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Enhanced extraction of phenolic compounds using biobased solvents as hydrotropes

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Bruna de Paula Soares

**Extração de alta eficiência de compostos fenólicos usando solventes de base biológica
como hidrótropos**

Tese submetida ao Programa de Pós-Graduação em Engenharia Química da Universidade Federal de Santa Catarina como requisito parcial para a obtenção do título de Doutora em Engenharia Química.

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"Você deve perder o seu coração e buscá-lo em todos os lugares. Quando você o encontrar, você o descobrirá como o coração de todas as coisas"
(Sri Ram).

RESUMO

O aumento da solubilidade de substâncias pouco solúveis em água desempenha um papel importante na extração e purificação de compostos bioativos, como os compostos fenólicos, bem como em sua formulação e estudos de biodisponibilidade. A extração hidrotrópica está emergindo como uma alternativa promissora para a recuperação de compostos bioativos de matrizes orgânicas e para o desenho de novos processos de extração mais sustentáveis, uma vez que é baseada em soluções aquosas. Nesse cenário, o objetivo deste trabalho é estudar o mecanismo de ação de novos hidrotropos baseados em éteres de glicerol e alcanodíóis, para aumentar a solubilidade de compostos fenólicos. Para esse efeito, medições de solubilidades, espectroscopia de ressonância magnética nuclear de prótons (^1H -RMN), índice de polaridade I_1/I_3 envolvendo fluorescência e parâmetros empíricos Kamlet-Taft forneceram as informações que foram racionalizadas usando os modelos cooperativo-hidrotropo e Setschenow para esclarecer os mecanismos moleculares, e levar ao design aprimorado de novos hidrotropos. Em uma etapa final, os melhores hidrotropos identificados foram aplicados na extração de compostos fenólicos da fruta juçara. As medições de solubilidade usando éteres de glicerol e alcanodíóis como hidrotropos demonstraram a capacidade excepcional desses solventes para aumentar a solubilidade de compostos hidrofóbicos. As medições de ^1H -RMN racionalizadas por um modelo computacional demonstram que a apolaridade do soluto e do hidrotropo é a força motriz da hidrotropia. O modelo cooperativo-hidrotropo descreveu satisfatoriamente os dados experimentais e os parâmetros calculados corroboram o modelo de apolaridade proposto, assim como as constantes de Setschenow. Por fim, as extrações utilizando sistemas aquosos de alcanodíóis e éteres de glicol como solventes para recuperação de antocianinas da fruta de juçara demonstraram que esses solventes são tão eficientes quanto o etanol, um dos solventes mais utilizados nos processos de extração de biomassa, gerando extratos com alta concentração de compostos fenólicos, alta atividade antioxidante e prontamente disponíveis para aplicações cosméticas.

Palavras-chave: hidrotropos; compostos fenólicos; solubilidade; mecanismos de ação.

RESUMO EXPANDIDO

Introdução

Hidrótropos são substâncias orgânicas anfífilas que, quando presentes em água, aumentam significativamente a solubilidade de compostos hidrofóbicos. Em contraste com os surfactantes, os hidrótropos têm uma tendência fraca de auto agregação em água. Apesar dos muitos trabalhos abordando a hidrotropia, seu mecanismo de ação ainda não é totalmente compreendido. A quebra da estrutura da água e a formação de complexos foram algumas das teorias propostas. Mais recentemente, foi proposto o modelo cooperativo de hidrotropia, que é baseado na termodinâmica estatística. Nesse, é demonstrado que existe uma forte correlação entre hidrotropia e os volumes das estruturas apolares do soluto e do hidrótropo. Portanto, famílias de compostos em que a sua apolaridade pode ser facilmente ajustada, como os éteres de glicerol e alcanodióis, têm elevado potencial para desenvolver ferramentas que permitem entender melhor o mecanismo geral da hidrotropia, e consequentemente desenhar hidrótropos mais eficazes. Além disso, o aumento de solubilidade obtido com soluções hidrotrópicas desempenha um papel muito significativo na extração e purificação de compostos bioativos, tais como os compostos fenólicos, bem como na sua formulação e estudos de biodisponibilidade. Nesse sentido, a extração hidrotrópica, está emergindo como uma alternativa promissora para a recuperação de compostos bioativos de matrizes orgânicas e para o desenho de novos processos de extração mais sustentáveis, uma vez que é baseada em soluções aquosas. Técnicas emergentes de extração, como a extração assistida por ultrassom (UAE) e a extração por líquidos pressurizados (PLE) combinadas com solventes sustentáveis, têm vindo a ser adotadas para superar limitações como os elevados tempo de processamento e elevadas temperaturas de extração (diretamente relacionados com custos energéticos) e o impacto ambiental dos solventes empregados nas técnicas convencionais de extração. Embora os éteres de glicerol e os alcanodióis sejam solventes industriais bem conhecidos, e tenham sido adotados para muitas aplicações, incluindo a indústria alimentar e cosmética, têm sido pouco explorados como solventes para extração hidrotrópica. Deste modo, o seu uso em tais processos, direcionados para formulações industriais, podem conferir amplas vantagens nomeadamente ao nível da bioatividade dos extratos.

Objetivos

Esta tese tem como objetivo estudar o mecanismo de ação de novos hidrótropos baseados em éteres de glicerol e alcanodióis para aumentar a solubilidade de compostos fenólicos e outros solutos hidrofóbicos. Posto isto, pretende-se ainda utilizar os hidrótropos mais promissores no desenvolvimento um processo de extração hidrotrópica de compostos bioativos do fruto juçara.

Metodologia

Neste contexto, foram efetuadas medições de solubilidades (segundo a metodologia analítica do frasco-agitado), espectroscopia de ressonância magnética nuclear de prótons (^1H -RMN), índice de polaridade I_1/I_3 envolvendo fluorescência e parâmetros empíricos Kamlet-Taft (π^*), que forneceram informação relevante para o estudo dos modelos cooperativo de hidrotropia e empírico de Setschenow, e ainda avaliar a qualidade preditiva do modelo COSMO-RS, no sentido último de contribuir para o esclarecimento dos mecanismos moleculares, e auxiliar o desenho mais eficaz de novos hidrotropos. Inicialmente procedeu-se ao estudo das séries de mono e di-éteres de glicerol, e 1,2 e 1,n-alcanodióis. O efeito de temperatura também foi avaliado para hidrótropos não-iônicos, como o cireno, éteres de glicol e de glicerol, e

alcanodióis. Ao nível da aplicação prática, técnicas emergentes de extração como a assistida por ultrassom (UAE) e a de líquidos pressurizados (PLE), combinadas com soluções aquosas dos hidrotropos estudados (éteres de glicerol ou alcanodióis), sendo que as condições de extração foram otimizadas para a recuperação de antocianinas e outros compostos fenólicos da juçara. Por fim, os extratos obtidos utilizando 1,2-propanediol foram incorporados em formulações cosméticas de sabonetes naturais e cremes.

Resultados e Discussão

As medições de solubilidade usando éteres de glicerol e alcanodióis como hidrotropos demonstraram capacidade excepcional desses solventes para aumentar a solubilidade de compostos hidrofóbicos. Coadjuvadas por medições de ^1H -RMN, e aplicando um modelo computacional, foi demonstrado que a similaridade entre a apolaridade do soluto e do hidrótopo é a força motriz da hidrotropia. O aumento do tamanho da cadeia lateral alquílica dos éteres de glicerol forneceu informações importantes sobre esse mecanismo de hidrotropia, revelando que a magnitude do aumento da solubilidade de um determinado soluto se correlaciona diretamente com a apolaridade do hidrótopo, e atinge um máximo quando a área de superfície apolar do hidrótopo e do soluto são similares. Este máximo é discutido em termos das interações entre soluto-hidrótopo e hidrótopo-hidrótopo.

Para duas famílias diferentes de alcanodióis (1,2-alcanodióis e 1,n-alcanodióis), observa-se que na região diluída (baixa concentração de hidrótopo), a posição relativa dos grupos hidroxila dos alcanodióis não afetam as suas performances. Contudo, a sua capacidade em aumentar a solubilidade do ácido sirínico é notavelmente correlacionada com o tamanho das cadeias alquílicas. No entanto, a concentrações mais elevadas de hidrotropos o mecanismo é diferente, sendo que os 1,2-alcanodióis apresentam um desempenho, em geral, melhor do que os 1,n-alcanodióis. Esses resultados contraditórios foram reconciliados usando modelos e técnicas experimentais, nomeadamente o modelo cooperativo de hidrotropia e medições de escalas de polaridade. Os resultados sugerem que a auto agregação é mais prevalente nos 1,n-alcanodióis, o que afeta negativamente o seu desempenho como hidrotropos.

As escalas de polaridade π^* (Kamlet-Taft) e I_1/I_3 (fluorescência) foram utilizadas para avaliar os diferentes mecanismos de interação (dipolaridade/polarizabilidade, ligações de hidrogênio, etc.) que permeiam a solubilização do soluto por hidrotropos. A determinação das composições locais, através do parâmetro I_1/I_3 , sugerem um ponto de transição entre a agregação hidrotrópica e a formação de micelas que resulta do aumento da cadeia alquílica dos alcanodióis.

O efeito de temperatura foi avaliado para hidrotropos não-iônicos e os resultados mostram duas regiões bem definidas com mecanismos de ação bastante distintos. Na região diluída, controlada pela dissolução em água do soluto, as constantes de Setschenow são similares e os aumentos de solubilidade também são análogos, apesar das variações de temperatura. Entretanto, na região concentrada de hidrótopo, resultante da dissolução hidrotrópica do soluto, os fenômenos de auto-agregação são mais evidentes e o aumento de solubilidade é mais significativo a temperaturas mais baixas. Com base na equação de van't Hoff, as propriedades termodinâmicas da solução foram estimadas, indicando que o processo de dissolução do ácido sirínico em soluções aquosas de hidrotropos é endotérmico, mas controlado por via entrópica.

Em todos os estudos de solubilidade efetuados, o modelo cooperativo de hidrotropia descreveu satisfatoriamente os dados experimentais, e os parâmetros calculados suportam o modelo de apolaridade proposto, assim como as constantes de Setschenow.

Finalmente, os estudos de extração, utilizando alcanodióis e éteres de glicerol como solventes para recuperação de compostos fenólicos da fruta de juçara, demonstraram que esses solventes são tão eficientes quanto o etanol, um dos solventes mais utilizados nos processos de extração de biomassa, gerando extratos com alta concentração de compostos fenólicos e com potencial

atividade antioxidante. Esses extratos por sua vez geraram formulações cosméticas (sabonetes e cremes) com capacidade antioxidante e com propriedades inerentes ao 1,2-propanediol, considerado, dentre outras propriedades, como um ingrediente emoliente em cosméticos.

Considerações Finais

Os dados experimentais apresentados nesse trabalho são inéditos e suportam o modelo cooperativo de hidrotropia, recentemente proposto. A partir de uma perspectiva de aplicação, os resultados sugerem que os hidrótropos devem ser desenhados considerando a apolaridade do hidrótropo e soluto; o seu volume apolar disponível para as interações; e a sua tendência de auto-agregação. Fatores como a temperatura, e a possível formação de sistemas com separação líquido-líquido devem também ser considerados para aplicações específicas, como extrações, purificações e processos de cristalização. A aplicação dos hidrótropos estudados nesta tese (éteres de glicerol e alcanodióis) podem ser utilizados para extração de compostos bioativos de matrizes orgânicas e, podem ser diretamente utilizados em formulações cosméticas, em concentrações regulamentadas.

