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Salting-out Phenomenon and Clathrate Hydrate Formation in Aqueous Solution of Polar Nonelectrolyte

Tadashi Kato,* Masako Yudasaka, and (the late) Tsunetake Fujiyama
Institute for Molecular Science, Myodaiji, Okazaki 444

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The Salting-out Phenomenon has been carefully observed by adding ammonium chloride into aqueous solutions of *t*-butyl alcohol, acetone, dioxane, and 2-butoxyethanol. The experimental results have been well explained by considering the formation of a clathrate hydrate-like local structure in the aqueous solution of each nonelectrolyte. In addition, the role of the chloride ion has been found to be the promotion of the aggregation of the clathrate hydrate-like local structures. The line-width measurement of ^{35}Cl-NMR spectra has been shown to be an extremely powerful method for studying the state of mixing of ions in solutions.

The mutual solubility of a nonelectrolyte and water is decreased by the addition of inorganic salts. The interpretation of this salting-out phenomenon from a molecular viewpoint originated in the work of Debye *et al.*[1] They presented an equation which relates the solubility of a nonelectrolyte with the salt concentration in terms of the dielectric constant of the solution. Later on, some modifications of Debye's theory have been reported.[2–4] These theories are based on the idea that the solubilities of nonelectrolytes diminish by adding salts into aqueous solutions because the ions draw water molecules around themselves and, therefore, rob the water molecules from the nonelectrolytes. The scaled particle theory has been proposed from a completely different point of view.[5] This theory does not imply any model for the structure of the solvent and calculates thermodynamic quantities through the estimation of a free energy which is necessary first to make cavities in solvents and, then, to introduce nonelectrolyte molecules into the cavities. There are currently many attempts to explain salting-out phenomena by the use of this theory.[6,7]

In general, the solubilities of polar nonelectrolytes in water are much larger than those of nonpolar molecules; sometimes the two components are miscible in all proportions. Therefore, the salt concentration required to cause a phase-separation is higher than that in the case of nonpolar molecules. However, the application of the foregoing theories has been limited to the very low concentration range of salt or nonelectrolyte. Solubility curves such as those shown in Fig. 1 cannot be explained by these theories even qualitatively.

In our previous reports, the existence of clathrate hydrate-like local structures has been suggested in aqueous solutions of polar nonelectrolytes such as *t*-butyl alcohol (TBA) and acetone.[8–10] Although these nonelectrolytes are miscible with water in all proportions, the addition of salt causes phase-separation.

In the present work, we consider the salting-out phenomena based on the results of the above studies and emphasize the role of a clathrate hydrate-like local structure formed in the aqueous solutions of a nonelectrolyte. It will be shown that the line-width measurement of the ^{35}Cl-NMR spectrum is a useful method for the clarification of these problems. Ammonium chloride was chosen as a salt because it has been known that ammonium chloride changes the structure of water very little.[11]

Experimental

Materials. Dioxane, *t*-butyl alcohol, 2-butoxyethanol, and ammonium chloride were commercially available in a guaranteed grade; they were used without further purification. Water was distilled and passed through an exchange resin.

Determination of Solubility Curve. Weighed ammounts of water, nonelectrolyte, and ammonium chloride were sealed in glass tubes. The phase-separation temperature, T_p, of each ternary solution was determined in a water bath by the observation of the temperature at which the solution became turbid. The accuracy of T_p was ± 0.5 °C. The solubility curves at various temperatures were determined by the observation of T_p at various concentrations.

NMR Measurement. The ^{35}Cl-NMR spectra were recorded on a Varian FT80A spectrometer at 7.794 MHz. For a spectral width of 4000 Hz and an aquisition time of 0.4 s. 8000 data points are available in the time domain spectra. The alpha delay was kept at 1000 s, while the Fourier number was kept at 16384. No weighting function was applied; this avoided artificial broadening of the line-widths. The accuracy of the half-line-width was ± 1.5 Hz.

