

Nitroprusside–PEO Enthalpic Interaction as a Driving Force for Partitioning of the $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ Anion in Aqueous Two-Phase Systems Formed by Poly(ethylene oxide) and Sulfate Salts

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Ions are known to concentrate in the salt-enriched phase of aqueous two-phase systems, with the only known exception being the pertechnetate anion, TcO_4^- . We have discovered a second ion, nitroprusside anion (NP), which is markedly transferred from the salt phase to the polymer phase. The partitioning behavior of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ anion was investigated in ATPS formed by poly(ethylene oxide) of molar mass 3350 and 35000 g mol^{-1} , and different sulfate salts (Na_2SO_4 , Li_2SO_4 , and MgSO_4). On the basis of a model, the nitroprusside high affinity for the macromolecular phase was attributed to an enthalpic specific interaction between the anion and ethylene oxide unit. Partition coefficients increased exponentially with tie-line length increase, reaching values as high as 1000 and showing a relationship very dependent on the salt nature, but independent of the polymer molar mass.

Introduction

The physics controlling the interactions between ions and flexible chain macromolecules is reflected in such diverse phenomena as the complexation of polymer and micelles,¹ polymer–salt blends,² phase separation in systems containing polymer,³ and the partitioning and extraction of charge species (proteins, cations, anions) in aqueous two-phase systems (ATPS).^{4–6} In the last several years there has been a considerable effort toward improving the understanding of ion partitioning behavior in these biphasic systems.⁷

An ATPS consists of a mixture of aqueous solutions of two structurally different polymers (or a polymer and an electrolyte), which separate into two phases above a critical thermodynamic condition.⁸ In such biphasic systems one polymer is found predominately in one phase and the other polymer or salt in the other phase.

An ion added to the ATPS system will be distributed unevenly between the phases and this partitioning has been studied since the pioneer works of Zvarova et al.⁹ Since then some investigations have studied the extraction of different ions in aqueous two-phase systems formed mainly by poly(ethylene oxide) (PEO) and different electrolytes.¹⁰ In general, without the use of any extractant or complexing agent, the majority of ions spontaneously concentrate in the salt-rich phase, the only exception being the anion pertechnetate (TcO_4^-) with a partition coefficient as high as 610 in PEO/sulfate and PEO/carbonate aqueous two-phase systems.¹¹ The partitioning of any solute, including ions, in ATPS is dependent on various variables such as the chemical nature of the components, charge and size of polymer(s) and of salts, tie-line length, temperature, pH, and addition of neutral and charged species.¹²

To increase the efficiency of ATPS to ion or any other charged solute separations, we need to better understand the driving force(s) governing the partition behavior of these chemical species.

Our aim with the present work is to identify the driving forces associated with the partitioning of pentacyanonitrosylferrate(II) anion, $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$, also known as nitroprusside (NP), in ATPS formed by PEO and different sulfate salts. This ion was chosen because our group discovered that it transfers preferentially to the PEO-rich phase and makes a specific interaction with the ethylene oxide unit, which could be followed by infrared spectroscopy measurement. We have investigated the partitioning behavior of nitroprusside anion in ATPS as a function of molar mass of the PEO, electrolyte nature, and tie-line length. The results obtained have been discussed on the basis of a fundamental model, derived by Haynes et al.,¹³ and based on the Flory–Huggins mean field theory for polymer solutions, which describes the dependence of solute partitioning on system variables and configuration.

Experimental Section

Reagents. Poly(ethylene oxide) samples, with average molar masses (according to the manufacturer) of 3350 (PEO3350) and 35000 g mol^{-1} (PEO35000), were supplied by Sigma Chem. Co. (USA). Na_2SO_4 , Li_2SO_4 , MgSO_4 , $\text{Na}_2[\text{Fe}(\text{CN})_5\text{NO}]$, and $\text{Na}_3[\text{Fe}(\text{CN})_6]$ were purchased from Merck (Darmstadt, Germany). All the chemicals were of analytical grade; deionized water ($R \geq 18 \text{ M}\Omega \text{ cm}^{-1}$) was used throughout all the experiments.

Determination of Phase Diagrams. Ternary phase diagrams of the PEO/ $\text{M}_2\text{SO}_4/\text{H}_2\text{O}$ ($\text{M} = \text{Li}^+$, Na^+ , or Mg^{2+}) system were determined at 25.0 °C with use of a water bath (Microquímica, model MQBTC99-20, Brazil) with an accuracy of ± 0.1 °C. The prepared samples were left for 24 h to phase separate, but approximately 0.5 h was often enough to get two macroscopic

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well-defined phases. The polymer concentrations in the separate phases were determined in two steps. First, after separating the two phases from each other and after suitable dilution, the sulfate salt concentration in both phases was determined by the classic gravimetric method with BaCl_2 as precipitant agent.¹⁴ The precipitate mass measurements were performed with an analytical balance (Gehaka, AG200, Brazil). After the concentration of the poly(ethylene oxide) in the phases was suitable dilution then determined by measuring the total refractive index of the solution and then subtracting the refractive index contribution due to sulfate salt at this concentration. To do this, standard curves were established for aqueous solutions of PEO3350, PEO35000, Li_2SO_4 , Na_2SO_4 , and MgSO_4 . The validity of refractive index additivity was also confirmed by using test samples. A 09-2001 model Analytic Jena AG Abbe refractometer (Germany) was used for the refractive index measurements.

