# Salting-In and Salting-Out of Water-Soluble Polymers in Aqueous Salt Solutions

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Ⓢ *Supporting Information*

ABSTRACT: To obtain further experimental evidence for the mechanisms of the salting effect produced by the addition of salting-out or sating-in inducing electrolytes to aqueous solutions of water-soluble polymers, systematic studies on the vapor−liquid equilibria and liquid−liquid equilibria of aqueous solutions of several polymers are performed in the presence of a large series of electrolytes. Polymers are polyethylene glycol 400 (PEG400), polyethylene glycol dimethyl ether 250 (PEGDME250), polyethylene glycol dimethyl ether 2000 (PEGDME2000), and polypropylene glycol 400 (PPG400), and the investigated electrolytes are KCl, NH_4Cl, $MgCl_2$, $(CH_3)_4NCl$, $NaCl$, $NaNO_3$, Na_2CO_3, Na_2SO_4, and Na_3Cit

(tri-sodium citrate). Aqueous solutions of PPG400 form aqueous two-phase systems with all the investigated salts; however, other investigated polymers form aqueous two-phase systems only with Na_2CO_3, Na_2SO_4, and Na_3Cit. A relation was found between the salting-out or sating-in effects of electrolyte on the polymer aqueous solutions and the slopes of the constant water activity lines of ternary polymer−salt aqueous solutions, so that, in the case of the salting-out effect, the constant water activity lines had a concave slope, but in the case of the salting-in effects, the constant water activity lines had a convex slope. The effect of temperature, anion of electrolyte, cation of electrolyte, and type and molar mass of polymers were studied and the results interpreted in terms of the solute−water and solute−solute interactions. The salting-out effect results from the formation of ion (specially anion)−water hydration complexes, which, in turn, decreases hydration, and hence, the solubility of the polymer and the salting-in effect results from a direct binding of the cations to the ether oxygens of the polymers.
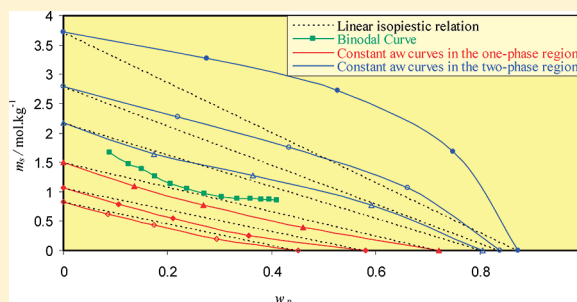
■ INTRODUCTION

Ternary aqueous solutions of water-soluble polymers and certain electrolytes are separated into a polymer-rich phase and a salt-rich phase with water as solvent in both phases over part of the composition space. The aqueous nature of both phases as well as the difference in their properties makes it possible to use them for the partitioning and separation of biological materials such as cells, organelles, enzymes, proteins,[1−3] metal ions,[4] dyes, drug molecules, and small organic species,[5,6] and nano- and microsolid particles[7,8] from the complex mixtures in which they are produced. Although determination of phase equilibrium data of these aqueous two-phase systems and their application to purification of a variety of biomolecules has been studied extensively,[9] experimental efforts aiming at the understanding of the underlying reasons for the formation of aqueous two-phase systems are considerably fewer and there remains considerable uncertainty as to the details of the mechanism of this phase separation. In fact, the effect of the addition of salts on solutions of polymers is very complex, primarily because a large number of different types of intermolecular interactions come into play between the ion and water, ion and polymer, and polymer and water. This is further complicated by the fact that the extent of the interactions varies in relation to the types of ions and polymers involved. Ananthapadmanabhan and Goddard[10,11] performed a

systematic investigation on the aqueous two-phase formation characteristics of polyethylene glycol (PEG) and electrolytes in water. They found that the effectiveness of various salts to form two-phase systems with PEG is similar to their tendency to depress the cloud point of polyethoxylates. They also found that for univalent sodium salts, hydroxide and fluoride caused aqueous two-phase system formation, whereas sodium chloride and bromide were not capable and for a given anion, the cation efficacy followed the order $Na^+ > Mg^{2+} > Zn^{2+} > Li^+$. These differences have been described on the basis of specific interactions between the polymer and ions. Rogers[4] and co-workers reported a correlation between the efficacy of a series of cations in inducing aqueous two-phase system formation and their hydration enthalpies and Gibbs energies. da Silva and Loh[12] investigated the evolution of ternary mixtures containing water, PEG, and dextran or electrolytes toward the formation of aqueous two-phase systems by calorimetric measurements of enthalpies of solution for one of the components in water and in an aqueous solution of the other component. They found that phase separation was accompanied by an enthalpy increase, indicating that entropy increase was the driving force for

aqueous two-phase systems formation and based on the obtained enthalpy data, they also assigned different behavior to electrolytes capable of inducing aqueous two-phase systems formation (lithium and sodium sulfates), and sodium chloride, which did not induce phase separation. Recently, Freire et al.[13] have shown that distinct pairs of PEG and ionic liquids can induce phase separation in aqueous media when dissolved at appropriate concentrations. They observed that contrary to the typical ionic liquid-inorganic salt aqueous biphasic systems, in which the phase behavior is dominated by the formation of the hydration complexes of the ions, the interactions between the PEG polymers and ionic liquids control the phase demixing in these systems and depend on the structural features of the ionic liquids, both the ionic liquids and PEG polymers can act as the salting-out species.

In an attempt to obtain further evidence about the salting effect produced by the addition of different salts to aqueous solutions of water-soluble polymers, a series of ternary systems containing water-soluble polymers and electrolytes capable or not of inducing phase separation were investigated in the present work. To cover a range of kosmotropic and chaotropic behaviors, the polymers polyethylene glycol 400 (PEG400), polyethylene glycol dimethyl ether 250 (PEGDME250), polyethylene glycol dimethyl ether 2000 (PEGDME2000), and polypropylene glycol 400 (PPG400) and electrolytes KCl, NH_4Cl, $MgCl_2$, $(CH_3)_4NCl$, NaCl, $NaNO_3$, Na_2CO_3, Na_2SO_4, and Na_3Cit were investigated. To make a thorough analysis of the salting-out effects in aqueous polymer solutions and to obtain a relation between the vapor–liquid equilibria and liquid–liquid equilibria behavior of the polymer–salt aqueous biphasic systems, isopiestic equilibrium molalities of binary polymer/salt + water and ternary polymer + salt + water systems in both one-phase and two-phase areas together with liquid–liquid equilibria phase diagrams of the polymer–salt aqueous two-phase systems containing the above water-soluble polymers and electrolytes were determined. These studies show for the first time that there is a relation between the salting effects of electrolyte on the polymer aqueous solutions and the slopes of the constant water activity lines of the corresponding ternary polymer–salt aqueous solutions and the salts influence on the water miscibility of the polymers qualitatively follows the Hofmeister series.