Results and Discussion

Solubility Curve. The solubility curves of ternary solutions of water, ammonium chloride, and various nonelectrolytes (TBA, acetone, dioxane, and 2-butoxyethanol) at 40 °C are shown in Fig. 1. We define two kinds of concentrations, X_2' and X_3,

$$X_2' = \frac{n_2}{n_1 + n_2}$$

and

$$X_3 = \frac{n_3}{n_1 + n_2 + n_3},$$

where n_1, n_2, and n_3 are the number of moles of a water, nonelectrolyte, and ammonium chloride, respectively. In Fig. 1, X_2' is plotted as abscissa, while X_3 as ordinate. We define here a new quantity, $X_3^{\min}$ as the minimum value of the mole fraction of ammonium chloride which is necessary to make the ternary solution separate into two liquid phases. The figure shows that $X_3^{\min}$ decreases rapidly as X_2' increases in the low concentration range. For each system, there exists a concentration range where ammonium chloride is precipitated before phase-separation occurs. This concentration range is defined as $0 \leq X_2' \leq X_2^p$.[12] For example, X_2^p is about 0.03 for the TBA system. Figure 2 shows the

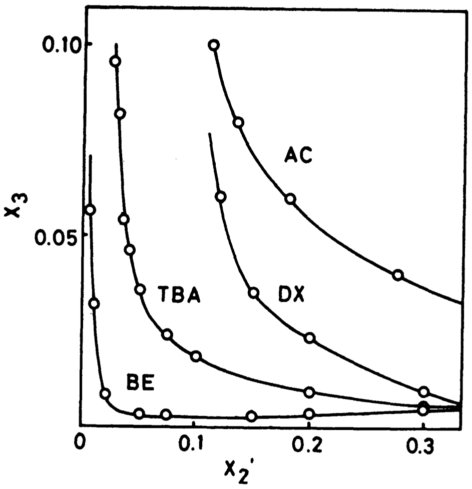

Fig. 1. Solubility curve for ternary solutions of water, ammonium chloride, and various nonelectrolytes (acetone(AC), dioxane(DX), *t*-butyl alcohol(TBA), and 2-butoxyethanol(BE)) at 40 °C.

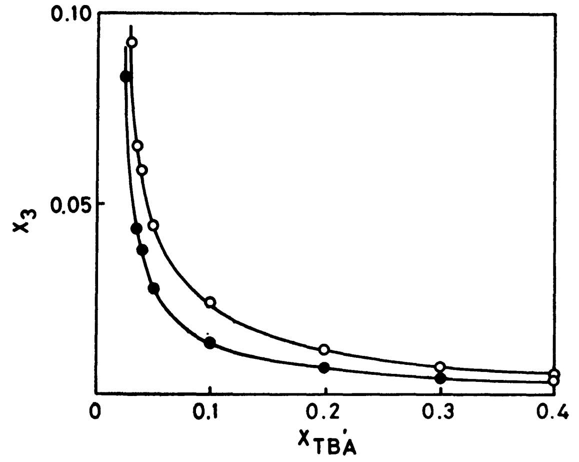

Fig. 2. Solubility curve for TBA–water–ammonium chloride solution at 30 °C (○) and 50 °C (●).

solubility curves for the TBA system at 30 °C and 50 °C. The figure shows that X_3^{min} decreases as the temperature increases, but that X_2^s is only slightly dependent on temperature. These results may depend on the mixing state of the binary solution of water and the nonelectrolyte. We will thus compare the solubility curves with the mean-square concentration fluctuations for the binary solution, which sensitively reflect the mixing state.

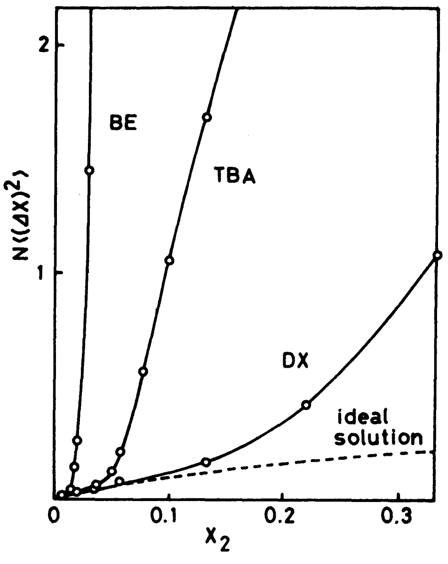

Fig. 3. Concentration dependence of mean-square concentration fluctuation for aqueous solutions of dioxane (DX), *t*-butyl alcohol(TBA), and 2-butoxyethanol(BE) at 40 °C.