Partitioning Experiments. Aqueous two-phase systems were prepared in 40 mL graduated centrifuge tubes by weighing the 50% (w/w) PEO solution and a stock solution of sulfate salt (Na_2SO_4 , 20% (w/w); Li_2SO_4 , 28% (w/w); MgSO_4 , 50% (w/w)) and mixing it to obtain a desired total composition. Aluminum foil sheets protected all tubes to avoid the photodecomposition of the inorganic complex. Approximately 40.0 mg of $\text{Na}_2[\text{Fe}(\text{CN})_5\text{NO}]$ was added to the system, which was made up to 10.0 g by the addition of water. The systems were mixed for 60 s and then brought to equilibrium in a thermostatic bath for 48 h, since earlier tests indicated that this period of time was enough to ensure thermodynamic equilibrium. After this time, the two phases became reddish with a well-defined interface. After equilibrium both phases were sampled with syringes, with care to leave a layer of phase at least 2.0 cm distant of the interface. The bottom phase was withdrawn by using a syringe with a long needle. After dilution, the anion content in each phase was determined by measuring the absorbance at 260 nm. Correspondingly diluted phases from the aqueous two-phase system, not containing the complexes, were used as blanks. The partition coefficient was defined as

$$K = \frac{[\text{NP}]_{\text{top}}}{[\text{NP}]_{\text{bottom}}} \quad (1)$$

where $[\text{NP}]_{\text{top}}$ and $[\text{NP}]_{\text{bottom}}$ are equilibrium concentrations of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ in the PEO and sulfates salt-rich phases, respectively. In all cases the concentrations were analyzed in triplicate. For the partition coefficients the relative standard deviation was $\pm 2.0\%$.

For all samples, the nitroprusside stability after the partitioning experiments was confirmed by recording infrared and UV–visible spectra from the upper and bottom phases. The nitroprusside molar absorptivity (ϵ) was determined in pure water, PEO solutions (5.0%, 10%, and 20% (w/w)), and salt solution (1.0%, 5.0%, and 10% (w/w)), and in all systems the same value was obtained ($\epsilon = 2.2 \times 10^3 \text{ L mol}^{-1} \text{ cm}^{-1}$).

Infrared Measurements. FTIR spectra of all samples (aqueous solutions) with a resolution of 2.0 cm^{-1} were taken on a Perkin-Elmer 1000 FTIR spectrometer with a DTGS (deuterium-triglycine sulfate) detector, Beaconsfield, England. All spectra were recorded at $25.0 \text{ }^\circ\text{C}$. To obtain a good quality of the spectra, 64 scans were accumulated and a cell with CaF_2 windows has been used. The concentration of nitroprusside in each solution has been over 1.0% (w/w).

Calorimetric Measurements. Solution enthalpies $\Delta_{\text{sol}}H$ of $\text{Na}_2[\text{Fe}(\text{CN})_5\text{NO}]$ were measured in a Thermometric 2225

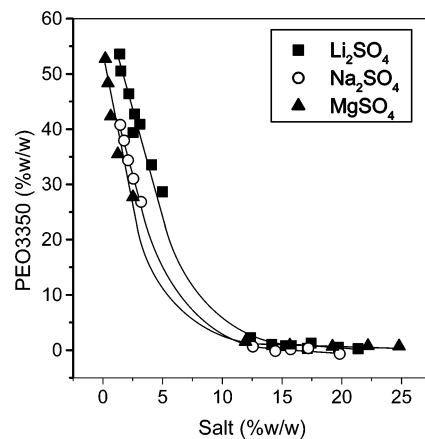


Figure 1. Phase diagram for the PEO3350– M_xSO_4 ATPS at $25 \text{ }^\circ\text{C}$.

Precision Solution calorimeter by the ampule breaking technique. It is a semiadiabatic macrocalorimeter with a 100 mL glass reaction vessel. This unit was inserted into a thermostatic water bath operating at 298 K , with a stability of 0.0001 deg . Glass sample ampules of cylindrical shape and with thin end walls were charged with 300–400 mg of sodium pentacyanonitrosylferrate and the thin-walled necks were sealed under flow flame. The measured enthalpy change was done by breaking the ampule in the top or bottom phase of the ATPS. With the amounts of substance used, the minimum energy measured in the experiment was 3 J, well above the energy exchanged by breaking an empty ampule. The accuracy of the equipment and of the procedure was confirmed by measuring solution enthalpies of KCl, with agreement better than 1% with respect to literature data. All of the reported enthalpies are averages of the two independent experiments.