■ EXPERIMENTAL SECTION

Materials. PPG400 was obtained from Fluka. PEGDME250, PEGDME2000, PEG400, NaCl, KCl, NH_4Cl, $MgCl_2$, $(CH_3)_4NCl$, $NaNO_3$, Na_2CO_3, Na_2SO_4, and Na_3Cit were purchased from Merck. The polymers and salts were used without further purification. Double distilled and deionized water was used.

Experimental Procedures. All the solutions were prepared by mass on a Sartorius CP124S balance precisely within $\pm 1 \times 10^{-7}$ kg. The experimental apparatus employed for determination of liquid–liquid equilibrium data is essentially similar to the one used previously.[14] A glass vessel, volume 50 cm³, was used to carry out the phase equilibrium determinations. The glass vessel was provided with an external jacket in which water at constant temperature was circulated using a thermostat. The temperature was controlled to within ± 0.05 K. The binodal curves were determined by a titration method. A salt solution of known concentration was titrated with the polymer solution or vice versa, until the solution turned turbid.

In this study, the improved isopiestic method was used to obtain the water activities of different systems. The isopiestic apparatus employed in this work was similar to the one used by Ochs et al.[15] This apparatus its operation and the experimental conditions and techniques have been essentially identical to that discussed by Ochs et al.[15] The only difference between our isopiestic apparatus and those of Ochs et al.[15] is in the number of flasks. The apparatus used for determination of water activity in the ternary PEGDME + salt + water solutions consisted of a nine-leg manifold attached to round-bottom flasks. One flask contained the standard pure NaCl solution, the other flask contained the pure PEGDME250 solution, one flask contained the pure PEGDME2000 solution, the other one contained the pure salt solution, two flasks contained the PEGDME250 + salt + water solutions, two others contained the PEGDME2000 + salt + water solutions, and the central flask was used as a water reservoir. However, the apparatus used for determination of water activity in the ternary PEG/PPG + salt + water solutions consisted of an eight-leg manifold attached to round-bottom flasks. Two flasks contained the standard pure NaCl solutions, one contained the pure PEG/PPG solution, another one contained the pure salt solution, three others contained the PEG/PPG + salt + water solutions and the central one was used as a water reservoir. The apparatus was held in a constant-temperature bath at least 5 days (depending on the solutes concentration) for equilibrium. Equilibrations were performed generally within 5–10 days for intermediate and concentrated solutions and about 15 days for low molalities. During the equilibration process the manifold was removed at least once a day and the samples were agitated. After the third day, samples were not agitated, but left in the bath to approach their final equilibrium conditions. The temperature was controlled to within ± 0.05 K. After equilibrium had been reached, the manifold assembly was removed from the bath, and each flask was weighed with an analytical balance with a precision of $\pm 1 \times 10^{-4}$ g. From the weight of each flask after equilibrium and the initial weight of salt and polymer, the mass fraction of each solution was calculated. At equilibrium, the chemical potentials of the solvent in each of the solutions in the closed system are identical. The equality of the solvent chemical potential implies the equality of the solvent activity. Because the solvent activity is known for one or more standard solutions, it will be known for each solution within the isopiestic system. The osmotic coefficients for the standard NaCl aqueous solutions have been calculated from correlation given in the literature.[16] The uncertainty in the measurement of solvent activity was estimated to be $\pm 5 \times 10^{-4}$.

■ RESULTS AND DISCUSSION

In the present work, two sets of experiments with more than 1600 different measured data were carried out to achieve further understanding about the salting effect produced by the addition of different salts to aqueous solutions of water-soluble polymers. In this respect, to individualize the effect of the anion and the cation, a large series of salts based on either Cl^- or Na^+ were used. To study the vapor–liquid equilibria behavior, the isopiestic equilibrium measurements at $T = 298.15$ K were carried out for the ternary PEGDME250 + salt + H_2O, PEGDME2000 + salt + H_2O, PEG400 + salt + H_2O, and PPG400 + salt + H_2O solutions containing the salts KCl, NH_4Cl, $MgCl_2$, $(CH_3)_4NCl$, NaCl, $NaNO_3$, Na_2CO_3, Na_2SO_4, and Na_3Cit. For the systems that form aqueous biphasic systems, the phase diagram measurements at $T = 298.15$,

308.15, and 318.15 K were also carried out to study their liquid−liquid equilibria behavior. The measured vapor−liquid and liquid−liquid equilibria data are presented in the Supporting Information of this manuscript.

Liquid−Liquid Equilibria Properties. The solubility of a water-soluble polymer in water may be decreased (salting-out effect and formation of aqueous two-phase system) or increased (salting-in effect) by the presence of an electrolyte, depending on the cation and anion of the electrolyte used, their concentration and the type and molar mass of the polymer. The results show that, aqueous solutions of PPG400 form aqueous two-phase systems with all investigated salts; however, other investigated polymers are capable only with Na_2CO_3, Na_2SO_4, and Na_3Cit.

To infer about the cation and anion effects on the mutual liquid−liquid mixing, several chloride and sodium-based salts were used, respectively. Figure 1 shows that the effectiveness of the cations in the salting-out of PPG400 follows the sequence $(NH_4)^+ < (CH_3)_4N^+ < K^+ < Na^+ < Mg^{2+}$ and that of the anions follows the order $NO_3^- < Cl^- < (H_2PO_4^-) < (HPO_4^{2-}) <$ SO_4^{2-} (low salt concentration) $< CO_3^{2-} < SO_4^{2-}$ (high salt concentration) $< (PO_4^{3-}) < Cit^{3-}$. Furthermore, the mutual liquid−liquid demixing in the studied aqueous PPG−electrolyte systems is promoted by increasing the temperature in the whole concentration range of solutes. In this figure, the data for the sodium phosphate salts were taken from the literature.[17,18] Because anions are typically more polarizable than cations[19,20] (due to their more diffuse valence electronic configuration), their hydration is usually stronger than that of cations; therefore, as can be seen from Figure 1, their salting-out effects are more pronounced. The salting-out inducing ions form preferentially hydration complexes with water and then are excluded from the near surface region of the polymer in solution. With an increase in the concentration of the solutes, the extent of exclusion will increase. Ultimately, the system could reach a state where, for entropic reasons, phase formation would become favorable, and therefore, these systems separate into a salt-rich phase and a polymer-rich phase over part of the composition space. Formation of aqueous two-phase systems can only involve partial dehydration of solutes. However, the salting-in inducing ions exclude water from their vicinity and interact with the polymer. Figure 1 shows that in the case of the higher the valency of the ion the lower salt concentration is required to form a two-phase system.