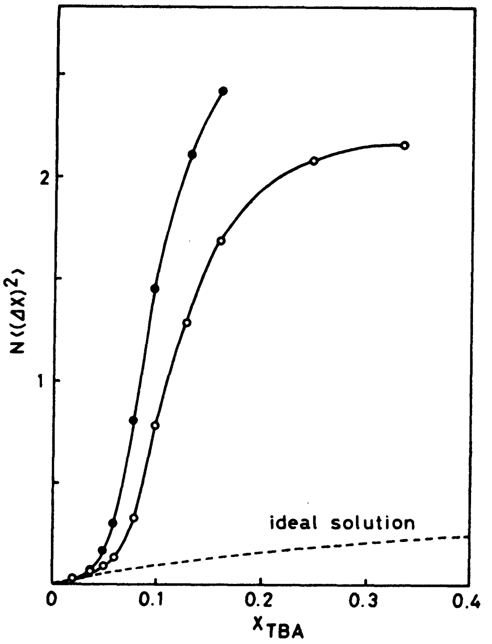

Fig. 4. Concentration dependence of mean-square concentration fluctuation for TBA–water solution at 24 °C (○) and 50 °C (●).

Concentration Fluctuations for Binary Solution and Solubility Curve. Figure 3 shows the mean-square concentration fluctuation, $N\langle(\Delta X)^2\rangle$, for the aqueous solutions of TBA,[8] dioxane,[8] and 2-butoxyethanol[13] at 40 °C where N is the mean number of total molecules included in the field within which the concentration fluctuations are considered. It is seen from the figure that the $N\langle(\Delta X)^2\rangle$ value is nearly equal to that for an ideal solution in the low concentration range and begins to increases abruptly at a certain mole fraction, X_2^s. For example, X_2^s is about 0.04 for the TBA system. Figures 1 and 3 show that X_2^s is nearly eaual to X_2^p. Figure 4 shows the temperature dependence of the $N\langle(\Delta X)^2\rangle$ values for the TBA–water system. $N\langle(\Delta X)^2\rangle$ increases in magnitude with the elevation of temperature in the concentration range of $X_{TBA}>X_{TBA}^s$. Similar results have been obtained for other systems. These results suggest that the solution is rapidly approaching the phase-separation with the increase of the temperature. In fact, 2-butoxyethanol(BE)–water system has a lower critical temperature. The phase diagram (T-X_2 curve) for this system is shown in Fig. 5.[14] Figures 3 and 5 show

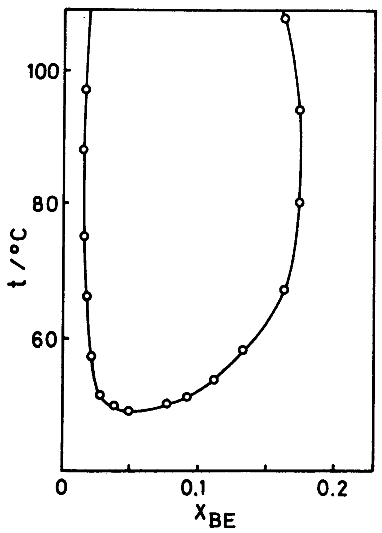

Fig. 5. Phase diagram for 2-butoxyethanol–water solution.