The enthalpy of transfer ($\Delta_{\text{tr}}H$) of the anion between the bottom and top phases, obtained by calorimetric measurements, was defined as

$$\Delta_{\text{tr}}H = \Delta_{\text{sol}}H^{\text{top}} - \Delta_{\text{sol}}H^{\text{bottom}} \quad (2)$$

Results and Discussion

Phase Diagrams. The partitioning of ions between two aqueous phases and the equilibrium distributions of the ATPS components can be understood in terms of molecular interactions in the mixture. At constant pressure and temperature, these different kinds of interactions are functions of the concentrations of polymers, water, and salt in the aqueous two-phase systems, generally expressed in its phase diagrams.

We have determined phase diagrams for PEO/ M_xSO_4 / H_2O systems using different cations: Na^+ , Li^+ , or Mg^{2+} and PEO of distinct molar mass (3350, 35000 g mol^{-1}). Additionally attempts were made to establish a relationship between the tie-line length and the partitioning behavior of Nitroprusside anion in these ATPS. Furthermore, we have not found in the literature phase diagrams for the systems formed by PEO3350 or PEO35000 with different sulfate salts. Figure 1 shows the influence of the electrolyte type on the phase diagram for the aqueous two-phase systems formed with PEO3350.

The formation of these aqueous two-phase systems clearly indicates the mutual exclusion of the salt and polymer and their high affinity for water. This is a general behavior in ATPS formed by sulfate salts and poly(ethylene oxide).^{15–18} The efficacy of the three salts in inducing the formation of aqueous two-phase systems with PEO is different and follows the order $\text{MgSO}_4 > \text{Na}_2\text{SO}_4 > \text{Li}_2\text{SO}_4$. da Silva and Loh,¹⁹ based on

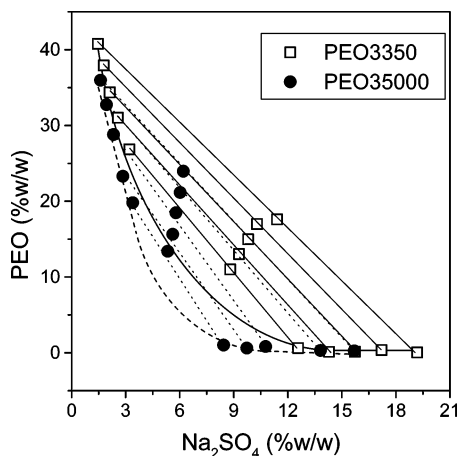


Figure 2. Phase diagram for the PEO–Na₂SO₄ ATPS at 25 °C.

calorimetric measurements, attribute the trend in efficacy of sodium and lithium sulfates in inducing ATPS formation to cation–polymer interactions. Their proposed model is that, when PEO and sulfate salts are mixed, the cations and the polymer interact, releasing some water molecules that were solvating them in a process that is driven by the entropy increase. This cation binding continues as more electrolytes are added, until a saturation point, after which no more entropy gain may be attained and phase splitting becomes more favorable. Therefore, the picture that arises from this proposed model is that the polymer-rich phase contains macromolecules bound to cations, forming a pseudopolycation, which is capable of interacting with negatively charged species.

The influence of the poly(ethylene oxide) molar mass on phase equilibrium is shown in Figure 2. The size of the macromolecule has an effect on the composition of the two phases, where with an increase in the molar mass an increase in the two-phase region is observed. The influence of molar mass is commonly verified in ATPS systems and has been attributed to a decrease of the configurational entropic contribution caused by enlargement of the polymer.^{20,21}

On the basis of our results and on data from the literature, we can affirm that in some range of temperature, pressure, and compositions, poly(ethylene oxide) molecules could show a strong segregation from some cations and anions.

Nitroprusside Anion Partitioning. The nitroprusside anion was partitioned at 25.0 °C in six distinct PEO/sulfate/H₂O systems. The three salts Na₂SO₄, Li₂SO₄, and MgSO₄ were used and two different PEO (3350 and 35000 g mol⁻¹). In partitioning studies with ATPS it is conventional to express the difference in the intensive thermodynamic properties of the two phases by the tie-line length (TLL) parameter, which is determined by the difference in concentration of the systems forming components. TLL, expressed in weight percentage of polymers and salts, is calculated according to

$$\text{TLL} = [(C_P^T - C_P^B)^2 + (C_S^T - C_S^B)^2]^{1/2} \quad (3)$$

where C_P and C_S are the polymer and salt concentrations in % (w/w), and T and B are the top and bottom phases, respectively.