Figure 2 illustrates the results for the other investigated polymers with Na_2SO_4 or Na_2CO_3 at 298.15 and 318.15 K. As shown, PPG400 can be more easily salted-out by a salting-out-inducing ion to from the polymer-based aqueous two-phase system than PEG or PEGDMEs. The abilities of the investigated polymers for the formation of aqueous two-phase systems with salts follow the sequence PPG400 > PEGDME2000 > PEGDME250 > PEG400. The results show that in the case of the more hydrophobic polymers the lower the concentration is required to form a two-phase system. In fact, more hydrophobic polymers can be more easily salted-out by a salting-out-inducing ion to from the polymer-based aqueous two-phase system. The investigated polymers have the structures $HOCH_2[CH_2OCH_2]_nCH_2OH$ (PEG), $CH_3OCH_2[CH_2OCH_2]_nCH_2OCH_3$ (PEGDME), and HOCH-$(CH_3)[CH_2OCH(CH_3)]_nCH_2OH$ (PPG). PPG contains a greater proportion of hydrocarbon in its molecule. Furthermore, the side chain methyl groups in PPG hinder hydrogen bonding between water molecules and the ether oxygen atoms.

(a)

(b)
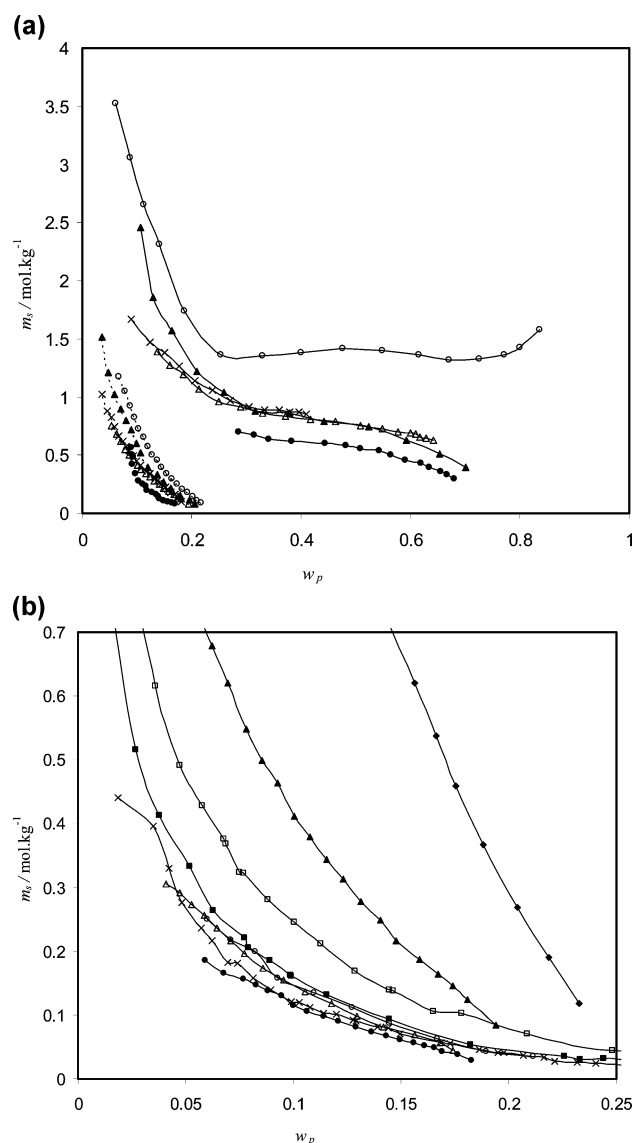
Figure 1. (a) Binodal curves for the PPG400 (p) + salt (s) + H_2O (w) two-phase systems at 298.15 K (solid line) and 318.15 K (dotted line): ●, $MgCl_2$; △, NaCl; ✕, KCl; ▲, $(CH_3)_4NCl$; ○, NH_4Cl. (b) Binodal curves for the PPG400 (p) + salt (s) + H_2O (w) two-phase systems at 318.15 K: ●, Na_3Cit; ○, Na_2CO_3; △, Na_2SO_4; ▲, NaCl; ◆, $NaNO_3$; □, NaH_2PO_4;[17] ■, Na_2HPO_4;[18] ✕, Na_3PO_4.[18]

Therefore, PPG can be more easily salted-out by a salting-out-inducing ion to from the polymer-based aqueous two-phase system. Although PEG and PEGDME have similar repeating units, the end groups of PEGDME (OCH_3) are still more hydrophobic than that of PEG (OH), and therefore, PEGDME can be more easily salted-out than PEG. PPG is a thermosensitive polymer and the effect of temperature on the phase forming ability of PPG is larger than those of other investigated polymers. PPG becomes more hydrophobic by increasing temperature,[21] and therefore, at higher temperatures, lower salt concentration is required to form a two-phase system. The binodal isotherms of the PEG or PEGDME + salt aqueous two-phase systems intersect at a certain concentration. For these systems, in the polymer-rich region, an increase in temperature causes the expansion of one-phase area, whereas, for the salt-rich region, expansion of the two-phase area is observed with increasing the temperature. However, for

