitive in distinguishing the effects of different salts on the molecular organization of bulk H_2O, different from our findings and the century long biochemical experiences since Hofmeister (1987)? When we could provide answer to this question, we would be, no doubt, a step closer to the reality of aqueous solutions of electrolytes.

Spectroscopies/diffractions are generally regarded as a superior technique to thermodynamic investigations in order to advance the molecular level understandings. Or are they? At the risk of being simplistic, we gain structural information by these techniques. After all, the structure is governed, in terms of thermodynamics, by the Gibbs energy, G, which is the net results of all the possible higher derivative quantities. To begin with, G is the difference of the H and TS terms, and the more detailed information given in H and TS separately could be obscured by the H-S compensation prevalent in aqueous solutions.[11,12] H and S are consisting of the partial molar enthalpies and entropies, which in turn compensate with each other obscuring the details in the resulting chemical potentials. And so on. Thus, in order to advance the molecular level understanding, it is mandatory to obtain higher order derivative quantities experimentally for thermodynamic studies. And a further development of a finer non-linear spectroscopy is awaited. There is no doubt that the afore-mentioned techniques have provided the first step towards this end.

Acknowledgement

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要　旨

いくつかの陰イオンが水の分子凝集構造に与える影響を，私らが開発した1－プロパノール（1P）・プロービングの方法で調べた。1P－サンプル塩（S）－H_2Oにおいて，三次微分量である1P－1Pエンタルピー相互作用$H^{E}_{1P\text{-}1P}$を実験的に求めた。サンプル塩を加えたことにより，$H^{E}_{1P\text{-}1P}$のパターンが変化する。その変化の仕方から，サンプル塩が水にどのような影響をあたえたか調べた。SO_4^{2-}，F^-，Cl^-，Br^-，I^-およびClO_4^-のNa塩をサンプル塩として用いた。SO_4^{2-}，F^-およびCl^-は基本的にHydration Centerとして働き，それぞれ7，14および2.3個の水分子が水和する。一方，Br^-, I^-およびClO_4^-は基本的に親水的な働きをし，水の水素結合網に加担するかたちになる。そのため，水本来の水素結合網内で不純物センターとなり，揺らぎの度合いをさげる。その効果は$Br^- < I^- < ClO_4^-$の順であった。いわゆるHofmeister効果（塩析・塩溶効果）の序列と比較すると，Br^-とCl^-を境にして，塩析側ではHydration Centerであり，かつ水和の度合いが強いものほど，塩析効果が強い。塩溶のほうでは，より親水的なイオンほど塩溶効果が強いことが明らかになった。

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研究テーマ：水溶液
趣味：テニスはJake (76)，司馬遼太郎は「街道を行く」，バッハは全曲