



Salt Effect on the Aqueous Two-Phase System PEG 8000 - Sodium Sulfate: Physico-Chemical Characterization of the Systems

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In recent studies there is one issue that seems to be one of the most interesting in regard to partitioning of biomolecules, from both theoretical and practical viewpoints - the effects of salt additives on partition [1]. Zaslavsky [2] studied the effect of several salts on polymer-polymer ATPSs and stressed on the role of "water structure" as an important factor controlling two-phase formation. In polymer-salt ATPSs, this issue has never been examined systematically. The main goal of this work is to study the effects of different salt additives (NaCl or KCl), with concentration up to 1.0 M, on PEG 8000 - Na₂SO₄ ATPS, containing 0.01 M of sodium phosphate buffer, pH 7.4, at 296.15 K. Phase diagrams determined by the cloud point method, including tie-lines assigned from mass phase ratios according to the lever arm rule, are presented. The results indicate that the salting-out ability of the cations follows the Hofmeister series (Na⁺>K⁺) and can be related to the ions Gibbs free energy of hydration (ΔG_{hvd}).

The Gibbs free energy of transfer of a methylene group between the coexisting phases, $\Delta G(CH_2)$, was been used to characterize the difference between the hydrophobic character of the equilibrium phases of those particular ATPSs. The $\Delta G(CH_2)$ was determined by partitioning of a homologous series of five sodium salts of dinitrophenylated - amino acids with aliphatic side chains in different tie-lines of each biphasic system. The results show that, within each system, there is a linear relationship between the $\Delta G(CH_2)$ and the tie-line length.

References

- [1] Azevedo AM, Rosa PAJ, Ferreira IF, Aires-Barros MR, "Integrated process for the purification of antibodies combining aqueous two-phase extraction, hydrophobic interaction chromatography and size-exclusion chromatography", *J. Chromatogr. A* (2007) **1213**: 154-161.
- [2] Zaslavsky BY, "Aqueous two-phase partitioning: physical chemistry and bioanalytical applications", Marcel Dekker, New York, 1994.