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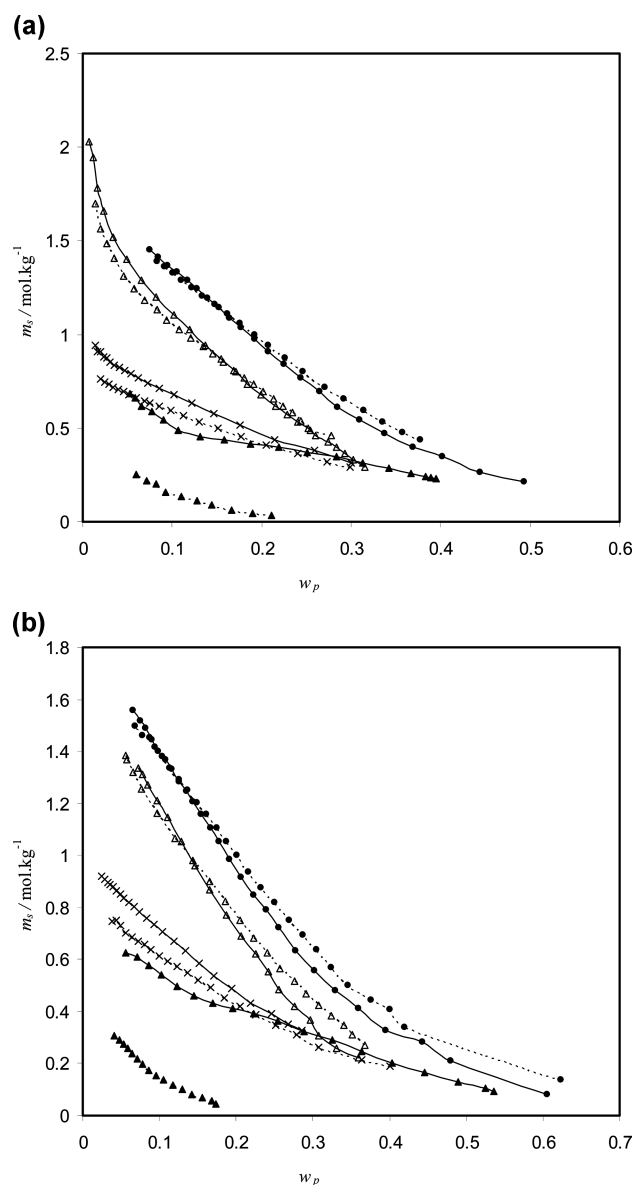
Figure 2. (a) Binodal curves for the Polymer (p) + Na_2CO_3 (s) + H_2O (w) two-phase systems at 298.15 K (solid line) and 318.15 K (dotted line): ●, PEG400; △, PEGDME250; ×, PEGDME2000; ▲, PPG400. (b) Binodal curves for the Polymer (p) + Na_2SO_4 (s) + H_2O (w) two-phase systems at 298.15 K (solid line) and 318.15 K (dotted line): ●, PEG400; △, PEGDME250; ×, PEGDME2000; ▲, PPG400.

systems containing PPG400 no crossing of binodal isotherms was observed. For these systems an increase in temperature causes an expansion of the two-phase area in the whole concentration range. The polymer and salt concentration in which the crossing of binodal curves occurs depend on the polymer molar mass, type of polymer and salt. The crossing point salt concentration for the PEG + Na_2SO_4 aqueous two-phase system is larger than those for the PEGDME + Na_2CO_3 aqueous two-phase system and decreases by increasing polymer molar mass. The reverse order was observed for the crossing point polymer concentration.

Vapor−Liquid Equilibria Properties. For the understanding of interactions in liquids, the activities of different components are of great interest. They are the most relevant thermodynamic reference data, and they are often the starting point of any modeling. The isopiestic method is the most

accurate, simple experimental technique available for measuring the solvent activity of solutions. It is based on the phenomenon that different solutions, when connected through the vapor space, approach equilibrium by transferring solvent mass by distillation. Equilibrium will be established once the temperature, pressure, and solvent chemical potential are uniform throughout the system, provided that no concentration gradients exist in the liquid phase. In this work, in an attempt to obtain further evidence about the salting effect produced by the addition of different salts to aqueous solutions of water-soluble polymers, and to obtain a relation between the vapor−liquid equilibria and liquid−liquid equilibria behavior of the polymer−salt aqueous biphasic systems, isopiestic equilibrium molalities of ternary aqueous systems containing water-soluble polymers and electrolytes capable or not of inducing phase separation in both one-phase and two-phase areas were determined. The water activity in aqueous solutions is affected by the solute−water interactions. Thus, if the mechanism of the salting effects involves dehydration of the polymer or association of ions with polar groups, the water activity is be expected to be strongly dependent on different ions.

As an example, the constant water activity lines of different (polymer + salt + H_2O) systems investigated in this work are plotted in Figures 3−5. In fact, all points on each line in these figures have a constant water activity or chemical potential and thus these points are in equilibrium. To further scrutinize the vapor−liquid equilibria behavior of the investigated systems, the linear isopiestic relation (Zdanovskii−Stokes−Robinson rule) derived using the semi-ideal hydration model was tested by comparing to the isopiestic measurements for the studied systems. Zdanovskii[22] proposed the following empirical rule for ternary aqueous electrolyte solutions under isopiestic equilibrium:

$$\sum_i \frac{m_i}{m_i^0} = 1 \quad \left(a_w = \text{constant and } 0 \le \frac{m_i}{m_i^0} \le 1\right) \tag{1}$$