The phase incompatibility and consequently the uneven solute partitioning increase with increase of the tie-line length.⁴ For each ATPS system, the partition coefficient of the [Fe(CN)₅NO]²⁻ anion, K , was determined in five different tie-line lengths. Partition coefficients of the nitroprusside ion determined in ATPS composed of PEO35000 and the different sulfate salts are shown in Figure 3.

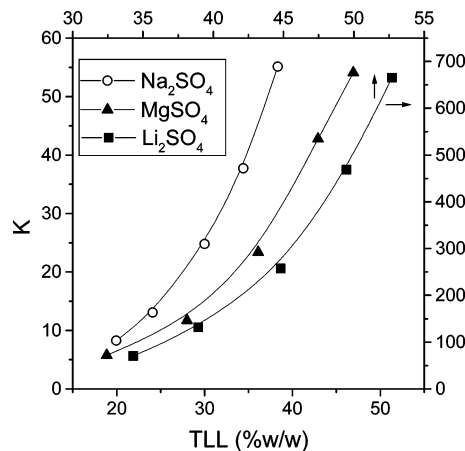


Figure 3. Partitioning coefficient of [Fe(CN)₅NO]²⁻ plotted against the tie-line length in the PEO35000–M_iSO₄ ATPS at 25 °C.

The [Fe(CN)₅NO]²⁻ anion prefers partitioning to the top phase, and the increase in tie-line length causes an exponential increase in partitioning coefficient, reaching experimental values as high as 1000, indicating a great potential for separation of nitroprusside anion from other ions. This result is very important because nitroprusside is a strategic material with applications in molecular communication,²² storage information devices,²³ and important medical applications.²⁴ Hence, there are current needs for the discovery of separation technologies capable of obtaining pure nitroprusside salts. Furthermore, the linear relationship between $\ln K$ and TLL provides a convenient and easy probe of the system composition. To improve this separation method we have investigated in detail the nature of the interaction of the [Fe(CN)₅NO]²⁻ anion with PEO-based ATPS.

As clearly observed in Figure 3, there is a strong influence of the electrolyte nature in the partitioning coefficient values. Nitroprusside anion transfers from the bottom phase to the top phase, both with PEO3350 and PEO35000, following the sequence Li₂SO₄ ≫ Na₂SO₄ > MgSO₄. These trends in [Fe(CN)₅NO]²⁻ distribution ratios do not follow the salting-out ability of the sulfate salts observed in the phase diagrams. Na₂SO₄ is more effective to induce phase separation than Li₂SO₄, but the lithium sulfate salt is much more efficient in pushing nitroprusside to the polymer-rich phase. It is well-known that a strong interaction exists between the lithium salts and PEO macromolecules, as indicated by NMR measurements,²⁵ electrochemical measurements,²⁶ and molecular simulations.²⁷ Hence, the larger K values observed in the Li₂SO₄/PEO/H₂O ATPS could be attributed to the existence of great positive charge densities in the PEO macromolecule, caused by the Li–EO interaction, and consequently an electrostatic interaction between [Fe(CN)₅NO]²⁻ and the pseudopolycation formed can occur.

As already stated, most of the ions in aqueous two-phase systems prefer to concentrate in the salt-rich phase, necessitating the use of some kind of extractant to transfer them to the polymer-rich phase.²⁸ At present, to the best of our knowledge, the only ion that has a large partitioning coefficient toward the polymer-rich phase is the pertechnetate anion, TcO₄⁻. Rogers' group^{11,29} has shown that pertechnetate anion can be partitioned from simulate nuclear wastes almost quantitatively to the PEO-rich phase, from a variety of salt solutions including OH⁻, CO₃²⁻, SO₄²⁻, and PO₄³⁻, in the absence of any other extractant. Contrary to the [Fe(CN)₅NO]²⁻ partitioning, $K_{TcO_4^-}$ values follow the order of the sulfate salting-out efficiency,³⁰ indicating probably some difference in its driving partitioning force.

TABLE 1: Difference between H_2O Concentration in the Top and Bottom Phases of the ATPS^a

TLL	$\Delta[\text{H}_2\text{O}] = [\text{H}_2\text{O}]_{\text{bottom phase}} - [\text{H}_2\text{O}]_{\text{top phase}} (\% \text{ w/w})$					
	Na_2SO_4 PEO3350	Na_2SO_4 PEO35000	Li_2SO_4 PEO3350	Li_2SO_4 PEO35000	MgSO_4 PEO3350	MgSO_4 PEO35000
1	16.85	13.75	22.47	23.64	16.73	12.20
2	19.88	15.77	27.95	25.60	20.16	17.66
3	20.58	19.51	29.89	28.64	23.10	20.75
4	22.15	20.56	31.80	30.92	25.81	23.09
5	22.98	21.62	33.37	31.85	27.43	25.07

^a Temperature: 25 °C.

