

Phase Equilibria Determinations for Separation Processes Involving Natural Products

KEYWORDS: Ionic liquids; Ethyl lactate; CAAILS; Green solvents; Aqueous Two-Phase Systems; Liquid-Liquid Equilibria.

The liquid-liquid equilibria (LLE), *i.e.*, binodal curves and tie-line compositions, of Aqueous Two-Phase Systems (ATPS) of the types ethyl lactate / salt, ionic liquid / salt or polymer / salt were determined at 298.15 K and 0.1 MPa. These systems provide biodegradable, non-toxic and mild extractive conditions for the extraction of natural products, such as biomolecules (e.g., pigments, proteins, fibres) and active pharmaceutical ingredients (e.g., antibiotics, anti-inflammatories). The obtained data were described with common empirical correlations and modelled using excess Gibbs free energy (G^E) models and equations of state (EoS).

Introduction

The most common feedstocks for natural vitamins and antioxidants, *i.e.*, chemicals which delay the oxidative damage by reactive oxidative species, are fruits and vegetables, but using food as raw material raises moral issues and contributes to several environmental and social problems, such as larger farming areas and global hunger. Therefore, the use of biowaste is a more feasible option for the production of dietary supplements and cosmetics, contributing simultaneously to a more circular economy and to more sustainable therapeutics.

Aqueous Two-Phase Systems (ATPS), or Aqueous Biphasic Systems (ABS), constitute a popular liquid-liquid extraction technique for the purification of biomolecules, pharmaceuticals and metals. They are well-known for their high water-content, low interfacial stress and high capacity. The most applied ATPS are composed of polymer + polymer, polymer + salt or organic solvent + salt and water systems. Generally, organic salts are preferred to the inorganic given the more sustainable production of the former, but some types of ionic liquids do not present immiscible liquid zones with organic salts.

Ionic liquids, or molten salts, are salts with melting points below 373.15 K (at 0.1 MPa) and are being increasingly applied in ATPS due to their low volatility and high solvent capacity. However, the eco-friendliness of some ionic liquids has been questioned, for which novel ionic liquids had to be developed based on biomaterials. The most popular green ionic liquids are the ones based on the cholinium cation and on amino acids (choline-amino acid ionic liquids, CAAILS), which present increased biodegradability and non-toxicity.

On the other hand, one of the most used organic solvents in ATPS is ethyl lactate, which is a bio-renewable and biodegradable solvent with low toxicity towards humans and animals. Ethyl lactate can be produced from biomass fermentation (e.g., corn residues) by the esterification reaction between ethanol and lactic acid and has been successfully applied in the extraction of flavonoids.

To develop extractive processes based on sustainable solvents for the recovery of natural products from biowaste, it is required to know the phase equilibria diagrams (e.g., liquid-liquid equilibria data) of the involved species. These data should refer to conditions which do not cause denaturation nor degradation of the target components, with ambient temperature and pressure being the most applied. The detailed knowledge of the phase diagrams is crucial for

choosing and ordering unit operations and for a proper sizing and economical assessment of the chemical processes.

Current Development

The solubility curves of green Aqueous Two-Phase Systems (ATPS) were determined following the cloud-point method at 298.15 K and 0.1 MPa. This method requires the titration of a solution of one of the water-soluble components (e.g., polyethylene glycol, PEG) by a solution of the other component (e.g., polyvinylpyrrolidone, PVP) until turbidity (or "cloudiness") is observed, as Fig. 1 shows.



Fig 1. Example of application of the cloud-point method: turbid mixture – left; clear mixture – right.

Then, if enough immiscible area was available in the phase diagrams, tie-line compositions were determined by performing isolines of thermophysical properties (e.g., conductivity and density) and/or lyophilisation, and modelled using excess Gibbs free energy models, such as UNIversal QUAsiChemical (UNIQUAC) and Non-Random Two-Liquid (NRTL), as Fig. 2 shows. These thermodynamic models were combined with Pitzer terms to describe long-range interactions, and an improved phase equilibria description was noticed by considering total dissociation of the present salts. Moreover, UNIQUAC-based approaches always outperformed the NRTL ones, which was justified by a stronger theoretical basis of the former.

The studied ATPS included ethyl lactate- (e.g., {ethyl

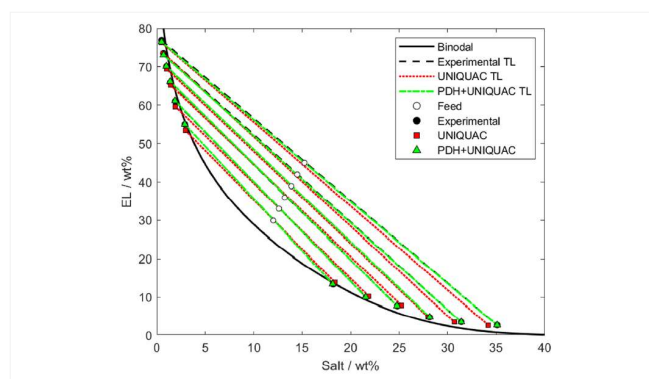


Fig 2. Experimental and correlated tie-line compositions of the ATPS {ethyl lactate (1) + NaH₂PO₄ (2) + water} at 298.15 K and 0.1 MPa.

lactate (1) + sodium potassium tartrate (2) + water (3)), PEG- (e.g., {PEG 4000 g·mol⁻¹ (1) + sodium formate (2) + water (3)}) and PVP-based (e.g., {PVP 29 000 g·mol⁻¹ (1) + disodium tartrate (2) + water (3)}) systems. Moreover, ATPS based on

ionic liquids, such as cholinium alaninate ([Ch][Ala]), cholinium leucinate ([Ch][Ley]) and cholinium glycinate ([Ch][Gly]), were also evaluated, as Fig. 3 illustrates.