where m_i is the molality of solute i in the ternary solution and m_i^0 is the molality of solute i in the binary solution of equal a_w. Stokes and Robinson[23] theoretically derived this equation for isopiestic mixed nonelectrolyte aqueous solutions from the semi-ideal hydration model. In this case, the semi-ideality means that the solute−solute interactions can be either mutually self-canceled or neglected and the solute−solvent interactions can be simply described by a hydration number. In fact, thermodynamic behavior of a mixed solution conforming to eq 1 is as simple as that of an ideal solution, that is, the constituent binary solutions mix ideally under isopiestic equilibrium. In this work the following form of the linear isopiestic relation was tested by comparing to the experimental constant water activity lines of the investigated systems.
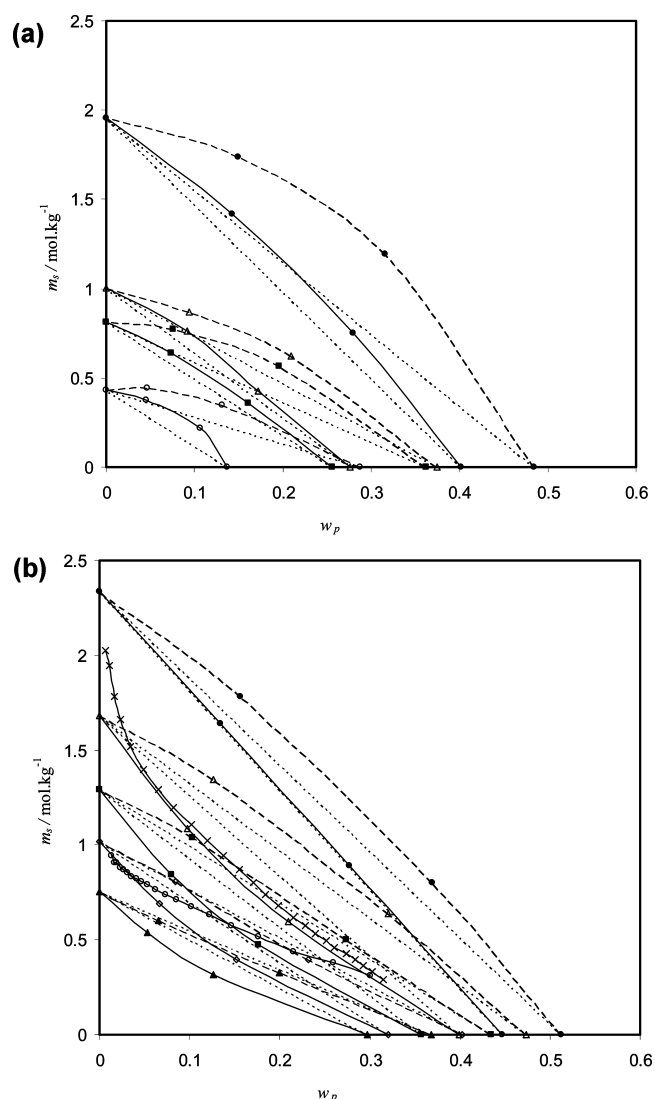
$$\frac{m_s}{m_s^0} + \frac{w_P}{w_P^0} = 1$$

$$\left(a_w = \text{constant}, 0 \le \frac{m_s}{m_s^0} \le 1 \text{ and } 0 \le \frac{w_P}{w_P^0} \le 1\right) \tag{2}$$

The results of this equation were also shown in Figures 3−5. As can be seen, the experimental constant water activity lines of the PEG400 + NH_4Cl + H_2O and PEGDME + $NaNO_3$ + H_2O systems (which do not form aqueous two-phase systems) show large positive deviations from eq 2. Similar behavior was

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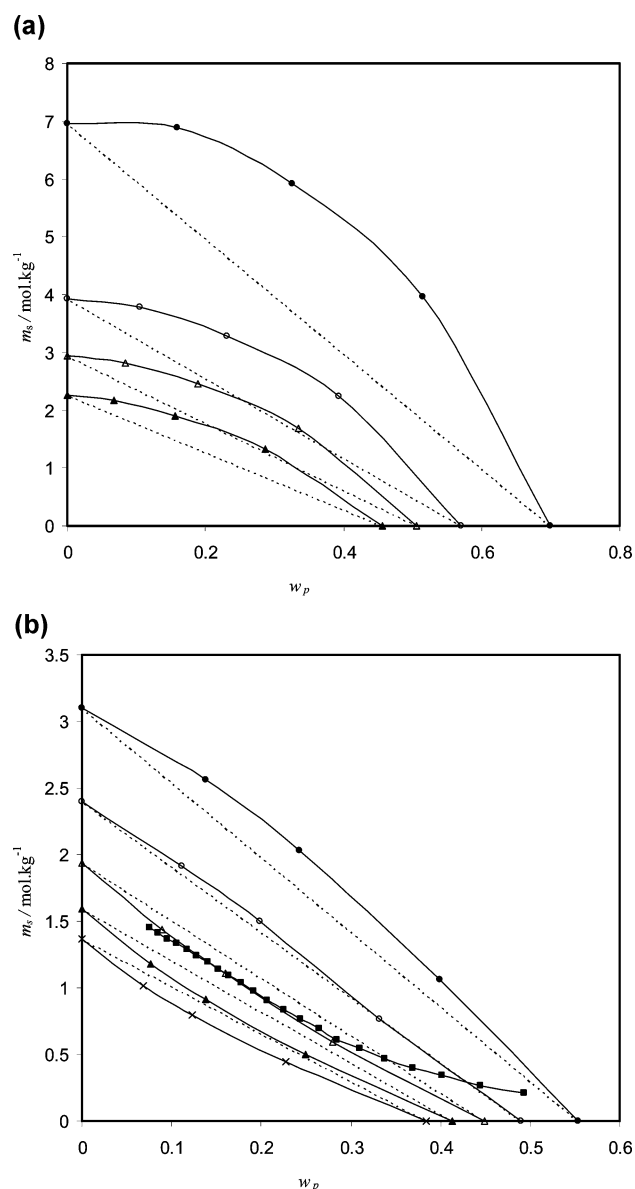
Figure 3. (a) Plot of molality of NH_4Cl, m_s, against mass fraction of PEG400, w_p, for constant water activity curves of PEG400 + NH_4Cl + H_2O system at 298.15 K: ●, 0.7791; ○, 0.8719; △, 0.9040; ▲, 0.9263. (b) Plot of molality of Na_2CO_3, m_s, against mass fraction of PEG400, w_p, for constant water activity curves of PEG400 + Na_2CO_3 + H_2O system at 298.15 K: ●, 0.8827; ○, 0.9139; △, 0.9311; ▲, 0.9434; ✕, 0.9499; ■, binodal curve; ⋯, calculated by eq 2.