However, similarly to nitroprusside behavior, they have observed a linear relationship for $\log K_{\text{TCO}^+}$ versus either weight percent salts or weight percent of polymers. Those authors attributed pertechnetate anion partitioning behavior to it preferring the hydration environment afforded by the PEG-rich phase rather than the highly ordered water structure in the salt-rich phase. However, recent studies suggested that the addition of Na_2SO_4 or PEO has no effect on the rotational dynamics of H_2O molecules outside their first hydration shell.³¹ If this is the case, the polymer and the salt-rich phases would have almost the same bulk hydration environment.

To explain the nitroprusside partition behavior, an approach using thermodynamics and Flory–Huggins theories can be applied to describe the transference process in terms of molecular interactions. In the Haynes model,¹³ a simple mean-field approach, the partitioning behavior of solutes in ATPS is interpreted in terms of enthalpic and entropic contributions, both written explicitly in simple analytical equations. The entropic contribution to partition coefficient is obtained by

$$\ln K = \frac{M}{\rho} \left(\frac{n^{\text{T}}}{V^{\text{T}}} - \frac{n^{\text{B}}}{V^{\text{B}}} \right) \quad (4)$$

where M is the molar mass of partitioning solute, n^{T} and n^{B} are the total number of molecules in the top and bottom phase, respectively, which produce the phase number density when divided by the phase volume, V^{T} and V^{B} , and ρ is the number of lattice sites per unit volume.

Basically, eq 4 shows that the transfer entropy change will make the solute partition to the phase with a large number of molecules per volume unit. In the PEO–sulfate ATPS systems the number density of the PEO-rich phase is smaller than that of the salt-rich phase mainly due to the lower water content of the top phase. Table 1 shows the difference in water concentration between the bottom and top phases in each ATPS used for $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ partitioning.

On the basis of eq 4 and Table 1, but contrary to experimental data, the entropic contribution to the nitroprusside anion distribution would favor its transfer to the salt-rich phase, increasing this entropic effect with the tie-line length increase. Consequently the cause of the concentration change of the anion in the polymer-rich phase could only be due to an enthalpic contribution, which is expressed, in the Haynes model, by

$$\ln K = - \frac{M_S}{RT} \left[\sum_{i=1(i \neq S)}^m (\Phi_i^{\text{T}} - \Phi_i^{\text{B}}) w_{\text{NP}-i} - \sum_{i=1}^2 \sum_{j=2}^3 (\Phi_i^{\text{T}} \Phi_j^{\text{T}} - \Phi_i^{\text{B}} \Phi_j^{\text{B}}) w_{i-j} \right] \quad (5)$$

where Φ_i^{T} and Φ_i^{B} are the volume fraction of the ATPS forming compounds on the top phase and on the bottom phase, respectively, w_{i-j} is the effective pairwise interchange energy

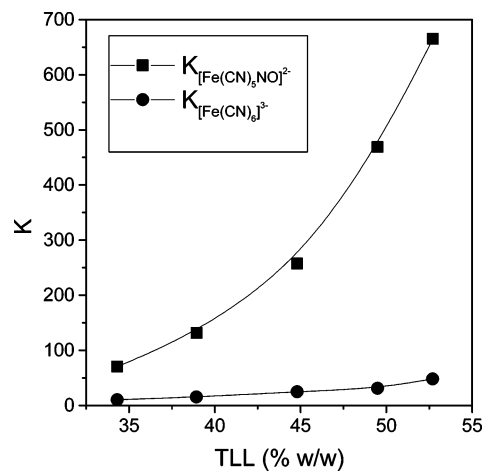


Figure 4. Partitioning coefficient of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ and $[\text{Fe}(\text{CN})_6]^{3-}$ plotted against the tie-line length in a ATPS formed by PEO35000– Li_2SO_4 at 25 °C.

defined as $w_{i-j} = z[\epsilon_{ij} - (1/2)(\epsilon_{ii} + \epsilon_{jj})]$, where z is the number of nearest neighbors and ϵ_{ij} is the potential energy of an $i-j$ pair.

The second term on the right-hand side of eq 5 represents the contributions for solute transfer enthalpy originating from unlike molecular enthalpic interactions between all forming ATPS phase components, without taking into account the interactions between nitroprusside anion and these components. This energetic term, called difference in the self-phase energy, is proportional to the molar volume of the solute.³² Solute transfer from the bottom to the top phase promotes the closing of a cavity in the higher density phase, causing interaction between forming components of the bottom phase and at the same time requires the breaking of interactions among top phase components to create a cavity for its insertion. To assess the contribution of self-energy of each phase to transfer of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$, another anion complex, hexacyanoferrate, $[\text{Fe}(\text{CN})_6]^{3-}$, with the same molar volume, was partitioned. Figure 4 shows the partitioning coefficient for both anions in ATPS formed by PEO35000 and Li_2SO_4 .