ABSTRACT

Enhancement of solubilization of poorly soluble substances in aqueous media plays a significant role in the extraction and purification of bioactive compounds, such as phenolic compounds, as well as in their formulation and bioavailability studies. Hydrotropic extraction is emerging as a promising alternative for the bioactive compounds recovery from organic matrices and the design of novel sustainable extraction processes since it is based on aqueous solutions. In this scenario, the principal objective is to study the mechanism of action of novel hydrotropes based on glycerol ethers and alkanediols, to enhance the solubility of phenolic compounds. For that purpose, solubilities, proton nuclear magnetic resonance spectroscopy (^1H -NMR), polarity index I_1/I_3 involving fluorescence, and Kamlet-Taft empirical parameters measurements provided the information rationalized using the cooperative-hydrotrope and Setschenow models to clarify the molecular mechanisms and lead to the enhanced design of novel hydrotropes. In a final step, the best hydrotropes identified will be applied to the extraction of phenolic compounds from juçara's fruit. Solubility measurements using glycerol ethers and alkanediols as hydrotropes have demonstrated the exceptional ability of these solvents to increase the solubility of hydrophobic compounds. ^1H -NMR measurements rationalized by a computational model demonstrate that the apolarity of both solute and hydrotrope is the driving force of hydrotropy. The cooperative model of hydrotrope satisfactorily described the experimental data, and the calculated parameters corroborate with the proposed apolarity model, as well as the Setschenow constants. Finally, extraction experiments using alkanediols and glycerol ethers as solvents for anthocyanins recovery from juçara's fruit demonstrated to be these solvents as efficient as ethanol, one of the preferred solvents in extraction processes from biomass, generating extracts with a high concentration of phenolic compounds and antioxidant activity, and readily available for cosmetic applications.

Keywords: hydrotropes; phenolic compounds; solubility; mechanisms of action.

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LIST OF ABBREVIATIONS AND ACRONYMS

Abbreviations / Acronyms

ABTS – Antioxidant Activity Assay
CAS – Chemical Abstract Service
CC- Critical Concentration
CCRD - Central Composite Rotatable Design
CER - Constant Extraction Rate period
CMC - Critical Micellar Concentration
C-NMR - Carbon Nuclear Magnetic Resonance
COSMO-RS - Conductor-like Screening Model for Real Solvents
DAD - Diode-Array Detection
DC - Diffusion-Controlled
DES – Deep Eutectic Solvents
DLS - Dynamic Light-Scattering
DSC - Differential Scanning Calorimetry
FDA - Food and Drug Administration
FST - Fluctuation Solution Theory
GC - Gas Chromatography
GRAS - Generally Recognized as Safe
HBA - Hydrogen-bond acceptor
HBD - Hydrogen-bond donor
H-NMR - Proton Nuclear Magnetic Resonance
HPLC - High-Performance Liquid Chromatography
ILs – Ionic Liquids
KB - Kirkwood-Buff
KBI - Kirkwood-Buff Integrals
MAE - Microwave-Assisted Extraction
MHC - Minimum Hydrotrope Concentration
NADES - Natural Deep Eutectic Solvents
OEC - Overall Extraction Curve
PID - Proportional–Integral Derivative controller
PLE - Pressurized Liquid Extraction

RDF - Radial Distribution Function
 SANS - Small-Angle Neutron Scattering
 SAXS - Small Angle X-ray Scattering
 SFE - Supercritical Fluid Extraction
 SLE – Solid-Liquid Equilibrium
 TPC – Total Phenolic Content
 TZVP – Triple Zeta Valence Polarized basis
 UAE - Ultrasound-Assisted Extraction
 UV - Ultraviolet

Symbols

(ΔG_{dis}^0) - Apparent dissolution Gibbs energy
 (ΔH_{dis}^0) – Apparent enthalpy of dissolution
 (ΔS_{dis}^0) – Apparent entropy of dissolution
 $\bar{V}_i$ - Partial molar volume of compound i
 $\Delta_m h$ - Enthalpy of melting
 b - Facility of inserting m hydrotrope molecules in the vicinity of the solute
 C_i – Molar concentration of compound i
 $g_{ij}(r)$ - Radial distribution function between species i and j
 I_1/I_3 or Py - Polarity scale from pyrene
 K_H/K_S - Setschenow constant
 K_{ow} - Octanol/water partition coefficients
 k_T - Isothermal compressibility
 m - Average number of hydrotropes that aggregate around a given molecule of solute
 n_i - Density in molarity of compound i
 p - Pressure
 r - Distance between the centers of molecules i and j (radius)
 R – Ideal gas constant
 S – Solubility of the drug in the hydrotrope aqueous solution
 S_0 - Solubility of the drug in pure water
 T – Absolute temperature
 T_{hm} - Harmonic mean temperature

T_m – Melting temperature

x_i - Mole fraction of compound i

x_L or x_H^L – Local composition form *Py* scale

Greek symbols

α – Solvent hydrogen-bond donating ability

β – Solvent hydrogen-bond acceptor ability

γ_i - Activity coefficient of compound i

ζ_H – Enthalpy relative contribution

ζ_{TS} – Entropy relative contribution

μ_i - Chemical potential for each component

π^* - Solvent dipolarity/polarizability parameter

ν - Experimental wave number

Chemicals

(C₂P₁) 1-ethoxy-2- propanol

(C₄E₁) 2-butoxy-2-ethanol

[1.0.0] 3-methoxypropan-1,2-diol

[1.0.1] 1,3-dimethoxypropan-2-ol

[1.0.2] 1-ethoxy-3-methoxypropan-2-ol

[1.0.4] 1-butoxy-3-methoxypropan-2-ol

[2.0.0] 3-ethoxypropan-1,2-diol

[2.0.2] 1,3-diethoxypropan-2-ol

[2.1.1] - 1-ethoxy-2,3-dimethoxypropane

[2.2.2]- 1,2,3-triethoxypropane

[3.0.0] 3-propoxypropan-1,2-diol

[3.1.1] - 1,2-dimethoxy-3-propoxypropane

[4.0.0] 3-butoxypropan-1,2-diol

[4.1.0] 3-butoxy-2-methoxypropan-1-ol

[4.1.1] - 1-butoxy-(2,3)-dimethoxypropane

[5.0.0] 3-pentoxypropan-1,2-diol

[6.0.0] 3-hexyloxypropan-1,2-diol

[C₂C₁im] [N(CN)₂] - 1-ethyl-3-methyl- imidazolium dicyanamide

[C₂C₁im] Cl - 1-ethyl-3-methyl- imidazolium chloride
[C₄C₁im] [N(CN)₂] - 1-butyl-3-methylimidazolium dicyanamide
[C₄C₁im] [SCN] - 1-butyl-3-methylimidazolium thiocyanate
[C₄C₁im] [TOS] 1-butyl-3-methylimidazolium tosylate
[R.₀.₀] / [x.₀.₀] - Glycerol monoethers
[R.₀._R] / [x.₀._x] - Glycerol symmetric diethers
[R₁. R₂. R₃] / [x,_y,_z]- Glycerol asymmetric triethers
[R₁.₀. R₂] / [x,₀,_z]/ [x,_y,₀]- Glycerol asymmetric diethers
ABTS⁺ - 2,2-azinobis (3-ethylbenzothiazoline-6-sulfonic acid)
Al(OTf)₃ - Aluminum trifluoromethanesulfonate
BF₃Et₂O - Boron trifluoride diethyl etherate
Bi(OTf)₃ - Bismuth(III) trifluoromethanesulfonate
BSA - Bovine serum albumin
C_iE_j - Ethylene glycol ethers
C_iP_j - Propylene glycol ethers
CO₂ - Carbon dioxide
DMSO - Dimethyl sulfoxide
H₂SO₄ - Sulfuric acid
HCl - Hydrogen chloride
K₂S₂O₈ - Potassium persulfate
Na₂CO₃ - Sodium carbonate
NaOH - Sodium hydroxide
OH⁻ - hydroxyl group
SBS - Sodium benzene sulfonate
sc-CO₂ - supercritical carbon dioxide
STS - Sodium toluene sulfonate
SXS - Sodium xylene sulfonate

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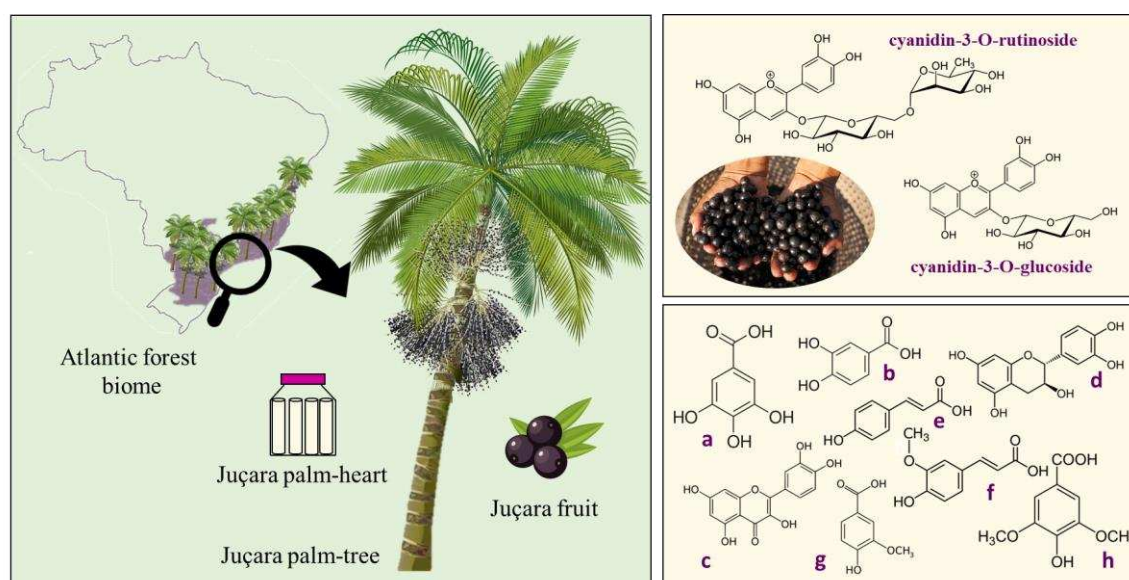
1. CHAPTER I - INTRODUCTION

In recent years, the concept of sustainable chemistry has acquired great importance for the design of chemicals and (BEHR *et al.*, 2008) processes less aggressive to the environment. The term Green Chemistry was first proposed in 1991 by Paul T. Anastas and John C. Warner and its twelve principles were developed by the same researchers in 1998 (ANASTAS; WARNER, 1998). Among the twelve principles of Green Chemistry are solvent minimization, substitution and recyclability (ERYTHROPEL *et al.*, 2018). Alternatives to replace solvents from the petrochemical industry, and adopt the fundamental chemistry deriving from aqueous solvent systems, use of biomass-derived solvents, and a more efficient chemical transformation for synthesis must be considered when thinking about solvent selection or how to design a sustainable process (BYRNE *et al.*, 2016; CLARKE *et al.*, 2018; ERYTHROPEL *et al.*, 2018). The high demand for petroleum products, the big wave of urbanization of cities, especially in developing countries, and the balance between human consumption and the depletion of natural sources, have created challenges and opportunities for the use of environmental resources. In this scenario, the concept of biorefinery emerges as an opportunity to use food by-products as compounds of commercial value, such as antioxidants as phenolic compounds.

Phenolic compounds are a widely used class of bioactive compounds, which can be extracted from industrial by-products. The depolymerization products of lignin, a by-product of cellulose pulping processes, contain a wide number of phenolic compounds such as vanillin, syringaldehyde, acetovanillone, vanillic acid, and syringic acid (FACHE; BOUTEVIN; CAILLOL, 2015; MOTA *et al.*, 2016; SOARES *et al.*, 2017). A large variety of phenolic compounds can also be extracted from biomass derived from food by-products. Ferulic, gallic and ellagic acids are the major phenolic compounds found in feijoa peel extracts (SANTOS *et al.*, 2019). The chestnut bur contains gallic acid esters of glucose, ellagic acid, and quercetin (VÁZQUEZ *et al.*, 2012). In grape berries, flavonols are present mainly in skins and the seeds are rich in flavan-3-ols, both flavonoids (MONTEALEGRE *et al.*, 2006). The juçara palm, a Brazilian endemic species, produces a noble type of palm heart, and also the juçara's fruit (Figure 1.1). With a high concentration of the anthocyanins cyanidin-3-O-rutinoside and cyanidin-3-O-glucoside, these are the main constituents of the phenolic juçara fruits' profile (around 73% of the total phenolic compounds content) (VIEIRA *et al.*, 2017), which confers to the fruit the intense dark purple color. In addition to anthocyanins, recent studies indicate

among the phenolic constituents (see Figure 1.1) a high content of gallic and protocatechuic acids and quercetin (AZEVEDO *et al.*, 2014; BICUDO; RIBANI; BETA, 2014; BORGES *et al.*, 2011). Juçara fruit is considered a super fruit since it has a high nutritional value due to its richness in nutrients and antioxidants (FELZENSZWALB *et al.*, 2013; SCHULZ *et al.*, 2016). Despite that, its valorization is of major importance, since the juçara palm tree is threatened with extinction due to the exploitation of the palm heart.