(a)

(b)

Figure 4. (a) Plot of molality of $NaNO_3$, m_s, against mass fraction of PEGDME250 (solid line) and PEGDME2000 (dashed line), w_p, for constant water activity curves of PEGDME + $NaNO_3$ + H_2O system at 298.15 K: ●, 0.9420; △, 0.9718; ■, 0.9746; ○, 0.9861. (b) Plot of molality of Na_2CO_3, m_s, against mass fraction of PEGDME250 (solid line) and PEGDME2000 (dashed line), w_p, for constant water activity curves of PEGDME + Na_2CO_3 + H_2O system at 298.15 K: ●, 0.9173; △, 0.9663; ■, 0.9711; ◇, 0.9749; ▲, 0.9790; ✕, binodal curve for PEGDME250 + Na_2CO_3 + H_2O; ○, binodal curve for PEGDME2000 + Na_2CO_3 + H_2O; ⋯, calculated by eq 2.

observed for all investigated ternary aqueous polymer + salt systems that were not capable of inducing phase separation. However, for the ternary aqueous systems containing water-soluble polymers and electrolytes capable of inducing phase separation, the constant water activity lines in the one-phase and two-phase areas respectively show the negative and positive deviation from eq 2. Therefore, we can conclude that there may be a relation between the salting-out ability of the salts and the vapor−liquid equilibria behavior of ternary aqueous polymer + salt systems. In fact, for the systems that conform to eqs 1 and 2, the solute−solvent interactions in the ternary solution are the same as those in the corresponding binary solutions and, therefore, constituent binary solutions mix ideally under isopiestic equilibrium. Positive and negative deviations from

the linear isopiestic relation (eq 1 or 2) show that the concentrations of solutes in a ternary solution that is in isopiestic equilibrium with certain binary polymer + water and salt + water solutions respectively are larger and smaller than those we expect in the case of semi-ideal behavior.

In the case of ternary aqueous polymer + salt solutions that do not form aqueous biphasic systems (salting-in effect), because of the interactions between the salting-in inducing ions and polymer, the water molecules are excluded from their vicinity. As a consequence of these preferential interactions (polymer−ion interactions), it would be expected that the water molecules are allowed to relax to the bulk state and, therefore, more free water molecules would be available in respect to the semi-ideal behavior in which the solute−solvent interactions in the ternary solution are the same as those in the
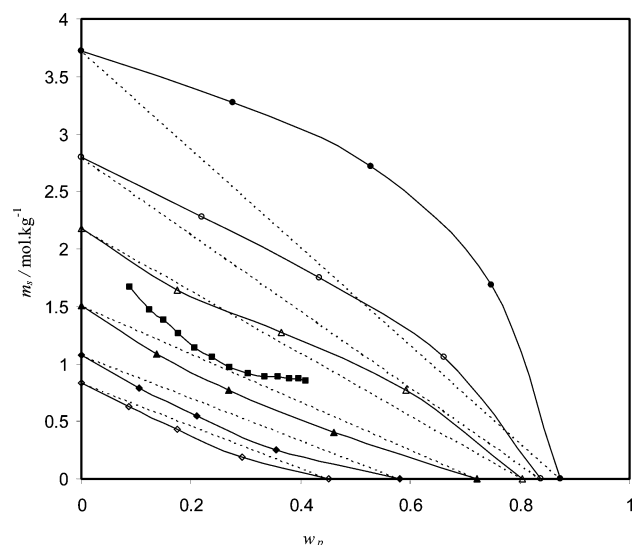
Figure 5. Plot of molality of KCl, m_s, against mass fraction of PPG400, w_p, for constant water activity curves of PPG400 + KCl + H$_2$O system at 298.15 K: ●, 0.8810; ○, 0.9112; △, 0.9312; ▲, 0.9529; ◆, 0.9663; ◇, 0.9739; ■, Binodal curve; ⋯, calculated by eq 2.

corresponding binary solutions (equal a_w). In these systems, because of the favorable polymer−salt interactions, the interaction of each solute with water becomes less favorable in the presence of the other solute. Therefore, the concentrations of solutes in a ternary solution are larger than those in the corresponding ternary semi-ideal solution and then the salting-in effects can be expected in the case of these systems.

However, for the ternary aqueous systems containing water-soluble polymers and electrolytes capable of inducing phase separation (salting-out effect), the solute−water interaction in the ternary solutions is stronger than those in the binary solutions and solute 1−water interaction becomes more stable in the presence of solute 2. In these systems, because of the unfavorable polymer−salt interactions, the interaction of each solute with water becomes more favorable in the presence of the other solute. Therefore, the concentrations of solutes in a ternary solution that is in isopiestic equilibrium with certain binary polymer + water and salt + water solutions are smaller than those we expect in the case of semi-ideal solutions and these systems show the negative deviation from eq 2. Salting-out inducing ions as well as polymer are favorably hydrated in aqueous polymer solution and interact with water molecules rather than with each other. The association of ions with polymer is a highly unfavorable process, so they exclude themselves from the vicinity of each other due to their preferential hydration. When the concentration of the solutes is increased, this exclusion will increase and ultimately, the system could reach a state where, for entropic reasons, phase formation would become favorable. Exclusion can also occur by repulsive interaction between the anions and the anionic-like polyether functionality of polymer. Similar behavior was obtained in the one-phase region of the ionic liquid−salt−water systems.[24] Similar to the polymer−salt interactions, formation of aqueous two-phase system involves partial dehydration of solutes and, therefore, the interaction of each solute with water becomes less favorable by phase separation. Therefore, as can be seen, in the two-phase region the positive deviation from eq 2 is observed.
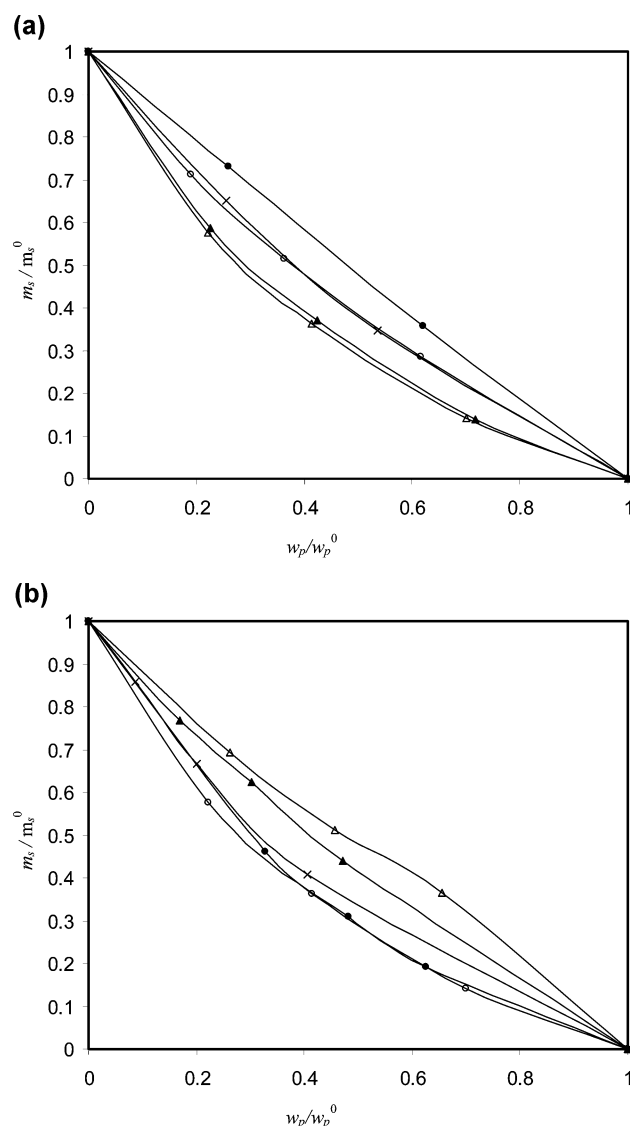
Water is the main component in these systems and dissolved components have high affinity for it. Therefore, water activity in these systems can give us useful information about the mechanisms of the salting effect produced by the addition of salting-out or sating-in inducing electrolytes to aqueous solutions of water-soluble polymers. The ethylene oxide units (in the case of PEG and PEGDME), propylene oxide units (in the case of PPG), and ionic species in aqueous solution are known to be hydrated. The higher valency of anions can be expected to be more effective than the lower valency of anions in the salting-out of the polyethers because of competition for water. In the case of cations, one can expect a competition between two opposing effects, namely, hydration and the tendency to complex with polyether oxygen.[10] If cations do interact with polymer, one can expect them to salt-in rather than salt-out the polymer. In the case of aqueous PEG or PEGDME + sodium salt solutions, two-phase formation occurs with anions sulfate, carbonate, and citrate, which have a marked salting-out effect on non-electrolytes, but that for large singly-charged anions such as chloride and nitrate has only one phase and the weak salting-out effects of these anions are offset by cations that are believed to coordinate to the ether oxygens to produce a salting-in effect. The side chain methyl groups in PPG hinder ion−dipolar interaction between cations and the ether oxygen atoms, and therefore, there is no salting-in effect in the case of aqueous PPG−salt solutions. Furthermore, PPG because of its higher hydrophobic characteristic can be more easily salted-out by a salting-out-inducing ion to form the polymer-based aqueous two-phase system than PEG and PEGDMEs.