Hexacyanoferrate anion transfers preferentially to the top phase; however, this trend is smaller than that for $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$, demonstrating that the demand of energy associated with cavity formation is not the only contribution factor to the transfer of these ions, making it necessary to analyze contributions from their molecular interactions with all ATPS components. The same trends were obtained for aqueous two-phase systems with other smaller cations and PEO, as shown in Table 2.

From the above discussion, nitroprusside transfer must be related to a specific enthalpic interaction between the anion and PEO molecules, as represented by the first term in eq 5. The basic concept of this term is that $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ will partition to the phase enriched in that component with more energetic interaction with this anion, i.e., with $w_{\text{NP}-i}$ more negative (or

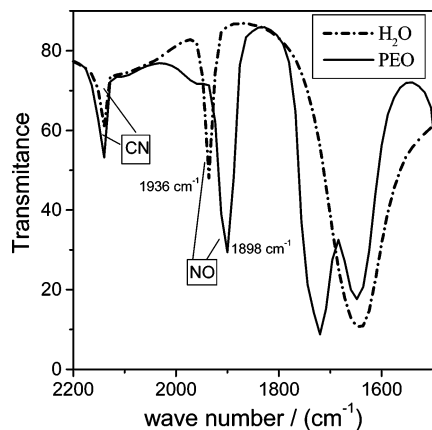
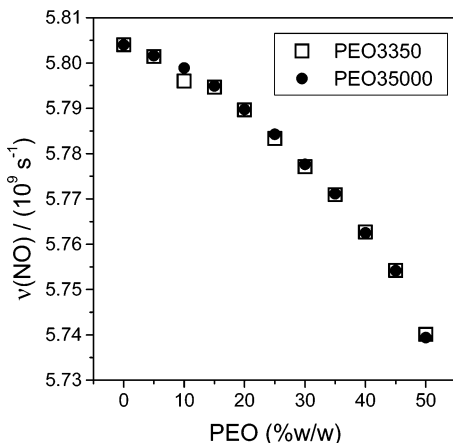
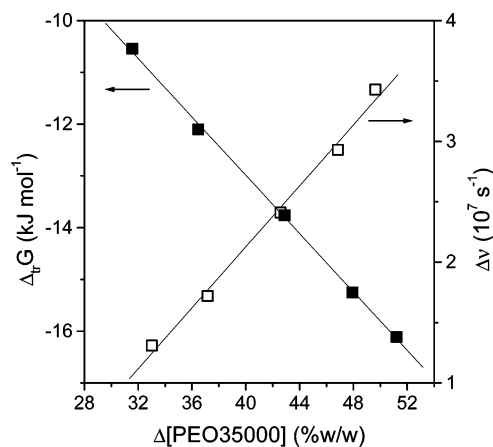
TABLE 2: $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ and $[\text{Fe}(\text{CN})_6]^{3-}$ Partition Coefficients in ATPS Formed by PEO35000/ Na_2SO_4 and PEO35000/ MgSO_4

PEO35000/ Na_2SO_4			PEO35000/ MgSO_4		
TLL (% w/w)	$K_{\text{Fe}(\text{CN})_6^{3-}}$	K_{NP}	TLL (% w/w)	$K_{\text{Fe}(\text{CN})_6^{3-}}$	K_{NP}
19.95	3.16	8.25	18.87	2.33	5.74
24.10	4.44	13.07	28.01	3.43	11.73
30.00	5.51	24.78	36.11	4.75	23.36
34.42	7.39	37.71	42.92	6.30	42.78
38.34	9.71	55.11	46.95	7.33	54.07

less positive). The main difference between both ions, which could lead to different partitioning behavior, is the presence of the NO group, assuming that the charge difference does not influence so much the partitioning behavior of ions in ATPS.³³

The existence of a direct interaction causing a favorable enthalpy of transfer is supported by infrared spectroscopy measurements of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ dissolved in water and in PEO aqueous solutions (Figure 5). The NO stretching band is very sensitive to PEO concentration, as shown in Figure 6, while the other normal-mode frequency values remained constant.

When the sodium nitroprusside is dissolved in pure water this NO wavenumber is observed at 1936 cm^{-1} . However this value decreases when the PEO concentration increases, reaching a limit value of 1898 cm^{-1} in pure liquid PEO. There is no dependence of the NO stretching frequency in relation to the size of PEO, indicating a site-specific interaction caused by an increased local concentration of the anion in the vicinity of the polymer. This increased local anion concentration must be promoted by the adsorption of the cation onto the PEO chain.

**Figure 5.** Change in NO stretching band when $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ is dissolved in water and in PEO 400.**Figure 6.** Effect of the PEO concentration on the NO stretching frequency. Temperature: $25\text{ }^\circ\text{C}$.**Figure 7.** Δ_rG and $\Delta\nu$ as a function of the difference in PEO concentration between the top and the bottom phase for ATPS formed by PEO35000– Li_2SO_4 at $25\text{ }^\circ\text{C}$.