that the phase diagram is strongly correlated with the concentration dependence of the $N<(\Delta X>)^2$ value. The phase-separation temperature decreases with the increase of $N<(\Delta X)^2>$ in the concentration range of $X_{BE}<X^{s}_{BE}$ and the phase-separation does not occur in the concentration range of $X_{BE}<X^{s}_{BE}$. Similar results have been obtained for other systems, such as, the 2,6-lutidine–water system.[13] The relation between the X_3-X_2' curve (Fig. 1) and the concentration dependence of the $N<(\Delta X)^2>$ value is similar to that between the T-X_2 curve and the concentration dependence of the $N<(\Delta X)^2>$ value. X_2^{min} rapidly decreases with the rapid increase of the $N<(\Delta X)^2>$ value and salting-out does not occur in the concentration range of $X_2<X_2^s$. It can be concluded that an addition of salt causes phenomenologically the same change of the mixing state of the binary aqueous solutions as that caused by an elevation of temperature.

The temperature dependence of the mixing state for the TBA–water system has been discussed based on the quantitative analysis of the concentration fluctuations.[8,10] The conclusion is as follows. In the concentration range of $0<X_{TBA}<X^{s}_{TBA}$, water molecules form a polyhedron which encages a TBA molecule in water. In the concentration range of $X_{TBA}>X^{s}_{TBA}$, the polyhedra gather together in TBA. As the temperature increases, the polyhedra continue gather in this concentration range. This suggest that microscopic phase-separation occurs in the solution. One is a TBA-rich phase and the other is a water-rich phase. In the water-rich phase, almost all the molecules take part in the formation of polyhedra.

The salting-out phenomenon is considered to be governed by a mechanism similar to that described above. Namely, on adding ammonium chloride into an aqueous solution of TBA, the polyhedra formed in the TBA–water solution gather together; this results in the separation of the water-rich layer.

Surroundings of Ion in Aqueous Solution of 'Nonelectrolyte. We now consider the state of mixing of ammonium chloride in the aqueous solution of a nonelectrolyte based on the results of the measurement of the ^{35}Cl-NMR spectra. The half-line-width of ^{35}Cl-NMR spectra, $\Delta\nu$, is proportional to the product of the square of the electric field gradient at the nucleus, q^2, and the correlation time of the fluctuation of the electric field gradient at the nucleus, τ_c.[15] The magnitude of q^2 depends on the local asymmetry of the local structure around the chloride ion. If τ_c is assumed to be proportional to viscosity η, we can discuss the concentration or temperature dependence of the asymmetry of the local structure around the chloride ion from the concentration or temperature dependence of the quantity, $\Delta\nu^\circ \equiv \Delta\nu/(\eta/\eta_0)$, where η_0 is the viscosity of the solution whose $\Delta\nu$ value is chosen as reference.[16] As an absolute value of $\Delta\nu^\circ$ will not be discussed in the present report, the η_0 value will be fixed to 10^{-3} Pa s.

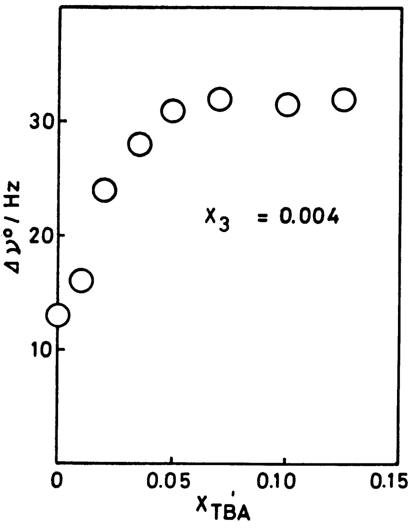

Fig. 6. Dependence of the $\Delta\nu^\circ$ value (see tle text) for TBA–water–ammonium chloride solution on the mole fraction of TBA at 32 °C (mole fraction of ammonium chloride is 0.004).