For the PEG or PEGDME + chloride or nitrate salt + water solutions, the polymer−cation interaction is the predominant factor that leads to the positive deviation of constant water activity curves from eq 2. However, for the systems that form aqueous biphasic systems the exclusion of ions from the near surface region of the polymer in solution is the predominant factor that leads to the negative deviation of constant water activity curves from eq 2. As can be seen in Figure 6, for the systems that do not form aqueous biphasic systems, the positive deviation from eq 2 decreases by increasing the water activity or decreasing the polymer molecular weight and for a same water activity this deviation follows the order $Mg^{2+} > NH_4^+ > Na^+ > K^+ > (CH_3)_4N^+$. Positive deviations from eq 2 increase by increasing the charge density of cations or increasing polymer molar mass. This is because the polymer−cation interactions increase as the charge density of cations or polymer molar mass increases. On the other hand, Figure 7 shows that in the one-phase region of the systems that form aqueous biphasic systems, the negative deviation from the eq 2 decreases slightly with water activity and for the water activity this deviation follows the order PPG400 > PEGDME250 > PEG400 > PEGDME2000. In fact, the obtained results in this work and those reported in the literature[15] show that at the same water activity the negative deviation from eq 2 decreases as the polymer molar mass increases. Similar to the case of the liquid−liquid equilibria, the effect of anions on the vapor−liquid equilibria behavior of aqueous polymer−salt solutions are larger than those of cations and, as mentioned above, the salting-out-inducing anions are predominantly responsible for the observed effect, whereas the cations have a measurable, but smaller, effect on the salting-out strength. Figure 7 shows that the negative deviation from eq 2 for the investigated anions follows the order $NO_3^- < Cl^- < CO_3^{2-} < SO_4^{2-} \approx Cit^{3-}$. But there is no
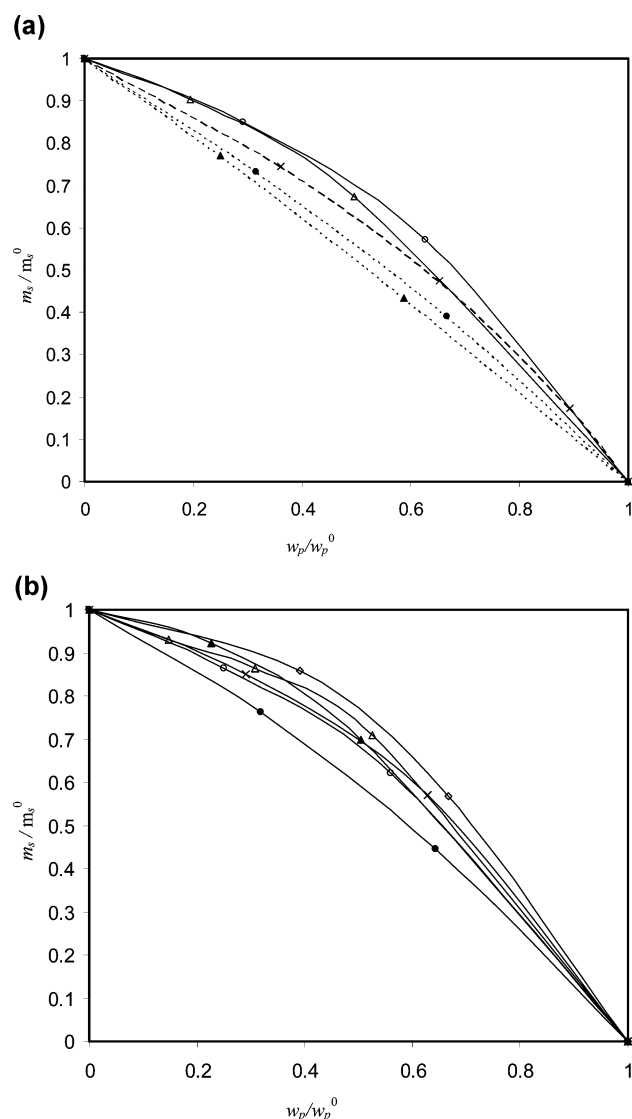
Figure 6. (a) Plot of m_s/m_s^0 against w_p/w_p^0 for constant water activity curves of PEGDME250 (dotted lines) + KCl + H_2O, PEGDME2000 (solid lines) + KCl + H_2O and PEG400 (dashed lines) + KCl + H_2O systems at 298.15 K: ▲ and △, $a_w = 0.9840$; ● and ○, $a_w = 0.9599$; ×, $a_w = 0.9575$. (b) Plot of m_s/m_s^0 against w_p/w_p^0 for constant water activity curves of PEGDME2000 + salt + H_2O system at 298.15 K: ●, $(CH_3)_4NCl$ ($a_w = 0.9602$); ○, NaNO$_3$ ($a_w = 0.9718$); ×, KCl ($a_w = 0.9599$); ▲, NaCl ($a_w = 0.9588$); △, NH$_4$Cl ($a_w = 0.9638$); ◇, MgCl$_2$ ($a_w = 0.9579$).