Figure 1.1. Scheme of the juçara palm-tree, juçara palm-heart and juçara fruit from Atlantic Forest and the main phenolic compounds constituents for juçara's fruit, in addition to anthocyanins: gallic acid (a), protocatechuic acid (b), quercetin (c), catechin (d), coumaric acid (e), ferulic acid (f), vanillic acid (g) and syringic acid (h).



In this line, the enhanced solubility of poorly soluble substances in aqueous media plays an important role in the extraction and purification of bioactive compounds, such as the phenolic constituents from juçara's fruit, as well as in their formulation and bioavailability studies. In this sense, the concept of hydrotropes appears, since they are compounds able to substantially increase the solubility of hydrophobic substances in water (EASTOE; HATZOPOULOS; DOWDING, 2011). The study of hydrotropes has been widely investigated since the concept of hydrotropy was proposed by Neuberg in 1916 (NEUBERG, 1916). Nevertheless, despite the large number of works addressing hydrotropy, its mechanism of action is not yet clearly understood (EASTOE; HATZOPOULOS; DOWDING, 2011). Despite that, the search for new solvents from

renewable sources, so-called biobased solvents, the need to produce less toxic and less aggressive solvents for the environment, and the search for compounds that facilitate the bioavailability and extraction of natural compounds are intimately related to both the concepts of biorefinery and hydrotropy.

1.1. OBJECTIVES

1.1.1. General Objective

This thesis is founded on the study of mechanisms of action of novel hydrotropes based on glycerol ethers and alkanediols to enhance the solubility of phenolic compounds and other hydrophobic solutes. The best hydrotropes were evaluated on the extraction of phenolic compounds from juçara's fruit, by emergent techniques of extraction.

1.1.2. Specific Objectives

- a) *Synthesize* the glycerol mono/di ethers and *select* the alkanediols for hydrotropy studies.
- b) *Measure* the solubility curves of phenolic compounds and other model molecules in aqueous hydrotrope solutions at 303.2 K and at different temperatures (ranging from 278 to 343K);
- c) *Rationalize* the solubility data by nuclear magnetic resonance spectroscopy (^1H -NMR) and polarity scales, namely the π^* Kamlet-Taft solvatochromic parameter and the P_y index;
- d) *Describe* the solubility data using the cooperative model of hydrotropy, Setschenow constants, and computational tools;
- e) *Interpret* the physical chemistry of the interactions from the experimental data and models;
- f) *Evaluate* the best hydrotropes on the extraction of phenolic compounds from juçara's fruit, by emergent extraction techniques and
- g) *Formulate* cosmetic products by using the phenolic-rich extracts obtained.

1.2. THESIS STRUCTURE

The thesis starts with the literature review in **Chapter 2**, which describes the hydrotropic phenomena, including some examples of biobased solvents that can act as hydrotropes, such as ionic liquids, deep eutectic solvents, glycerol ethers, and alkanediols.

Then, the Kirkwood-Buff Theory was scrutinized to make possible the understanding of thermodynamic models that address hydrotropy. Finally, some extraction procedures commonly used for the recovery of phenolic compounds were reported as well as emergent extraction methodologies.

Chapter 3 describes the investigation of glycerol ethers as hydrotropes. Some classes of neoteric solvents, such as ionic liquids and deep eutectic solvents, are shown to be promising classes of hydrotropes (CLÁUDIO *et al.*, 2015; SINTRA *et al.*, 2018; SOARES *et al.*, 2017) and recently, the glycerol ethers have also been proposed for the same effect (LEBEUF *et al.*, 2016; MOITY *et al.*, 2013; QUESTE *et al.*, 2006a). However, an integrated approach to study these compounds as hydrotropes, trying to achieve an understanding of the molecular level mechanism of hydrotropy that could lead to the design of improved hydrotropes has never been systematically evaluated. In **Chapter 3.1**, mono and dialkyl glycerol ethers were evaluated to increase the aqueous solubility of gallic and syringic acids, changing the glycerol ether alkyl side chain. Solubility curves provided the information that was rationalized using the Cooperative-Hydrotrope (SHIMIZU; MATUBAYASI, 2016) and Setschenow models (KUNZ; HOLMBERG; ZEMB, 2016; SETSCHENOW, 1989) to clarify the molecular mechanisms and lead to the enhanced design of novel hydrotropes. **Chapter 3.2** goes deeper into the study of these compounds, applying experimental ¹H-NMR studies and a new computational technique (based on COSMO-RS predictions) to evaluate the apolarity effect on hydrotropy. **Chapter 3.3** contains results obtained using glycol ethers, a common class of petrochemical-sourced hydrotropes (MOLINIER; AUBRY, 2014) (which have the glycerol ethers studied in **Chapters 3.1** and **3.2** as replacement alternatives). Interestingly, during the studies with glycol ethers as hydrotropes, and vanillic acid as a solute, the unexpected oiling-out effect occurred (DE ALBUQUERQUE; MAZZOTTI, 2014; DU *et al.*, 2016; SVÄRD; GRACIN; RASMUSON, 2007). Thus, the oiling-out phenomenon is described and discussed for vanillic acid solubilization and reconciled by COSMO-RS predictions of the solid-liquid phase diagram.

Given the dependence of hydrotropy on the apolar volumes of solute and hydrotrope, families of compounds with easily tuned apolarities, such as glycerol ethers and alkanediols, are a powerful tool to understand the overall mechanism and rationally design better green hydrotropes for specific applications. In this sense, in a continuous

line with the previous chapters, **Chapter 4** demonstrates the study of alkanediols as hydrotropes. To assess the impact of the alkyl chain length (size) and relative position of polar groups (shape) of hydrotropes on the solubility enhancement of hydrophobic solutes, the ability of two sets of alkanediols (1,2-alkanediols and 1,n-alkanediols) to increase the aqueous solubility of syringic acid was investigated in **Chapter 4.1**. The data were reconciled using theoretical and experimental techniques, namely the cooperative model of hydrotropy and chemical probes. In **Chapter 4.2**, to assess the contributions of the various interactions (dipolarity/polarizability, hydrogen bonding, etc.) that permeate the solute solubilization by hydrotropes, Kamlet–Taft solvatochromic parameters π^* and P_y polarity scale (I_1/I_3) were measured for all the alkanediols studied in **Chapter 4.1** and correlated with the enhanced solubility. The local composition calculations from I_1/I_3 scale were used to delineate a transition point from hydrotropic to micellar aggregation when increasing the alkanediols alkyl chain. COSMO-RS has been also used as a relevant tool to rationalize the experimental results for glycerol and glycol ethers in **Chapters 3.2** and **Chapter 3.3**, respectively. In **Chapter 4.3** the predictions by COSMO-RS, obtained for solubility curves using alkanediols as hydrotropes for different solutes (syringic acid and pyrene) are reported, and the future perspectives to improve the predictions are discussed.

Besides glycerol ethers and alkanediols, there are some main chemical families, or biobased compounds, that reveal hydrotropic properties, including anionic, cationic, or neutral freely soluble organic molecules. Among the non-ionic hydrotropes, cyrene was recently proposed as a green solvent and has also been studied as a hydrotrope (DE BRUYN *et al.*, 2019). Cyrene, together with the compounds previously studied in **Chapter 3** and **Chapter 4**, namely glycerol and glycol ethers, and alkanediols, enclose the non-ionic hydrotrope family. Classical aromatic anionic hydrotropes, such as sodium salicylate and sodium cumene sulfonate have been studied from the perspective of thermodynamic properties (DESAI; PARIKH, 2009; WAGLE; KOTHARI; GAIKAR, 2007), in particular on the effect of temperature. However, similar properties of aqueous non-charged or non-aromatic hydrotropes have not been explored to the same extent although these compounds are commonly used for industrial applications. In this sense, **Chapter 5**, which includes the single **Chapter 5.1**, presents the impact of temperature on hydrotropy, studying six selected non-charged compounds (cyrene, the glycerol ether [1.0.0], the glycol ether 2-ethoxyethanol and the alkanediols 1,2-propanediol, 1,3-

butanediol, and 1,2-pentanediol) to enhance the solubility of syringic acid as the model compound. In order to analyze the soluble procedure from a thermodynamic aspect, the apparent solution thermodynamic properties (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) of the syringic acid dissolution processes of the six investigated aqueous hydrotropes solutions were estimated by the van't Hoff equation.

One of the outcomes of the investigation of solubility enhancement is the extraction of valuable compounds. Hydrotropic extraction is emerging as a promising alternative for the extraction of bioactive compounds and the design of new sustainable extraction processes as it is based on aqueous solutions (EASTOE; HATZOPOULOS; DOWDING, 2011). In this sense, **Chapter 6.1** brought up an application of some of the best hydrotropes studied in this thesis in the extraction of bioactive compounds from an organic matrix, the juçara fruit. The valorization of juçara fruits becomes important since its tree (juçara palm-tree) is threatened with extinction due to the extraction of its palm-heart. The extractions were carried out using alkanediols and glycerol ethers aqueous solutions as solvents, combined with emergent extraction methodologies, such as pressurized liquid extraction and ultrasound-assisted extraction. Since some of the compounds under study are approved by FDA (Food and Drug Administration) and also regulated ingredients of cosmetic formulations, the resulting anthocyanins-rich extracts were directly used (without polishing steps) to produce natural soaps and creams.

Finally, the conclusions are addressed, and future work envisioned in **Chapter 7**.

2. CHAPTER II - LITERATURE REVIEW

2.1. HYDROTROPES

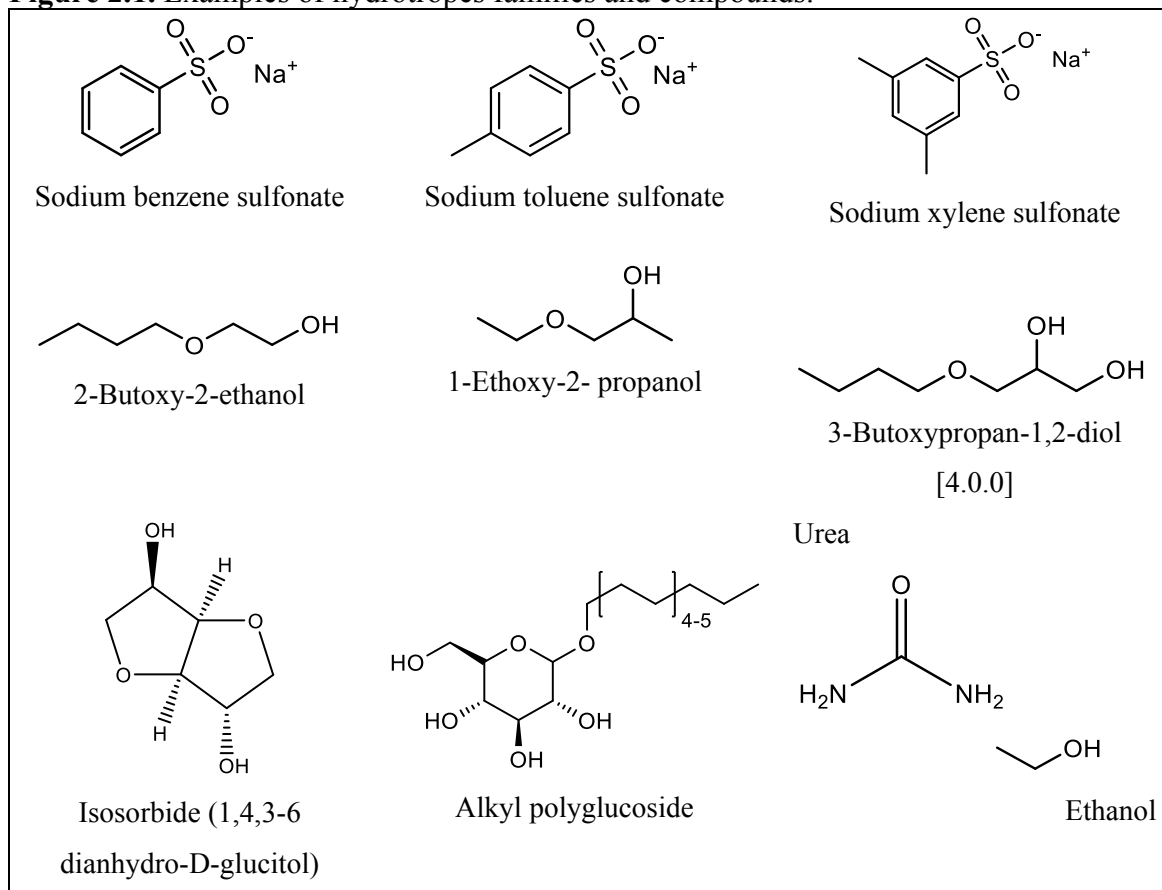
Hydrotropes are compounds used to increase the solubility of hydrophobic solutes in aqueous solvents. The concept of hydrotropy was mentioned for the first time in 1916 when Neuberg dissolved a variety of organic compounds into aqueous solutions containing, what would be called, hydrotropes (NEUBERG, 1916). Historically, the ability of the hydrotrope to solubilize organic compounds in water was associated with aggregate formation according to a stack-type mechanism, described for short-chain aromatic salts, as were reported, in a first moment, being the hydrotropes (KUNZ; HOLMBERG; ZEMB, 2016). Some of the compounds first mentioned were sodium benzoate, sodium xylene sulfonate, sodium p-toluene sulfonate, and sodium benzenesulfonate (FRIBERG; BLUTE, 2006).