The NO frequency shift could be explained considering the diamagnetic character of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$, usually described as a low-spin $\text{Fe}^{\text{II}}\text{NO}^+$ species, and its preferential solvation. So in PEO aqueous solutions, water molecules and EO segments will form the solvation shell of the anion, with a radial distribution that will depend on the polymer concentration. The acceptor–donor interaction between the ion species (mainly at the NO^+ site) and its solvation molecules (due to the electron lone pair present in the oxygen atom) will change the NO electron density, altering the force constants of the NO bond. A parameter that could express the PEO– $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ specific interaction is $\Delta\nu$, defined by eq 6. Naturally $\Delta\nu$ will be dependent on PEO concentration:

$$\Delta\nu = (\nu_{\text{H}_2\text{O}}^{\text{NO}} - \nu_{\text{PEO/solution}}^{\text{NO}}) \quad (6)$$

where $\nu_{\text{H}_2\text{O}}^{\text{NO}}$ and $\nu_{\text{PEO/solution}}^{\text{NO}}$ are the frequency of the NO stretching in pure water and in PEO aqueous solutions, respectively.

To support the specific NP–PEO interaction as a contribution to the anion transfer, we compared the change in Gibbs energy caused by the nitroprusside transfer, i.e., $\Delta_rG = -RT \ln K$, and the $\Delta\nu$, a parameter directly related to the NP–PEO interaction. The two variables were analyzed in dependence on the PEO concentration difference between the top and the bottom phase, $\Delta[\text{PEO}]$. Figure 7 shows an increasing ($\Delta\nu \times \Delta[\text{PEO}]$) and a decreasing ($\Delta_rG \times \Delta[\text{PEO}]$) linear relationship for the ATPS formed by PEO35000/ Li_2SO_4 / H_2O . It is clear from both curves that the increase in PEO concentration on the top phase and, consequently, the increase in NP–PEO interaction (increase in $\Delta\nu$) cause a proportional decrease in the free energy of transfer. The same behavior was verified for other ATPS, as shown in Figure 8.

As Δ_rG values are related to all molecular interactions pushing the $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ anion to the PEO-rich phase and $\Delta\nu$ is only due to the NP–PEO interaction, we can conclude that polymer–anion interaction should be the main driving force for the increased nitroprusside concentration in the top phase. Moreover, the linear relationship between Δ_rG and $[\text{PEO}]$ aids, to a certain extent, the model of a specific site interaction between the EO unit and the $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ anion.

As discussed before, our conclusion about an enthalpic motriz power for the transference process of the nitroprusside anion was based on the Haynes' model.¹³ To confirm this conclusion, calorimetric measurements were carried out. Enthalpies of solutions of $\text{Na}_2[\text{Fe}(\text{CN})_5\text{NO}]$ in the bottom and the top phase

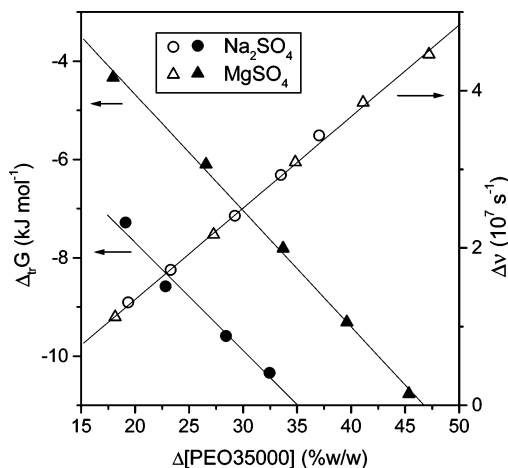


Figure 8. $\Delta_{tr}G$ and $\Delta\nu$ as a function of the difference in PEO concentration between the top and the bottom phase for ATPS formed by PEO35000– $M_x\text{SO}_4$ at 25 °C.

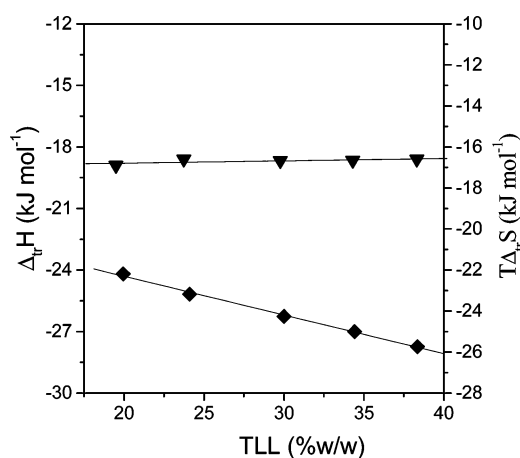


Figure 9. $\Delta_{tr}H$ and $T\Delta_{tr}S$ as a function of TLL for ATPS formed by PEO35000– Na_2SO_4 at 25 °C.