Figure 6 shows the relation between $\Delta\nu^\circ$ and X'_{TBA} for $X_3=0.004$ at 32 °C. It is seen from the figure that $\Delta\nu^\circ$ increases in magnitude as X'_{TBA} increases in the concentration range of $0<X'_{TBA}<0.05$. In the aqueous solution of ammonium chloride ($X'_{TBA}=0$), the $\Delta\nu^\circ$ value was observed to be only about 13 Hz for the wide salt concentration range from dilute to $X_3=0.11$. As the molar ratio of (ammonium ion) : (chloride ion): (water) at $X_3=0.11$ is about 1 : 1 : 9, there are only two layers of water molecules between ammonium and chloride ions. This result indicates that $\Delta\nu^\circ$ increases only when the symmetry of the local structure around a chloride ion is broken down. The increase of $\Delta\nu^\circ$ observed in the TBA–water–ammonium chloride solution may reflect the break-down of the local symmetry around the chloride ion. Some chloride ions probably take part in the formation of the polyhedra in the solution. The concentration dependence of $\Delta\nu^\circ$ confirms the validity of the above picture. As ammonium chloride is only slightly soluble in TBA by itself, the number of chloride ions which take part in the formation of the polyhedra is expected to be proportional to the number ratio, R, of water molecules which form polyhedra to all the water molecules. According to the results obtained from the analysis of concentration fluctuations, R can be calculated from the relations:[8)]

$$R = lX_{\mathrm{TBA}}/(1-X_{\mathrm{TBA}}) \quad \text{for } 0<X_{\mathrm{TBA}}<X^{\mathrm{s}}_{\mathrm{TBA}},$$

$$R = 1 \quad \text{for } X_{\mathrm{TBA}}>X^{\mathrm{s}}_{\mathrm{TBA}},$$

where $l(\simeq 20)$ is the number of water molecules which encage a TBA molecule. Figure 7 shows the concentration dependence of R calculated for $l=20$. It can be seen from Figs. 6 and 7 that $\Delta\nu^\circ$ is nearly proportional to R.

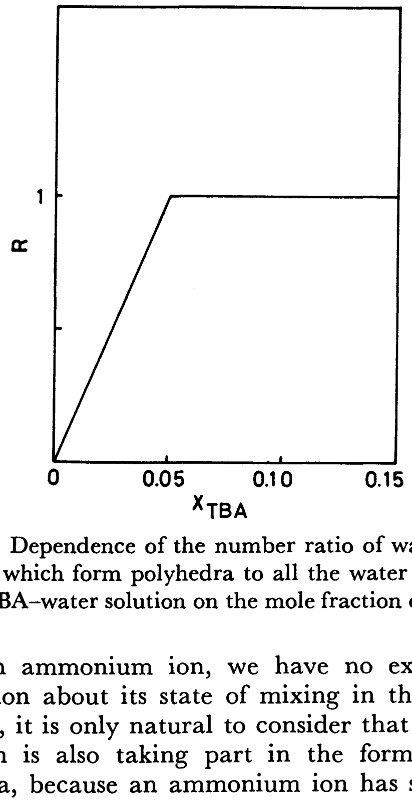

Fig. 7. Dependence of the number ratio of water molecules which form polyhedra to all the water molecules for TBA–water solution on the mole fraction of TBA.

For an ammonium ion, we have no experimental information about its state of mixing in this solution. However, it is only natural to consider that an ammonium ion is also taking part in the formation of a polyhedra, because an ammonium ion has similar size and property to those of a water molecule. Therefore, the water molecule which forms a polyhedra can be replaced by an ammonium ion much more easily than by a chloride ion.

Role of Ammonium Chloride in Phase-separation. Next, we focus our attention on the dependence of the $\Delta\nu^\circ$ value on salt-concentration. The acetone (AC) system was chosen for this experiment because the salt-concentration can be widely changed in a one-phase region. In Fig. 8, the $\Delta\nu^\circ$ values observed for $X'_{\mathrm{AC}}=0.3$ at 32 °C are plotted against the salt-concentration, X_3. It can be seen from the figure that $\Delta\nu^\circ$ decreases its magnitude with the increase of the ammonium chloride concentration. Note that the phase-separation occurs at $X_3>0.004$. These results cannot be explained if we consider that all the chloride ions are engaged in the formation of polyhedra, because the local symmetry around a chloride ion should attain higher symmetry with the increase of salt-concentration. Therefore, it may be reasonable to consider that, at high salt-concentration, some chloride ions can exist in this solution, surrounded by polyhedra. In other words, some chloride ions can attract polyhedra around themselves, which results in the formation of a high local symmetry around the chloride ion. As the aggregation of polyhedra is one of the necessary conditions for phase-separation, it can be said that chloride ions promote the phase-separation by attracting polyhedra around themselves.[17)]