In a second moment, 30 years later, McKee (MCKEE, 1946) realized that solutions of hydrotropes when diluted, induced the separation of the dissolved hydrophobic compound. Then, the aqueous solution of hydrotrope could be recovered and reused for extraction. During this period the first indications that the enhanced solubility is of the type "salting-in" effect was described by McKee (MCKEE, 1946), and supported by other authors (FRIBERG; BLUTE, 2006). A few years later, the phase diagrams for hydrotrope solutions studied by Lawrence (LAWRENCE, 1959) and Friberg and Rydhag (FRIBERG; RYDHAG, 1970) brought new insights into the phenomena involved in solubilization. The solubility enhancement would be caused by the influence of the hydrotrope molecule on the packing conditions in colloidal association structures, and their presence would prevent the formation of the lamellar liquid crystal region. The phase diagrams are still a tool for recent works related to the hydrotropic effect, and this topic will be discussed in more detail later.

There are some main chemical families or compounds that reveal hydrotropic properties, some of which are represented in Figure 2.1. The aromatics, represented by anionic salts of petroleum origin are the most common and usual hydrotropes, sodium benzene sulfonate (SBS), sodium toluene sulfonate (STS), and sodium xylene sulfonate (SXS) probably being the best examples (DHAPTE; MEHTA, 2015; HODGDON; KALER, 2007). The ethylene glycol ethers abbreviated by C_iE_j (i is the number of carbons of the hydrophobic part and j is the number of ethoxylates) and the propylene glycol ethers (C_iP_j) are the most common non-ionic alkyl-hydrotropes and their thermodynamic and toxicological properties have been studied extensively since they may present

reprotoxicity (BAUDUIN *et al.*, 2005; MOLINIER; AUBRY, 2014). Between the aliphatic hydrotropes, there are also the bio-sourced sugars and polyols derivatives, including the alkyl polyglucosides and the alkyl polipentosides, exhibiting good environmental and toxicological profiles (MOLINIER; AUBRY, 2014). As an alternative for the petro-sourced hydrotropes, esters and ethers of isosorbide and glycerol have been widely evaluated as an alternative to glycol ethers and routes of green synthesis have been increasingly studied for these compounds (DURAND *et al.*, 2009; HERBINSKI *et al.*, 2018; LEBEUF *et al.*, 2016; MOITY *et al.*, 2013; MOLINIER; AUBRY, 2014). Ethanol, urea, and other small molecules can also be considered hydrotropes. Urea has a particular biological function, acting both as a donor and an acceptor of hydrogen bonds (BOOTH *et al.*, 2015; FRIBERG *et al.*, 1999; KUNZ; HOLMBERG; ZEMB, 2016).

Figure 2.1. Examples of hydrotropes families and compounds.



The properties of hydrotropes are not only restricted to enhancing the solubility of hydrophobic compounds. They also stabilize aqueous solutions, modify their viscosity and limit low-temperature phase separation (SUBBARAO *et al.*, 2012). In contrast to surfactants, hydrotropes have a weak tendency to self-aggregate in water and this is a

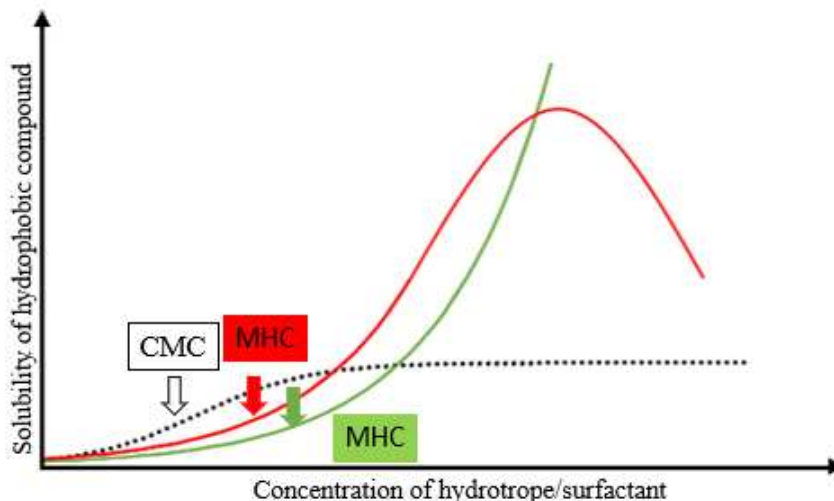
prerequisite for a number of applications such as drug solubilization, chemical reactions, liquid-liquid separation, and solid-liquid extraction, which make them powerful compounds for the extraction of natural products such as antioxidants, and great allies in the context of biomass valorization and biorefineries (ANSARI; GAIKAR, 2014; SUBBARAO *et al.*, 2012). Furthermore, hydrotropes, are interesting from a green chemistry perspective as they can present, in general, low toxicity and low bioaccumulation potential due to their low octanol-water partition coefficients (STANTON *et al.*, 2009).

Over the years, molecules without an aromatic ring and even without charge, as mentioned before, turned out to possess hydrotropic properties and for those, the stacking mechanism first proposed was not the most adequate (KIM *et al.*, 2010; KUNZ; HOLMBERG; ZEMB, 2016). Despite the large number of reviews addressing hydrotropy, its mechanism of action is not yet clearly understood (EASTOE; HATZOPOULOS; DOWDING, 2011). There are three main approaches to explain the mechanism of the hydrotropic solubilization phenomenon, namely (i) self-aggregation of hydrotrope molecules; (ii) disruption of water structure by the hydrotropes; and (iii) complex formations between solute and hydrotrope. Recently, some groups, have been proposing mechanisms over this concept: the co-aggregation of the solute with the hydrotropes above a minimum hydrotrope concentration (MHC), which correlate the stability of hydrotrope aggregation form relative to its monomeric form (BOOTH *et al.*, 2015; BOOTH; ABBOTT; SHIMIZU, 2012; LEE *et al.*, 2003; NEUMANN *et al.*, 2007; SHIMIZU; BOOTH; ABBOTT, 2013; SHIMIZU; MATUBAYASI, 2014, 2016). The MHC is the concentration at which the hydrotropic effect starts expressing and when some properties in solution also change, such as solubility, surface tension, viscosity, conductivity, and others (MOLINIER; AUBRY, 2014; SUBBARAO *et al.*, 2012). The measurements of MHC coincide with a change in the slope of the properties (e.g. solubility) as a function of the hydrotrope concentration (BAUDUIN *et al.*, 2005), although some authors have shown that for some short alkyl chain compounds, the clear change in behavior attributable to a critical phenomenon is not easily interpreted, since the solubility curves show continuous changes (CLÁUDIO *et al.*, 2015; SUBRAMANIAN *et al.*, 2011).

The hydrophobic part of hydrotropes is too small to induce micelle formation, because of that the aggregation does not occur at a critical micellar concentration (CMC)

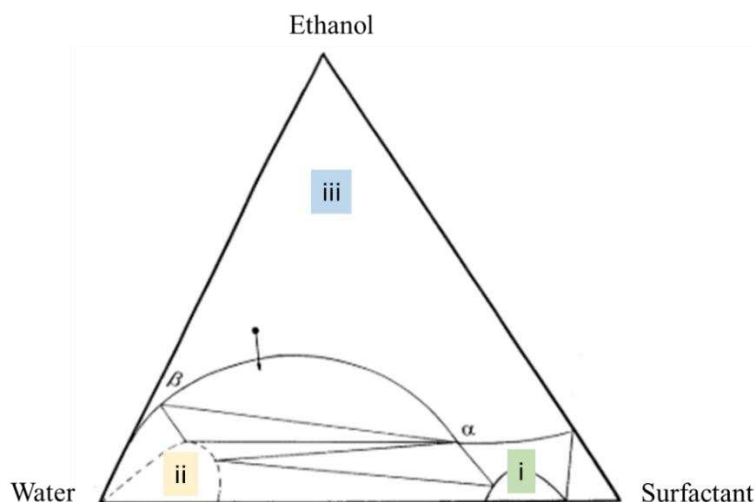
as observed in surfactants. The aggregates appear at much higher concentrations in the case of hydrotropes and occur continuously starting from the MHC or until reaching a maximum solubility (MOLINIER; AUBRY, 2014) as can be observed in Figure 2.2.

Figure 2.2. Representation of CMC and MHC solubilization curves for a hydrophobic compound.



As mentioned before, the colloidal association structure is the key to the superior solubilization in a hydrotrope solution compared to that in a surfactant solution since the addition of a hydrotrope destabilizes the lamellar structure (FRIBERG; BLUTE, 2006). The main difference between hydrotrope and micelle aggregation is the liquid crystal region in their phase diagram, which is observed for the micellar formation and do not observed for aqueous solutions of hydrotropes that display a single continuous isotropic liquid region. Friberg *et al.* (FRIBERG *et al.*, 1999) determined, by visual observation of the one-phases, the phase diagram for water + ethanol (as a hydrotrope) + surfactant. Figure 2.3 illustrates the lamellar liquid crystal regions for the system (i and ii) and the region of isotropic liquid (iii). The results show that the lamellar liquid crystal becomes disordered upon the addition of ethanol, forming an isotropic liquid.

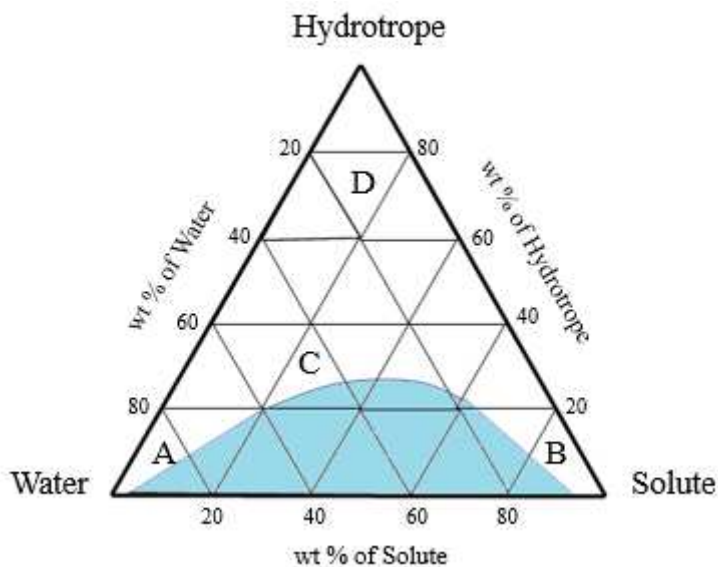
Figure 2.3. The phase diagram for the system water + ethanol + surfactant. (i and ii) lamellar liquid crystals. (iii) isotropic liquid solution.



Source: Adapted from Friberg *et al.* (FRIBERG *et al.*, 1999).