Figure 7. (a) Plot of m_s/m_s^0 against w_p/w_p^0 for constant water activity curves of polymer + Na$_2$SO$_4$ + H_2O system at 298.15 K: ●, PEGDME2000 ($a_w = 0.9727$); × , PEGDME250 ($a_w = 0.9727$); ○, PEG400 ($a_w = 0.9545$); ▲, PPG400 ($a_w = 0.9760$); △, PPG400 ($a_w = 0.9713$). (b) Plot of m_s/m_s^0 against w_p/w_p^0 for constant water activity curves of PPG400 + salt + H_2O system at 298.15 K: ●, Na$_3$Cit ($a_w = 0.9720$); ○, ($a_w = 0.9713$); ×, Na$_2$CO$_3$ ($a_w = 0.9717$); ▲, NaCl ($a_w = 0.9739$); △, NaNO$_3$ ($a_w = 0.9268$).

significant difference between the negative deviations from eq 2 for the investigated cations. The anions with higher charge density have a better ability to form ion/water complexes and also larger repulsive interaction with the ether oxygens of polymer (higher exclusion of ions from the polymer−water interface), and therefore, we can expect the negative deviations from eq 2 to increase by increasing the charge on the anion of electrolyte. As mentioned above, the side chain methyl groups in PPG hinder the PPG−cation interactions and, therefore, the salting-in effect of cations on PPG400 is smaller than those on PEG or PEGDMEs, which leads to a larger negative deviation of constant water activity curves from eq 2. Hydrophobicity increases with the increase of the molecular weight of the PEG or PEGDME polymer, and therefore, the exclusion of ions from the near surface region of the polymer in solution and thus the

negative deviation of constant water activity curves from eq 2 increases by decreasing the polymer molecular weight.

The isopiestic equilibrium in the two-phase region of these systems has three equilibrium phases: salt-rich liquid phase (bottom phase), polymer-rich liquid phase (bottom phase) and vapor phase. If we consider all three phases as one system, the positive deviation from eq 2 decreases as the water activity increases and for a same water activity this deviation for the investigated polymers follows the order PPG400 > PEGDME2000 > PEG400 > PEGDME250. At the same water activity, the effectiveness of the anions in the positive deviations from eq 2 in this region increase by increasing the charge on the anion of electrolyte.

■ CONCLUSIONS

To obtain further experimental evidence for the mechanisms of the salting effect produced by the addition of salting-out inducing electrolytes to aqueous solutions of water miscible polymers, vapor−liquid equilibria and liquid−liquid equilibria properties of aqueous solutions of PPG400, PEG400, PEGDME250, and PEGDME2000 were determined in the presence of large series of salts KCl, NH$_4$Cl, MgCl$_2$, (CH$_3$)$_4$NCl, NaCl, NaNO$_3$, Na$_2$CO$_3$, Na$_2$SO$_4$, and Na$_3$Cit. The results show that, aqueous solutions of PPG400 form aqueous two-phase systems with all the investigated salts; however, other investigated polymers form aqueous two-phase systems only with Na$_2$CO$_3$, Na$_2$SO$_4$, and Na$_3$Cit.

Detailed understanding of the molecular-level interactions obtained from the vapor−liquid equilibria behavior of the investigated systems allowed us to show that the phase behavior in aqueous polymer−salt two-phase systems is controlled by the net result of competition between the ability of the solutes to interact with water and the favorable (or nonfavorable) interactions that occur between the polymer and salt to a large extent. In the case of ternary aqueous polymer + salt solutions that do not form aqueous biphasic systems (salting-in effect), because of the favorable polyether oxygen−cation interactions, the interaction of each solute with water becomes less favorable in the presence of the other solute. Therefore, the concentrations of solutes in a ternary solution that is in isopiestic equilibrium with certain binary polymer + water and salt + water solutions are larger than those we expect in the case of semi-ideal solutions, and thus, these systems show the positive deviations from the linear isopiestic relation (Zdanovskii−Stokes−Robinson rule) derived using the semi-ideal hydration model and the salting-in effects can be expected in the case of these systems. However, for the ternary aqueous polymer + salt solutions that form aqueous biphasic systems (salting-out effect), because of the unfavorable polymer−salt interactions, the interaction of each solute with water becomes more favorable in the presence of the other solute. Therefore, the concentrations of solutes in a ternary solution that is in isopiestic equilibrium with certain binary polymer + water and salt + water solutions are smaller than those we expect in the case of semi-ideal solutions and these systems show the negative deviation from the linear isopiestic relation, and thus, a salting-out of the polymer by salt occurs and this leads to the entropically driven formation of aqueous biphasic systems above a certain critical concentration. Formation of aqueous two-phase systems involves partial dehydration of solutes, and therefore, the interaction of each solute with water becomes less favorable by phase separation. Therefore, in the two-phase region the positive deviations from the linear isopiestic relation are observed. The side chain methyl groups in PPG hinder the PPG−cation interactions, and therefore, the salting-in effect of cations on PPG400 is smaller than those on PEG or PEGDMEs, which leads to a larger negative deviation of constant water activity curves from the linear isopiestic relation.

■ ASSOCIATED CONTENT

ⓢ Supporting Information

Tables of isopiestic equilibrium concentrations, water activities, vapor pressures, and binodal data of the investigated polymer + salt + water systems examined in this study. This information is available free of charge via the Internet at http://pubs.acs.org

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Notes
The authors declare no competing financial interest.

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