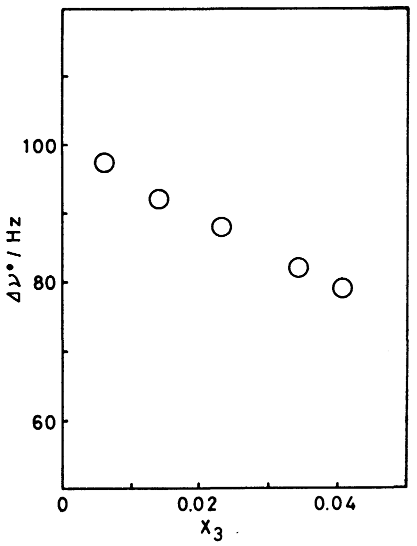

Fig. 8. Dependence of the $\Delta\nu^\circ$ value for acetone–water ammonium chloride solution on the mole fraction of ammonium chloride at 32 °C (mole fraction of acetone is 0.3).

From the above discussion, the picture for the salting-out phenomenon can be drawn as follows. When ammonium chloride is added to the aqueous solution of nonelectrolyte in the concentration range of $X_2>X^{\mathrm{s}}_2$, not only water molecules but also ammonium and chloride ions form polyhedra and each of them encages a nonelectrolyte molecule. As the concentration of ammonium chloride increases, the polyhedra begin to gather around a chloride ion. When the concentration of ammonium chloride is raised to X^{min}_3, the aggregate of the polyhedra grows into a macroscopic phase and the phase-separation occurs. This situation is schematically visualized in Fig. 9.

Lastly, we consider the temperature dependence of the mixing state at a constant salt-concentration. The temperature dependence of the $\Delta\nu^\circ$ value observed for $X'_{\mathrm{AC}}=0.3$ and $X_3=0.04$ is shown in Fig. 10. Note that phase-separation occurs at 33 °C at this concentration. It can be seen from the figure that the $\Delta\nu^\circ$ value decreases with the elevation of temperature. This experimental result corresponds to the fact that, as the temperature approaches the phase-separation temperature, polyhedra begin to gather together around a chloride ion, which in turn results in the formation of a symmetric local field around a chloride ion.

Concluding Discussion. We have shown the existence of the clathrate hydrate-like local structure plays an important role in the salting-out phenomena in the aqueous solutions of polar nonelectrolytes such as TBA and acetone. Even if the solution does not contain