In the work by Kunz *et al.* (KUNZ; HOLMBERG; ZEMB, 2016) the authors organized the tertiary phase diagram for the usual hydrotrope + water + solute system to clarify the main acting forces to the hydrotrope mechanism in different concentrations of each component (see Figure 2.4). Region A is where at low hydrotrope concentration the solubility of the organic solute is significantly enhanced, and it is the region for most of the hydrotrope applications. B is the region where hydrotrope-enhanced solubility of water in the solute is known as solvotropy, where there is symmetry between diluted domains. Region C represents a highly fluctuating cluster consisting of many solute molecules plus water and hydrotrope molecules, known as pre-Ouzo clustering, and finally, Region D is where the hydrotrope concentration is highest, and the molecular distribution of the three compounds is random.

Figure 2.4. A typical ternary phase diagram water-solute-hydrotrope. The numbers refer to different domains corresponding to different solubilization mechanisms. Region A: classical hydrotrope action; Region B: solvotrope action. Region C: pre-Ouzo clustering. Region D: random molecular distribution of all components.



Source: Adapted from Kunz *et al.* (KUNZ; HOLMBERG; ZEMB, 2016).

The hydrophobic group in the hydrotrope has been cited as the most determining factor for the hydrotropic interactions and the differences between surfactants, co-solvents, and hydrotropes is a topic of many works, but according to the relative concentrations in a ternary mixture, a component can act either as a co-solvent or a hydrotrope (KUNZ; HOLMBERG; ZEMB, 2016), as in the case of the system containing water-ethanol-octanol. Ethanol, short-chain alcohol unable to form micelles, is considered a hydrotrope since it can form defined surfactant aggregates. In the case of the ternary system, in which the ethanol partition forms an interface between the pseudo-phases, fluctuating structures appear alternating between well-defined aggregates in an oil-rich and oil-poor domains, as demonstrated by light scattering experiments (ZEMB *et al.*, 2016). Different from what was understood, Bauduin *et al.* (BAUDUIN *et al.*, 2005) testing two different dyes as hydrophobic solutes, showed the shape of the solubilization curves for cosolvents, hydrotropes, and solvosurfactants are similar, and the only difference between them is the minimum hydrotrope concentration (MHC), calculated through the measurements of surface tension as a function of the hydrotrope concentration. However, as previously reported, the MHC values could not be precisely determined for the short amphiphile and cosolvent molecules, since the solubility of the solute increases exponentially with

increasing concentration. Testing the two different dyes for solubility enhancement, these authors could also conclude that the aqueous solubilization is not sensitive to the hydrophobic solute, it is the hydrophobic chain of the hydrotrope the main responsible for the phenomena (BAUDUIN *et al.*, 2005).

2.1.1. Biobased Solvents as Hydrotropes

Most of the substances that present hydrotropic properties are petrochemical-based. Their study and replacement by green solvents and the development of sustainable technology processes using biobased solvents in extraction and purification processes is of great importance on the grounds of the Green Chemistry concept. Among the green solvents that have been studied, there are many works addressing ionic liquids and deep eutectic solvents (CLARKE *et al.*, 2018), but there are only a few studies that investigate their hydrotropic effect. Glycerol ethers, although already mentioned for some time as hydrotropes (QUESTE *et al.*, 2006a) are also little explored for sustainable extraction processes.

2.1.2. Ionic Liquids

Ionic liquids (ILs) are a novel class of solvents that at room temperature are molten salts. They are composed of two distinct parts (anionic and cationic), resulting in synthetic flexibility, which has given them the “designer solvents” characteristic (CHEN *et al.*, 2014; FREIRE *et al.*, 2012). In this way, it is possible to tune their properties, such as density and viscosity, to synthesize compounds for specific applications (PANTELI; VOUTSAS, 2010). ILs have been proposed as alternatives to volatile organic solvents since they exhibit low vapor pressures at environmental temperatures, high chemical, and thermal stability, and high solvating capacity for organic, inorganic, and polymeric materials (EARLE *et al.*, 2006; ROCHA; COUTINHO; SANTOS, 2014; ROGERS; SEDDON, 2003), characteristics that give them great potential as alternative solvents and many applications in extraction processes (CLÁUDIO *et al.*, 2018; PASSOS; FREIRE; COUTINHO, 2014).

Recently, some ILs were shown as a promising class of catanionic hydrotropes since both parts of their structure play a role in the increase of the solubility of hydrophobic compounds in an aqueous solution. Rengstl *et al.* (RENGSTL *et al.*, 2014) showed the capacity of the short-chain choline carboxylates ionic liquids acting as hydrotropes for a hydrophobic dye. In their study, cytotoxicity tests were also performed

showing slightly higher toxicity for hydrotropes compared to the surfactants (the long-chain choline carboxylates).

Some recent studies go deeper into the hydrotropic phenomena through ionic liquids. Cláudio *et al.* (CLÁUDIO *et al.*, 2015), studied different ionic liquids for the solubilization of vanillin or gallic acid in aqueous solutions, with vanillin solubility enhancements of up to 40-fold for [C₂C₁im][N(CN)₂], [C₄C₁im]Cl and [C₄C₁im][TOS]. In this work, the authors disclose hydrotropy as a synergistic effect: both anion and cation contribute to the solubility enhancement of the hydrophobic compounds. Comparing a wide range of different ionic liquids, the non-aromatic ones also had hydrotropic effects, since they are not formed by $\pi \cdots \pi$ interactions, those driving forces would not be enough to explain the phenomenon behind the solubility improvement promoted by such compounds. In addition, the formation of complexes was not revealed by UV spectroscopy analyzes and the existence of co-aggregates between the hydrophobic solute and the IL ions in an aqueous solution was probed by molecular dynamics simulations. The authors also mentioned that there are significant differences in the solubility increase between vanillin and gallic acid, for the same hydrotrope, which shows the specificity character of each solute, which can be highly favorable.

In 2018, Sintra *et al.* (SINTRA *et al.*, 2018) investigated ten ionic liquids for ibuprofen solubilization. The results showed that the performance for some ILs was superior to conventional hydrotropes and salting-in inducing salts, the [C₄C₁im][N(CN)₂] and [C₄C₁im][SCN] displaying the best hydrotropic effect to enhance the solubility of ibuprofen. Molecular dynamics simulations for these compounds confirm the formation of ibuprofen-IL-aggregates like Cláudio *et al.* (CLÁUDIO *et al.*, 2015) reported for the antioxidants. From these studies, the ionic liquids become important tools for the understanding of the hydrotropy mechanism and a class of hydrotropes that allow the specific selection of cations and anions for the optimization of extraction processes.

2.1.3. Deep Eutectic Solvents

The deep eutectic solvents (DES) were introduced by Abbott *et al.* (ABBOTT *et al.*, 2004) and later presented as a combination of the type (Cat⁺X⁻zY), where X is usually a Lewis base, for instance, a halide ion, and Y a Lewis or Brønsted acids. That mixture should present a much lower melting point compared to the starting pure compounds melting temperatures. The DES are easy to prepare from the complexation of a halide

salt, which acts as a hydrogen-bond acceptor (HBA) as choline chloride, and a hydrogen-bond donor (HBD), such as urea, glycerol, and carboxylic acids, among others (MAUGERI; MARIA, 2012). In comparison to ILs, the DES formulation is low-cost and can reach high purity (DAI *et al.*, 2013). It is also possible to obtain DES through natural compounds such as organic salts, amino acids, sugars alcohols, sugars, choline, betaine, and urea, that result in the so-called Natural Deep Eutectic Solvents (NADES) (AROSO *et al.*, 2017; CRAVEIRO *et al.*, 2016; DAI *et al.*, 2013; PAIVA *et al.*, 2014).

The properties of NADES with water were studied since many times DES have high viscosities, which makes it difficult to perform the mass transfer, and solvent transfer operations are time-consuming (AROSO *et al.*, 2017). Dai *et al.* (DAI *et al.*, 2015) studied the effect of water on the viscosity and other properties of choline chloride, or betaine, based NADES. The use of DES in aqueous systems allows greater applicability in the extraction and solubilization process. DES have been increasingly applied in the extraction processes of bioactive compounds, such as phenolic compounds, polysaccharides, and proteins (DAI *et al.*, 2016; GARCÍA *et al.*, 2016; ZAINAL-ABIDIN *et al.*, 2017).

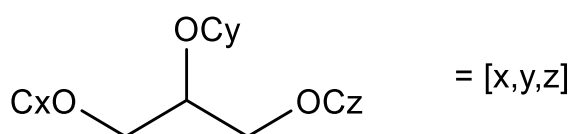
To the best of our knowledge, the first work applying DES as hydrotropes was developed by Soares *et al.* (SOARES *et al.*, 2017). In this work, monomeric lignin compounds in aqueous solutions of DES were dissolved, and these solvents presented a high solubility power for organosolv and kraft lignins since both the hydrogen bond donor and the hydrogen bond acceptor synergistically facilitate the solubilization enhancement. Dynamic light scattering measurements confirmed the hydrotropic mechanism as the driving force for the monomeric lignin model compounds solubilization.

A few other works which relate the increases in the solubility of hydrophobic substances in aqueous solutions using DES are reported in the literature (LI *et al.*, 2019; MOKHTARPOUR *et al.*, 2019; SHEKAARI *et al.*, 2018). However, an integrated approach to study these mixtures as hydrotropes, by investigating the solubility enhancement aiming at the extraction of valuable compounds has never been systematically evaluated. There is still a gap to be filled in the study of NADES as hydrotropes, extolling the potential of DES as green solvents for sustainable extraction processes.

2.1.4. Glycerol Derivatives

Among the derivatives of glycerol, glycerol ethers, are interesting class of hydrotropes (QUESTE *et al.*, 2006a). They are amphiphilic compounds, which contain the glycerol groups in their structure, which confer high solubility in water and allow the use of long alkyl chains that will aggregate at low concentrations and interact more strongly for the solubilization of hydrophobic compounds. Besides that, they are miscible in water and many other organic solvents, which gives them a certain hydrotropic character (QUESTE *et al.*, 2006a, 2007). Another main advantage of their use as bio-based solvents is also the possibility to tune their physicochemical properties by changing the number, size, and nature of their alkyl groups (LEAL-DUASO *et al.*, 2017a). The mono/di/trialkyl glycerol ethers are abbreviated by [x,0,0], [x,y,0]/[x,0,z], and [x,y,z], respectively, according to Figure 2.5.

Figure 2.5. Abbreviated nomenclature for mono/di/trialkyl glycerol ethers.

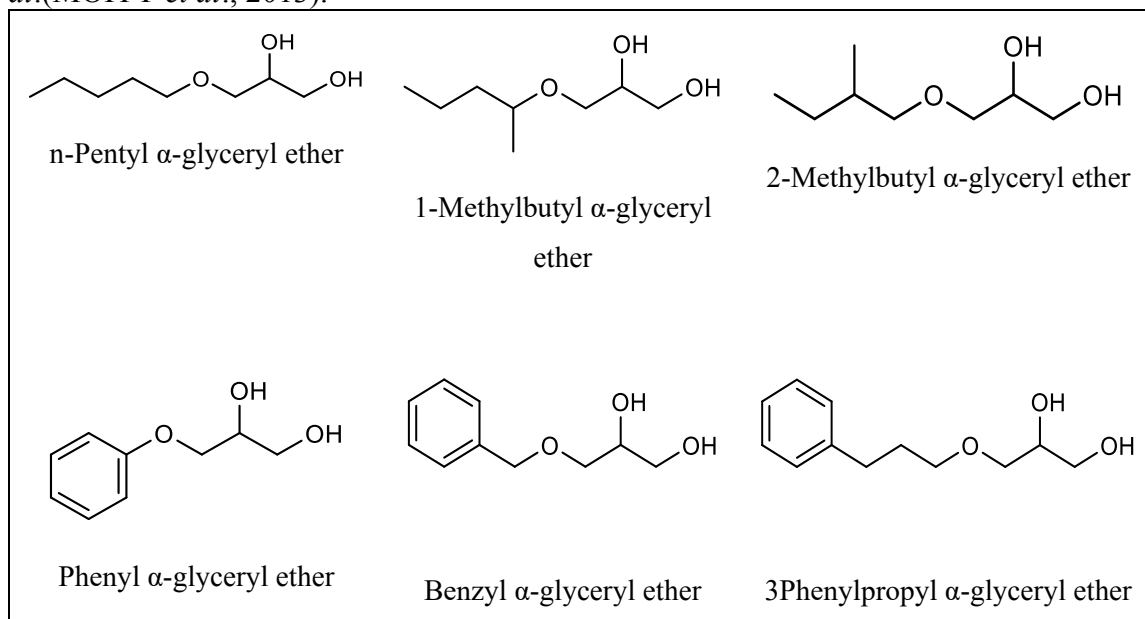


Since glycerol is an abundant by-product of biodiesel production (LEONETI *et al.*, 2012), its use also includes the valorization of unavoidable waste biomass generated, one of the principles of circular economy (SUTTER *et al.*, 2015). Besides being abundantly available, the glycerol ethers can be synthesized through a green synthetic pathway (LEAL-DUASO *et al.*, 2017a, 2017b), and they are a promising alternative to commonly used solvo-surfactants such as ethylene or propylene glycol ethers, which are produced from the petrochemical industry. Glycerol ether shows similar properties to glycol ethers, and despite that the glycerol monoethers are degradable at lower concentrations, they present lower vapor pressures and higher boiling points than their glycol counterparts, and also volatility much lower than common organic solvents (cyclohexane, acetone, and dimethylformamide) (QUESTE *et al.*, 2007).