of the PEO35000/ Na_2SO_4 were determined at 298 K. With these data it was possible to calculate the nitroprusside enthalpies of transfer, $\Delta_{tr}H$, by subtracting its enthalpy of solution in the top phase from its enthalpy of solution in the bottom phase. Therefore, $\Delta_{tr}H$ represents the enthalpic energy balance between the solvation of the sodium nitroprusside in the bottom and the top phases and would reflect any change occurring in molecular interactions during the partitioning processes. Using the classical thermodynamic equation, $\Delta_{tr}G = \Delta_{tr}H - T\Delta_{tr}S$, it was possible to calculate the transfer entropies of sodium nitroprusside.

The enthalpies and entropies of transfer for $\text{Na}_2[\text{Fe}(\text{CN})_5\text{NO}]$ from the bottom phase to the top phase are shown in Figure 9. For all TLL values, the process of transference was directed by enthalpic contributions, and it occurs with a decrease in the entropies of the systems, which is in line with Haynes' model¹³ propositions.

The Influence of Poly(ethylene oxide) Molar Mass. In general, the partitioning behavior of any solute in ATPS depends on the molar mass of polymeric components.³⁴ If the molar mass of one of the phase polymers is increased the partition of solute into the phase in which that polymer predominates decreases. The main cause of this feature of the partition behavior is that the increase in macromolecular size decreases the contribution of configurational entropy to minimize the solute transfer Gibbs energy.³⁵ As pointed out, the nitroprusside transfer is mainly enthalpically driven and, consequently, the influence of entropy

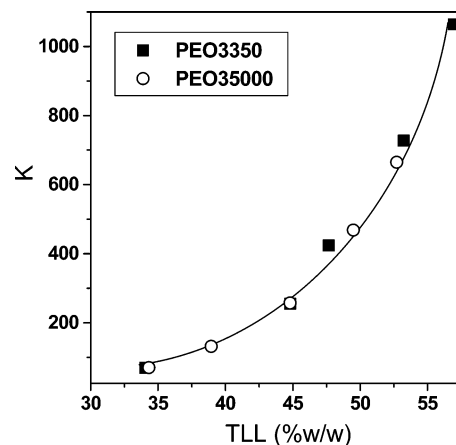


Figure 10. Partitioning coefficient of $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ plotted against the tie-line length in the PEO– Li_2SO_4 ATPS at 25 °C.

change, caused by any molecular process, is expected not to be dominant.

The results of nitroprusside partitioning in the PEO3350/ Li_2SO_4 and PEO35000/ Li_2SO_4 are shown in Figure 10, where K is plotted against TLL.

The relationship of K values in relation to TLL is independent of the PEO molar mass in agreement with a process governed by specific enthalpic interaction between EO segments and nitroprusside anions. Similar behavior was obtained with sodium sulfate and magnesium sulfate as the salt-forming phase (not shown). Different from the nitroprusside anion, the TcO_4^- transfer process shows polymer size dependence.³⁶ ATPS containing higher PEO molar masses, which are salted out with less salt, exhibit larger $K_{\text{TcO}_4^-}$ values. It must be stressed that, these experiments studying the dependence of pertechnetate partitioning coefficients in relationship to polymer size were carried out in ATPS with same polymer global composition but not in the same tie-line length. So it is difficult to separate the influence of molar mass from the influence of tie-line length differences.

Conclusion

The present investigation revealed a new ion, $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$, which partitions preferentially to the polymer-rich phase in ATPS formed by PEO/ $M_x\text{SO}_4/\text{H}_2\text{O}$ ($M = \text{Na}^+, \text{Li}^+, \text{Mg}^{2+}$). As the nitroprusside salts are strategic compounds, mainly due their applications, knowledge of new purification methods is very important. Furthermore, the partitioning behavior of this anion furnishes important insights on the major factors governing ions partitioning in the PEO/electrolyte ATPS. On the basis of our results and literature data, there should be a specific enthalpic interaction between the ion and the macromolecules, to concentrate the cation or anion in the polymer-rich phase. In the absence of this enthalpic contribution, the entropic force will predominate transferring the ions to the salt-rich bottom phase.

The enthalpic interaction between nitroprusside and poly(ethylene oxide) macromolecules probably occurs between the $[\text{Fe}(\text{CN})_5\text{NO}]^{2-}$ and ethylene oxide units and is very dependent on the nature of the electrolyte. On the other hand, the macromolecule size does not influence significantly the nitroprusside partitioning, corroborating the assumption of a small entropic contribution. In addition, calorimetric results confirm the insights related to enthalpic specific contributions that determine the nitroprusside partitioning in ATPS systems.

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