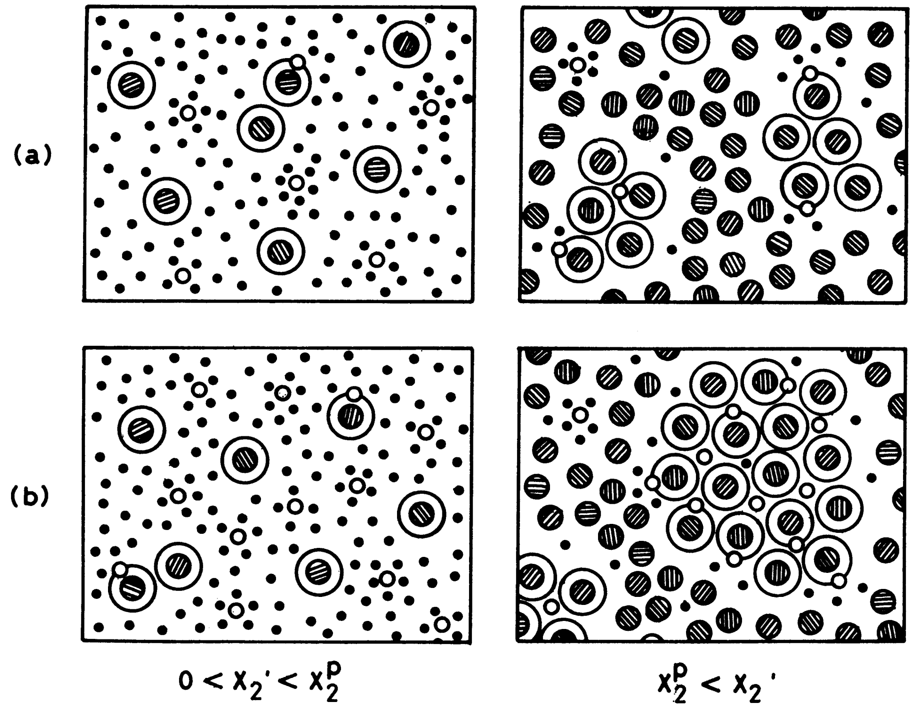

Fig. 9. Mixing state of ternary solution of water–ammonium chloride–nonelectrolyte (schematic) in the low (a) and high (b) concentration range of ammonium chloride. (•) and (●) represent a water and a TBA molecule, respectively. (◎) Represents the clathrate hydrate-like local structure. (○) Represents an ammonium or chloride ion.

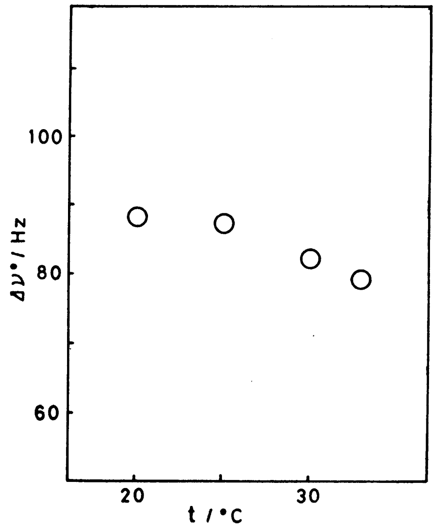

Fig. 10. Temperature dependence of the $\Delta\nu^\circ$ value for acetone–water–ammonium chloride solution (mole fractions of acetone and ammonium chloride are 0.3 and 0.04, respectively).

salts, the chlathrate hydrate-like local structure grows with the increase of the temperature. That is, the number of polyhedra which gather increases as the temperature increases. This property of the solution is considered to originate in the difference between the affinity of the polyhedron (strictly speaking, the water molecules which form a polyhedron) for nonelectrolyte and the affinity of the polyhedron for the polyhedron. The salt performs the task of promoting the gathering of the polyhedra. The present result shows that the ions take part in the formation of polyhedra; this may arise from the fact that the affinity of the ion for nonelectrolyte is weaker than that of the ion for water. The facts that the salting-out does not occur in the concentration range of $X_2 < X_2^s$ can be explained from this point of view. Roughly speaking, the polyhedra rarely contact with nonelectrolyte molecules in this concentration range because most of the nonelectrolyte molecules are encaged by water molecules. So, the phase-separation does not occur.

The above picture is rather different from the classical picture that ions rob the water molecules from the nonelectrolyte. Of course, we are not going to deny this classical picture wholly. In fact, the result that X_2^p is slightly smaller than X_2^s may arise from the fact that the number of water molecules which can form polyhedra is decreased by some ions which rob water molecules from the polyhedra. However, the rapid decrease of $X_3^{\min}$ with the increase of X_2', and the existence of the concentration range where salting-out does not occur cannot be explained by the classical picture.

The strong correlation between the solubility curve for the ternary solution and the concentration dependence of the concentration fluctuation for the binary solution indicates that the phase behavior is determined principally by the mixing state of the binary solution at least in the concentration range near X_2^s. So, the validity of our picture is considered to be dependent on the validity of the picture for the mixing state of the binary solution.

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17) Similar pictures can be drawn for other systems. As X_2^p is nearly equal to X_2^s, the difference in X_2^p values is considered to come from the difference in the size of the hydrophobic group of the molecule. See Ref. 9.