A few works have been developed regarding monoglycerol ethers as hydrotropes. Moity *et al* (MOITY *et al.*, 2013) prepared six alkyl and aryl monoglycerol ethers via esterification from glycerol (see Figure 2.6), which were investigated as hydrotropes. A comprehensive approach to the thermodynamic properties of the

compounds was evaluated by the authors, including studies related to the determination of octanol/water partition coefficient (obtained by COSMO-RS), volatility (by thermogravimetric analysis), DSC experiments, determination of phase diagram in water, surface tension and self-diffusion coefficient measurements, solubilization curves (for hydrophobic dye), and Fish plots from ternary hydrotrope/oil/water systems.

Figure 2.6. Alkyl and aryl monoglycerol ether structures studied in the work by Moity *et al.* (MOITY *et al.*, 2013).



Lebeuf *et al.* (LEBEUF *et al.*, 2016) have studied the extraction potential of mono, di, and tri-alkyl glycerol ethers, evaluating their thermodynamic properties such as the solubilization of Disperse Red 13 hydrophobic dye, their volatility from DSC measurements and COSMO-RS predictions, as well as their aqueous solubility from the phase diagram, and the presence or absence of cloud points, rationalizing information for a good solute recovery as well as hydrotrope recycling. The temperature binary phase diagrams for hydrotrope/water mixtures present solubility limits for some of the glycerol ethers, since some are not completely water-soluble, such as [6.0.0]. The di-alkyl ethers [4.0.1] and [4.1.0] have the highest dye solubilization potential compared to the others glycerol ethers and also compared to sodium xylene sulfonate (a typical hydrotrope). These compounds also showed a cloud point, suggesting the application of the hydrotropic cloud point extraction method as proposed by the authors.

The information available in the literature for the measurement of thermodynamic properties of glycerol ethers in a hydrotropic approach are scarce, and

the works of Moity *et al.* (MOITY *et al.*, 2013) or Lebeuf *et al.* (LEBEUF *et al.*, 2016) bring suggestions for the characterization of these compounds and are a tool for the study and application of glycerol ethers as hydrotropes in extraction processes or other industrial purposes (FRIBERG; BLUTE, 2006).

Recently, Sutter *et al.* (SUTTER *et al.*, 2015) have reviewed the synthesis of glycerol ethers. These compounds can be synthesized either directly from glycerol. However, as reported in the literature (CHANG *et al.*, 2012; GAUDIN *et al.*, 2011; GU *et al.*, 2008; LIU *et al.*, 2014; PARIENTE; TANCHOUX; FAJULA, 2008), it is difficult to perform reactions with good selectivities due to the formation of mono/di/triesters of glycerol, even using Brönsted and Lewis acids as catalysts with different alcohols as initial etherification reactants.

To the best of our knowledge, Liu *et al.* (LIU *et al.*, 2014) presented the best results so far for the reaction between glycerol and an alcohol. They achieved selectivities of 70% for glycerol ethers using a homogeneous Lewis catalyst, such as Bi(OTf)₃, starting from butanol alcohol and an excess of 4:1 of glycerol at 423.2 K. Veiga *et al.* (VEIGA *et al.*, 2017) described the same selectivity for the reaction using USY, HZSM-5, and H-Beta zeolites as catalysts, starting from tert-butyl or ethyl alcohols. The good results were achieved using high reaction temperatures and an excess of glycerol, but the high viscosity of these mixtures makes separation processes difficult at the end of the reaction.

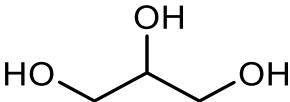
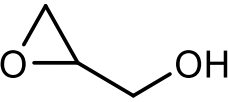
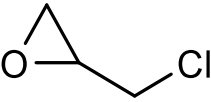
Some industrial building blocks, such as glycidol, epichlorohydrin, or 3-chloropropane-1,2-diol can also be the precursors, which can be produced from glycerol. These starting compounds have shown greater conversions and selectivities for the target products (SUTTER *et al.*, 2015). Cucciniello *et al.* (CUCCINIELLO *et al.*, 2016) and Ricciardi *et al.* (RICCIARDI *et al.*, 2017) described conversions, and product selectivities, over 90% from glycidol using Al(OTf)₃ or Bi(OTf)₃ as a catalyst. A very low catalyst loading of 0.01 mol % was used, and the reaction was carried out at 353.2 K.

In 2017, Pires and co-workers (LEAL-DUASO *et al.*, 2017b) presented the synthesis of glycerol ethers from glycidol and several alcohols in the presence of both homogeneous or heterogeneous basic catalysts obtaining excellent yields and conversions of 93% without the need of glycidol excess. Aiming at enhancing the sustainability of the process, one year later the same group described a green methodology using heterogeneous acid catalysts such as Nafion NR50 and K10-Montmorillonite, using low

catalyst loading (between 1 and 5 % mol) and experiments carried out at room temperature (PIRES *et al.*, 2018).

For the synthesis of glycerol diethers, epichlorohydrin has been described as the building block, used as a precursor of the reactions. Al-Lal *et al.* (AL-LAL *et al.*, 2012) described the synthesis of 1,3-di-tert-butoxypropan-2-ol from epichlorohydrin using $\text{BF}_3\text{Et}_2\text{O}$ as the catalyst. The process has three steps and includes the transformation of epichlorohydrin from glycerol. In the paper by García *et al.* (GARCÍA *et al.*, 2010) published in 2010 it is also described the use of glycidol for the formation of glycerol monoethers and the synthesis of symmetrical di and tri ethers from epichlorohydrin with basic catalysts. Figure 2.7 summarizes the main aspects of the reaction related to each precursor: glycerol, glycidol, and epichlorohydrin.

Figure 2.7. Comparison between the precursors for the glycerol ethers synthesis: glycerol, glycidol and epichlorohydrin.

 Glycerol	 Glycidol	 Epichlorohydrin
by-product low selectivity formation of mono/di/triesters high excess of glycerol high reaction temperatures	synthesized high selectivity high conversion low catalyst loading medium reaction temperatures	

Although glycerol is a by-product of chemical reactions, mainly from biodiesel production (LEONETI *et al.*, 2012), the use of glycidol and epichlorohydrin (obtained through glycerol) shows greater selectivity and conversion for glycerol ethers synthesis, achieved at lower temperatures and low catalyst loading.

2.1.4.1. Alkanediols from glycerol and others

Alkanediols are a class of di-alcohols widely used in the cosmetic, food, and pharmaceutical industries that can provide tunable solvent properties since they offer the opportunity to systematically study the combined effects of hydrophilic and hydrophobic interactions varying the number density of hydrogen-bonding sites and inert groups

(WANG; POTTEL; KAATZE, 1997). Among them, there are two alkanediols produced directed from glycerol namely, 1,2-propanediol (propylene glycol) and 1,3-propanediol (WOLFSON *et al.*, 2012).

Propylene glycol is commonly obtained through hydrogenolysis of glycerol, promoted by heterogeneous catalysts (BEHR *et al.*, 2008; GARCÍA; GARCÍA-MARÍN; PIRES, 2014). However, this chemical synthesis requires a large investment and generates a high flow of polluting waste. In the last years, some attractive biological processes using anaerobic microorganisms have been developed for the production of propylene glycol from the renewable resource at low prices and high quality (SAXENA *et al.*, 2010). 1,2-propanediol is considered a GRAS solvent (generally recognized as safe) by the US Food and Drug Administration (FDA) and is often used as an additive in nutritional products and cosmetics, solvent colourings, and flavours (BEHR *et al.*, 2008).

1,3-propanediol is an important compound in polymer production, and it is a raw material for the production of biodegradable plastics, films, solvents, adhesives, detergents, cosmetics, and pharmaceuticals. 1,3-propanediol is usually prepared from glycerol by fermentation and other biotechnological processes using natural and/or genetically engineered microorganisms (BEHR *et al.*, 2008; GARCÍA; GARCÍA-MARÍN; PIRES, 2014).

The 1,2, 2,3 and 1,4-butanediols can be produced as glycerol/ethanol surplus derivatives from biofuel processing by microbial fermentation, and the most common microorganism involved in the bioconversion are *Saccharomyces cerevisiae*, *K. pneumoniae*, *C. butyricum*, *E. coli* among others (ALMEIDA; FÁVARO; QUIRINO, 2012; ROSALES-CALDERON; ARANTES, 2019). 1,2-butanediol and 1,4-butanediol are compounds in the manufacture of polymers, solvents, and chemicals. 1,2-butanediol is also used to produce adhesive resins, or as a coolant, refrigerant, and hydraulic fluid. Its isomer 2,3-butanediol is used to manufacture printing inks, perfumes, fumigants, moistening and softening agents, explosives, plasticizers, foods, and medicinal (ROSALES-CALDERON; ARANTES, 2019).

The 1,2-alkanediol family is interesting to look at due to the increasing alkyl-chain length and to analyze also the impact of the OH groups' proximity. Besides 1,2-propanediol, 1,2-butanediol already mentioned, 1,2-pentanediol, 1,2-hexanediol, 1,2-octanediol, or 1,2-decanediol are frequently used in cosmetic applications due to their excellent moisturizing properties, they are soluble both in water and cosmetic esters (LEE

et al., 2011). These 1,2 and the 1,n-alkanediols (such as 1,3-propanediol, 1,5-pentanediol, and 1,6-hexanediol) are ingredients considered safe (up to certain concentrations) by FDA for cosmetic formulations (BELSITO *et al.*, 2014; JOHNSON *et al.*, 2012). Additionally, 1,2-alkanediols show increasing anti-microbial activity as their alkane chain length increases (LEE *et al.*, 2011).

In addition to industrial applications of alkanediols, there are numerous works concerning the use of alkanediols-based DES for the study of their properties and applications as the extraction and recovery of bioactive compounds from natural sources (RUESGAS-RAMÓN; FIGUEROA-ESPINOZA; DURAND, 2017; WAN MAHMOOD *et al.*, 2019; WANG *et al.*, 2019; WOJEICCHOWSKI *et al.*, 2021).

2.2. THE KIRKWOOD-BUFF THEORY

How the mechanism of action of hydrotropes works? The main hypotheses addressing this question involve the complex formation (π - π interactions), water structure breaking, and the self-aggregation of the hydrophobic compound, as mentioned before. But to deepen the analysis of the phenomenon, thermodynamic models have been described based on the Kirkwood-Buff (KB) theory (KIRKWOOD; BUFF, 1951), capable of relating properties such as solubility, partial molar volume, and water activity. In the KB theory, the radial distribution function (RDF) is the basis to give the physical-chemistry picture of the solution. RDF represents how many hydrotrope molecules are present at different distances of the solute in favorable and unfavorable interactions (ABBOTT; BOOTH; SHIMIZU, 2016). The RDF between species i and j designated by $g_{ij}(r)$ depends on the intermolecular separation, r . By integrating the RDF over the whole solution space, three scenarios can be found: zero, if the distribution has no special interactions, positive for strong interactions, and negative for systems with unfavorable interactions (ABBOTT; BOOTH; SHIMIZU, 2016).