hopefully leading to a better prediction of the partition coefficients of natural products, such as biomolecules (e.g., antioxidants and pigments) and active pharmaceutical

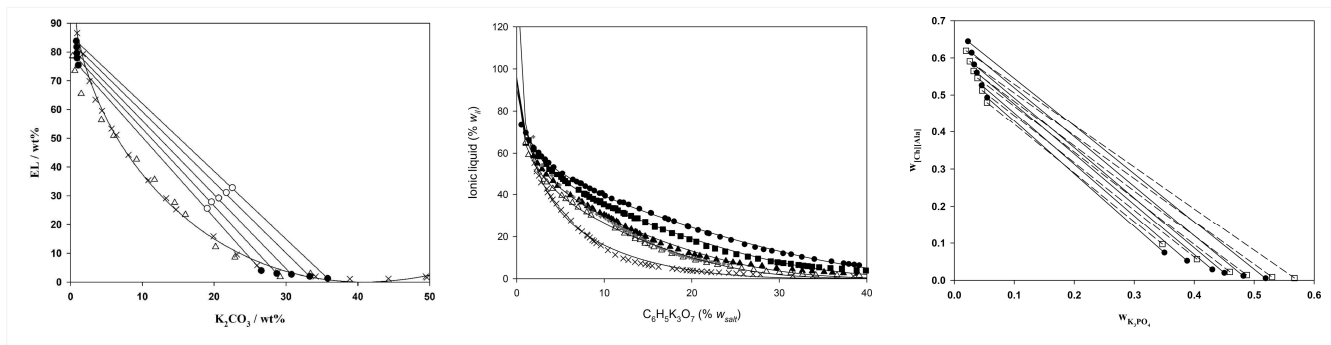


Fig 3. Liquid-liquid equilibria (binodal curve and/or tie-lines) at 298.15 K and 0.1 MPa for the ATPS: *left* – {EL (1) + K_2CO_3 (2) + water (3)}; *centre* – {● [C₂Mim][DCA] or ■ [C₃Mim][DCA] or ▲ [C₄Mim][DCA] or X [C₆Mim][DCA] or (Δ) [C₄Mpy][DCA] + $C_6H_5K_3O_7$ (2) + water (3)}; *right* – {[Ch][Ala] (1) + K_3PO_4 (2) + water (3)}.

Collaboration with the University of Las Palmas

A collaboration with the University of Las Palmas – Gran Canaria (Spain) was promoted for some months with the final purpose of drafting a book chapter on novel approaches to the fitting of parameters in the thermodynamic modelling of phase equilibria.

The final document was included as chapter 4 (“A Practical Fitting Method Involving a Trade-Off Decision in the Parametrization Procedure of a Thermodynamic Model and Its Repercussion on Distillation Processes”) in the book “Distillation: Modelling, Simulation and Optimization”, published in 2019 by IntechOpen. The chapter comprised a thorough review of some thermodynamic models, validation procedures for liquid-liquid equilibria (LLE) and vapour-liquid equilibria (VLE) data, and simulation alternatives.

Future Perspectives

The obtained liquid-liquid equilibria (LLE) data (binodal curves and tie-line compositions) always referred to 298.15 K and 0.1 MPa. Although these correspond to the ambient conditions, at which operation is less expensive and less energy-demanding, it would be interesting to study the effect of temperature and pressure on the phase diagrams, while preserving the feasibility of the tested conditions.

Due to the relevance of Aqueous Two-Phase Systems (ATPS) in the research group, LLE determination experiments represented more than half of the studied systems, with a complete absence of solid-liquid equilibria (SLE) studies. However, SLE data are essential to evaluate the feasibility of chemical processes, particularly to what pertains the extraction of biomolecules from biowaste, so they should be included in future works.

Furthermore, phase equilibria determinations involving other organic solvents besides ethyl lactate and other polymers besides polyethylene glycol and polyvinylpyrrolidone were not evaluated. Moreover, other types of green ionic liquids and a wider variety of inorganic salts could be tested to enlarge the number of assessed systems.

Finally, even though the research group has prior experience with the Cubic Plus Association (CPA) equation of state (EoS), the electrolyte-CPA EoS was never tested in the modelling of LLE data of ATPS. This equation of state could provide an enhanced predictive ability of the LLE data and deeper knowledge on the solvation of charged species,

ingredients (APIs), in ATPS.

Related Sustainable Development Goals



Outputs

PhD Theses

[1] Kamila Wysoczanska, Solubility and partitioning of biomolecules. Experimental studies and modeling, PDEQB, FEUP, 2020.

Master Dissertations

[1] C.S. Rebelo, Extraction of Antioxidants for Food Supplementation using Aqueous Two-Phase Systems with Low Environmental Impact, MEQ, FEUP, 2023.

Book Chapters

[1] A. Sosa et al., Chapter 4: A Practical Fitting Method Involving a Trade-Off Decision in the Parametrization Procedure of a Thermodynamic Model and Its Repercussion on Distillation Processes, pp. 47-70, Distillation: Modelling, Simulation and Optimization, IntechOpen, 2019.

Selected Publications

- [1] E. Gómez et al., J. Chem. Eng. Data 63, 1103-1108 (2018)
- [2] P.F. Requejo et al., J. Chem. Eng. Data 64, 4733-4740 (2019)
- [3] E. Gómez, E.A. Macedo, Fluid Phase Equilib. 484, 82-87 (2019)
- [4] P.F. Requejo et al., Ind. Eng. Chem. Res. 59, 21196-21204 (2020)
- [5] P. Velho et al., Sep. Purif. Technol. 275, 119155 (2021)
- [6] P. Velho et al., Fluid Phase Equilib. 556, 113389 (2022)

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