Among the mechanisms that influence the solubilization of a substance in water is the ability of an ion to cause the effects of salting-in and salting-out a solute, which is classified according to the Hofmeister series. The mechanism of the Hofmeister series is also not clear (KUNZ; NOSTRO; NINHAM, 2004). In the work of Shimizu and McLaren (SHIMIZU; MCLAREN; MATUBAYASI, 2006), Kirkwood-Buff theory was the main tool for correlating partial molar volume data of bovine serum albumin (BSA), at infinite protein dilution, in the presence of a wide range of cosolvents. The KB model allows the

measurement of the strength of the protein-water and protein-cosolvent interactions and the relation between these forces and the Hofmeister series. This statistical-mechanical analysis proposed by Shimizu and McLaren (SHIMIZU; MCLAREN; MATUBAYASI, 2006), starting from the Kirkwood-Buff integrals, are represented by Equations 2.1 and 2.2 (SHULGIN; RUCKENSTEIN, 1999):

$$G_{ij} = \int_0^\infty (g_{ij} - 1) 4\pi r^2 dr \quad (2.1)$$

$$v_{21}^c = \left(\frac{\partial n_1}{\partial n_2} \right)_{T,p,\mu_1} = n_1 (G_{21} - G_{13}) \quad (2.2)$$

where g_{ij} is the radial distribution integral function between species i and j , r is the distance between the centers of the two species, T is the absolute temperature and p is the pressure. Considering a solution that consists of water (1), protein (2), and cosolvent (3), G_{21} and G_{13} are the measure of the strength of the protein-water and water-cosolvent interactions. Concerning the v_{21}^c parameter, which is the so-called concentration-based preferential hydration parameter determined by dialysis or sedimentation equilibrium, is related to the KB parameters and with the density in molarity (n_i) of component i and its chemical potential (μ_i). When the cosolvents are of small size G_{21} can be simply calculated by $G_{21} = v_{21}^c/n_1$ while for G_{23} for the following relation (2.3) holds:

$$\bar{V}_2 = -n_1 \bar{V}_1 G_{21} - n_3 \bar{V}_3 G_{23} \quad (2.3)$$

where $\bar{V}_i$ is the partial molar volume of component i , which can be determined experimentally. From G_{21} and G_{23} is possible to calculate v_{21} , which expresses the effect of the presence of cosolvents on the chemical potential of the solute using Equation 2.4:

$$v_{21} = - \left(\frac{\partial \mu_2}{\partial \mu_1} \right)_{T,P,n_2} = n_1 (G_{21} - G_{23}) \quad (2.4)$$

being v_{21} chemical potential-based preferential hydration parameter, which is a way to quantify the Hofmeister series. Positives v_{21} for salts generally tend to stabilize proteins while negatives v_{21} generally tend to denature them. Protein-excluded partial molar volumes originate G_{21} and G_{23} negatives.

But how can the KB model describe the hydrotropic behavior? Booth *et al.* (BOOTH *et al.*, 2015) from experimental data on the solubilization of some drug models compounds, using nicotinamide and urea as hydrotropes, applied also the fluctuation solution theory (FST) to clarify some of the hypotheses behind the hydrotropy phenomena. Besides data on drug solubility dependence on hydrotrope concentration, activity coefficients, and density, it was possible to determine the preferential drug-hydrotrope interaction. Starting from Equation 2.1 and combining with FST, the authors could correlate the parameters into the following Equations 2.5 and 2.6:

$$G_{ij} = RT\kappa_T \frac{-\bar{V}_i \bar{V}_j (n_i + n_j)}{D} \quad (2.5)$$

where R is the ideal gas constant, κ_T is the isothermal compressibility of the solution, and D is defined as:

$$D = x_i \left(\frac{\partial \ln a_i}{\partial x_i} \right)_{T,P} \quad (2.6)$$

being a_i the activity of species i , and x_i is its mole fraction. From the activity, and partial molar volume $\bar{V}_i$ of species i , it is possible to obtain KB parameters from Equation 2.7:

$$n_3 \left(\frac{\partial \mu_3}{\partial \mu_3} \right)_{T,P,n_2 \rightarrow 0} = \frac{RT}{1 + n_3 (G_{33} - G_{31})} \quad (2.7)$$

From these equations and the literature data, the authors concluded that water activity depression increases the solubilization efficiency. In addition, KB parameters are capable

of describing the hydrotrope self-association, which reduces the hydrotrope efficiency per mol. Shimizu and co-authors (ABBOTT; BOOTH; SHIMIZU, 2016) also described how the solubility is correlated to Kirkwood-Buff Integrals (KBI) by Equation 2.8:

$$\left(\frac{\partial \ln c_2^*}{\partial c_3}\right)_{T,p,c_2 \rightarrow 0} = \frac{(G_{23} - G_{21})}{1 + c_3(G_{33} - G_{31})} \quad (2.8)$$

relating the solute molar solubility (c_2^*) with respect to the hydrotrope concentration (c_3).

Considering the complex mathematical form of the KB integrals what makes it difficult to be determined, and also due to the unexplained origin of the solubilisation plateau in the hydrotrope solubilization curves Shimizu and Matubayasi, in 2016, proposed a simple theory based on statistical thermodynamics (henceforth named cooperative hydrotropy model) to describe hydrotropy (SHIMIZU; MATUBAYASI, 2016). This theory enunciates that hydrotropy occurs due to the water-mediated aggregation of hydrotropes around the solute. The mathematical expression of the model can be expressed as:

$$\frac{x_S}{x_{S,0}} = \frac{1 + e^{b \left(\frac{x_S}{x_{S,0}}\right)_{max}} x_H^m}{1 + e^b x_H^m} \quad (2.9)$$

or in the linearized form

$$\ln \left[\frac{1 - \frac{x_S}{x_{S,0}}}{\frac{x_S}{x_{S,0}} - \left(\frac{x_S}{x_{S,0}}\right)_{max}} \right] = m \cdot \ln(x_H) + b \quad (2.10)$$

where x_S is the solute saturation mole fraction (solubility) in the hydrotropic system, $x_{S,0}$ is the solute saturation mole fraction in water and x_H is the mole fraction of the hydrotrope. From the definition of x_S and $x_{S,0}$ the term $x_S/x_{S,0}$ represents the relative solubility in mole fraction basis. As such, $(x_S/x_{S,0})_{max}$ (henceforth *max*) is the maximum of the relative solubility caused by a given hydrotrope, i.e. the value of the plateau in the sigmoidal solubility curve. Finally, m and b are parameters that give insights into the

molecular interactions between the solute and the hydrotrope. More specifically, m corresponds to the number of hydrotrope molecules involved in the solvation of solute, and b represents the facility of inserting m hydrotrope molecules in the vicinity of the solute (SHIMIZU; MATUBAYASI, 2016).

The Setschenow constant (SETSCHENOW, 1989), that quantifies the change in the solubility of a solute due to the presence of an hydrotrope in the dilute region can be defined as:

$$\ln(S_s) = K_H \cdot C_H \quad (2.11)$$

where S_s is the molar solubility of the solute, K_H is the Setschenow constant and C_H is the molarity of the hydrotrope. Equation 2.11 is valid from a hydrotrope molarity of zero up to a value where the variation of the natural logarithm of the solubility of the solute remains linear with the increase in the molarity of the hydrotrope, usually in a very limited diluted range.

Besides being useful to quantify the hydrotropic power of a substance (albeit in the dilute region), Setschenow constants can also be linked to statistical thermodynamics. The Setschenow constants calculated as per Equation 2.11 are related to Kirkwood–Buff Integrals (KBI) through the following expression (ABBOTT; BOOTH; SHIMIZU, 2016):

$$K_H = G_{S,H} - G_{S,W} \quad (2.12)$$

where $G_{S,H}$ is the KBI between solute and hydrotrope and $G_{S,W}$ is the KBI between solute and water. Equation 2.12 shows that the higher the Setschenow constant is, the higher the preference of the solute to interact with the hydrotrope instead of with water, and consequently, the solubility enhancement of the solute is higher.

Aiming to quantify the interactions between the species, the Kirkwood-Buff theory and the derived cooperative hydrotrope model and Setschenow constants allow predicting the relative contributions of each component (hydrotrope and solute) to the process of solubilization. In this sense, the models proposed above help interpret and rationalize the experimental data, and how thermodynamic properties can be used to express the hydrotropic behavior in different systems.

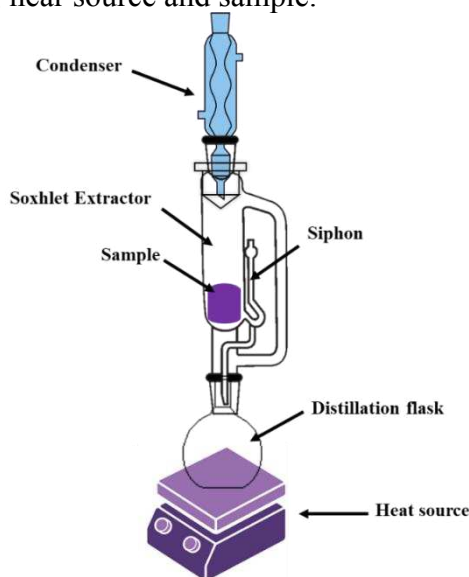
2.3. EXTRACTION PROCESS FOR PHENOLIC COMPOUNDS

Biorefinery can be defined as the use of biomass for conversion into energy and other chemical products, that is mainly biofuels and high-value-added compounds such as antioxidants (CHERUBINI, 2010; SÁNCHEZ-CAMARGO *et al.*, 2017). Biorefinery is an important and fundamental concept for the transition from a linear to a sustainable circular economy (CLARK *et al.*, 2016). In this context, the extraction of high-value compounds from natural matrices, such as phenolic compounds, can be performed with alternative solvents and innovative technologies, producing co-products instead of waste materials (SÁNCHEZ-CAMARGO *et al.*, 2017).

Conventional techniques used for the extraction of bioactive compounds from biological matrices include Soxhlet extraction, maceration, and infusion, among many others. These techniques extract in low yield, higher time consumption, and hazardous nature of solvents used such as dichloromethane and methanol (NN, 2015).

The most classical technique of leaching extraction was developed in 1879 by von Soxhlet and is known by its name, Soxhlet extraction. This technique has been considered a standard of leaching techniques. Since then many other methodologies have been optimized and implemented, but it is still being considered a validation model for the new techniques (CASTRO; PRIEGO-CAPOTE, 2010). The conventional Soxhlet consists of placing the sample in a thimble holder that is gradually filled with the extraction solvent from a distillation flask (Figure 2.8). A siphon aspirates the solute from the thimble-holder when the liquid reaches the overflow level, then unloads it back to the distillation flask carrying the extracted compounds into the bulk liquid, continuously repeating the operation until extraction is complete (CASTRO; PRIEGO-CAPOTE, 2010).

Figure 2.8. Conventional Soxhlet apparatus containing condenser, Soxhlet extractor, siphon, distillation flask, hear source and sample.



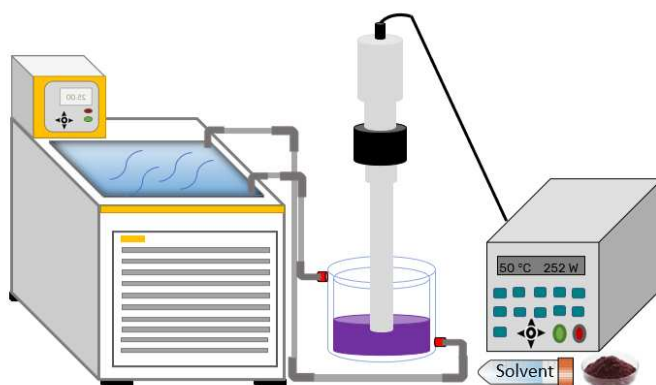
The extraction methods and the solvents to be used must be carefully selected according to the properties of the organic matrix and the target substances, aiming to provide an adequate balance to enhance yield and selectivity (SÁNCHEZ-CAMARGO *et al.*, 2017). Novel methods such as microwave-assisted extraction (MAE), (LIDIA *et al.*, 2017; VINATORU; MASON; CALINESCU, 2017) ultrasound-assisted extraction (UAE) (SHIRSATH; SONAWANE; GOGATE, 2012), supercritical fluid extraction (SFE) (BALLESTEROS-VIVAS; MENDIOLA; IBÁÑEZ, 2019; RUI; ROCHA-SANTOS; DUARTE, 2016) and pressurized liquid extraction (PLE) (NIETO *et al.*, 2010; VAZQUEZ-ROIG; PICÓ, 2015) have been developed aiming to increase yield at low cost, exploiting the wide range of benefits found in natural products. The most commonly used solvents for all these processes are ethanol, methanol, water, or an aqueous mixture of either ethanol or methanol (KAUR *et al.*, 2016; OROIAN; ESCRICHE, 2015; SILVA; ROCHA-SANTOS; DUARTE, 2016).

MAE utilizes microwave energy to facilitate particle breakdown into the solvent from the sample matrix. Microwave radiation interacts with the polar components of the matrix, generating heat on the surface of the sample (LIDIA *et al.*, 2017; VINATORU; MASON; CALINESCU, 2017). The heating of microwave energy acted directly on the molecules by ionic conduction and dipole rotation, both using the electric field to influence the migration of the charge carriers and alternate the movement of polar molecules to line up them. This targeted and selective effect over some compounds is

based on their dielectric constant or polarizability (CHAN *et al.*, 2011; WANG *et al.*, 2016; ZHANG; YANG; WANG, 2011).

UAE increases the surface contact between solvents and samples and allows the disruption of the plant cell wall in the case of plants matrices, increasing the mass transport of the solvents to the sample (SHIRSATH; SONAWANE; GOGATE, 2012). For UAE (see Figure 2.9), the cavitation generated by liquid bubbles collapse is an important driving force in the extraction since the cavitation results in extremely high-speed microjets of solvent that reach the vegetal matrix, facilitating the penetration of the solvent for swelling and solvation process (VINATORU, 2001; VINATORU; MASON; CALINESCU, 2017).

Figure 2.9. Scheme of the UAE apparatus composed of a titanium probe sonifier immersed in a covered glass balloon connected to a thermostatic bath.

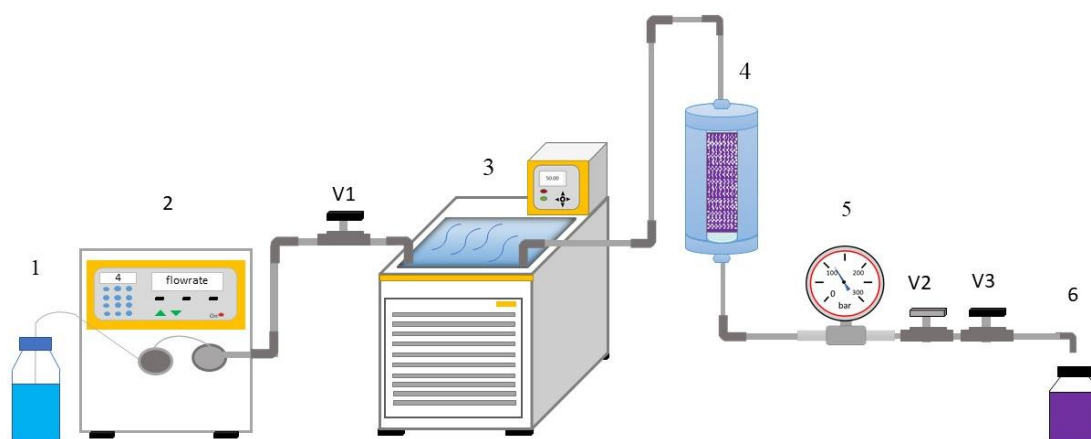


SFE is performed with supercritical fluids as solvents, such as CO₂, ethane, methanol, butene, or CO₂, with co-solvents, combining physical properties of both gas and liquid phases at its critical point to achieve high solubilization and diffusivity of the solute to improve the mass transfer. The most commonly used solvent for SFE is CO₂, basically for three reasons: it is an innocuous compound from the direct human health point of view, has a moderate critical temperature of 304.4 K (fundamental for the conservation of bioactive compounds), and preserves the extract from contact with the air avoiding oxidative reactions (BALLESTEROS-VIVAS; MENDIOLA; IBÁÑEZ, 2019; RUI; ROCHA-SANTOS; DUARTE, 2016; SILVA; ROCHA-SANTOS; DUARTE, 2016).

Finally, the PLE method employs organic or aqueous solvents at high pressure and temperature, which destroys the hydrogen-bond structures of the solvents, usually

water and ethanol, reducing the polarity of the solvent that allows a better dissolution of non-polar substances (NIETO *et al.*, 2010; VAZQUEZ-ROIG; PICÓ, 2015). The potential of PLE is correlated to (i) facilitate the disruption of analyte-matrix by exerting pressure, (ii) increase the solubility, and (iii) improve the mass transfer by the use of liquid solvents at temperatures above their atmospheric boiling point (MUSTAFA; TURNER, 2011; TURNER, 2006). Figure 2.10 shows an example of a self-assembled PLE unit.

Figure 2.10. Scheme of the self-assembled PLE unit: 1. Solvent reservoir; 2. HPLC pump; 3. Thermostatic bath; 4. Jacketed extraction cell; 5 Manometer; 6. Collection flask; V1 and V3. Regulator needle valve; V2. Back pressure needle valve.



The optimization and design of new methods using environment-friendly solvents and the use of consecutive extraction techniques to recover different fractions from the raw materials are being proposed continuously as in the work by Mazzutti *et al.* (MAZZUTTI *et al.*, 2018), in which a multi-task apparatus combining supercritical carbon dioxide (sc-CO₂) and pressurized liquid extraction with ethanol to recover phenolic compounds from cocoa bean hulls was developed. Aguiar *et al.* (AGUIAR *et al.*, 2019) also studied a sequential high-pressure extraction to recover bio-actives from *biquinho* peppers using the same techniques sc-CO₂ and PLE with ethanol and water, considered green solvents.

High-pressure extraction processes such as PLE and SFE have been showing extraction yields, total phenolic content, and antioxidant activities higher than the low-pressure techniques such as Soxhlet and UAE, as demonstrated in the work by Santos *et al.* (SANTOS *et al.*, 2019), for the extraction of bioactive compounds from feijoa peel,

and in the work by Zachová *et al.* (ZACHOVÁ *et al.*, 2018) for the extraction of stilbenes from grape cane using a combination between the methods to obtain the selectivity of SFE and efficiency of extraction with pressurized ethanol.

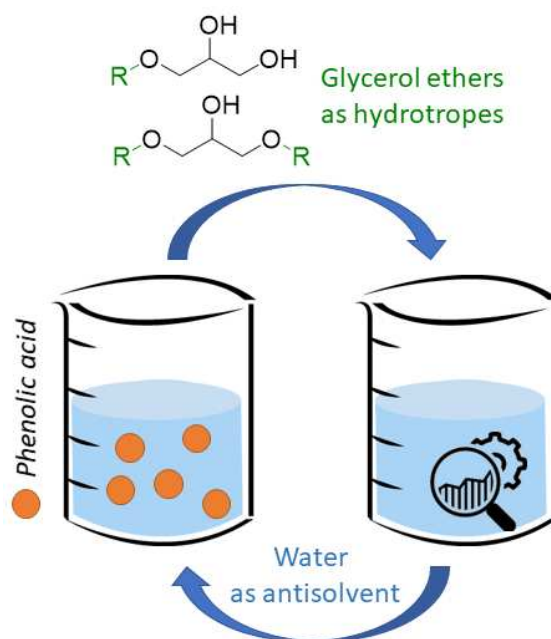
An ultrasound-assisted hydrotropic extraction was proposed by Thakker *et al.* (THAKKER; PARIKH; DESAI, 2018) for selective isolation of geraniol using sodium cumene sulfonate as a hydrotrope. Reduced extraction times and hydrotrope requirements were needed compared to common hydrotropic extraction. Aiming to increase the selectivity of the extraction of bioactive compounds, to reduce the time of extraction, and chemical consumption, hydrotropes are constituents of new efficient solvents to integrate into the conventional extraction methodologies.

3. CHAPTER III - GLYCEROL ETHERS AS HYDROTROPES

3.1. GLYCEROL ETHERS AS HYDROTROPES AND THEIR USE TO ENHANCE THE SOLUBILITY OF PHENOLIC ACIDS IN WATER

Chapter based on the published manuscript:

Bruna P. Soares, Dinis O. Abranches, Tânia E. Sintra, Alejandro Leal-Duaso, José Ignacio García, Elisabet Pires, Seishi Shimizu, Simão P. Pinho and João A. P. Coutinho, Glycerol ethers as hydrotropes and their use to enhance the solubility of phenolic acids in water, ACS Sustainable Chemistry and Engineering, v. 8, n. 14, p. 5742–5749, 2020.



Synopsis: Glycerol ethers are shown in this work to be excellent hydrotropes for the solubility enhancement of gallic and syringic acids in water

3.1.1. Abstract

The use of glycerol ethers (with alkyl side chain ranging from one to six methyl groups) as hydrotropes to enhance the solubility of gallic and syringic acids in water was here studied. These compounds were selected due to their biological and industrial applications and for serving as model molecules for lignin solubilization. The results obtained were compared against traditional co-solvents, demonstrating the exceptional hydrotropic ability of glycerol ethers. Setschenow constants show that the hydrophobicities of both solute and hydrotrope play an important role in the solubility enhancement by hydrotropy, shedding light into its molecular mechanism.

The solubility curves of gallic acid and syringic acid in the aqueous glycerol ether solutions were fitted using a recently proposed statistical thermodynamics-based model. This allowed the estimation of solute recovery from hydrotropic solution by using water as antisolvent. Unlike what is usually claimed it is here shown that in some conditions it is impossible to recover the solute by simply adding water. This analysis paves the way for a rational design and selection of hydrotropes, in which both solubility enhancement and solute recovery are critical parameters to be taken into account.

keywords: Green Chemistry, hydrotropy, phenolic acids, Setschenow, Kirkwood-Buff, cooperativity.

3.1.2. Introduction

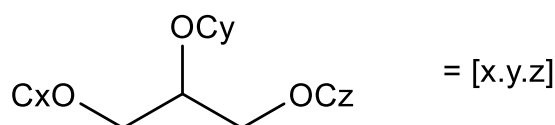
The concept of sustainable chemistry acquired great importance in the design of chemicals and processes less aggressive to the environment. Aiming at designing more sustainable extraction processes, the combination of alternative solvents from renewable and biological sources is an important cornerstone of Green Chemistry (ANASTAS; EGHBALI, 2010; DEVIerno KREUDER *et al.*, 2017), combined preferentially with water, the greener universal solvent (BYRNE *et al.*, 2016; CLARKE *et al.*, 2018; ERYTHROPEL *et al.*, 2018). However, the low solubility in water of many organic compounds, some of which presenting relevant bioactivity, is one of the main shortcomings regarding the use of water to extract valuable compounds (MOTA *et al.*, 2008). Increasing the solubility of poorly soluble substances in aqueous media, typically achievable by the addition of a co-solvent, plays an important role in the purification of bioactive compounds as well as in their formulation and bioavailability studies (SAVJANI; GAJJAR; SAVJANI, 2012; VELEVA; CUE; TODOROVA, 2018).

Hydrotropes are a class of water-soluble compounds with an amphiphilic structure that are able to increase the solubility of hydrophobic substances in water (HODGDON; KALER, 2007). Hydrotropy has been applied in several scientific fields, such as lignin and cellulose dissolution and fractionation, with great success (CAI *et al.*, 2020; CHEN *et al.*, 2017; GABOV; FARDIM; GOMES DA SILVA JÚNIOR, 2013; SOARES *et al.*, 2017, 2019; SONG *et al.*, 2019). Hydrotropes can extend the applicability of water as a solvent to water-insoluble compounds, thus being highly relevant to Green Chemistry. However, since proposed by Neuberg (NEUBERG, 1916) in 1916, most

compounds studied as hydrotropes are petrochemical-based, such as sodium benzene sulfonate (SBS), sodium toluene sulfonate (STS) or sodium xylene sulfonate (SXS). Non-ionic alkyl-hydrotropes such as ethylene glycol ethers and propylene glycol ethers have also been proposed (BAUDUIN *et al.*, 2005; HODGDON; KALER, 2007; LAI, 2005). Recently, glycerol ethers have been shown to behave as hydrotropes, making them a promising bio-based alternative for the commonly petrochemical-based hydrotropes (QUESTÉ *et al.*, 2006b), while possessing lower vapor pressures and higher boiling points than their glycol ethers counterparts (QUESTÉ *et al.*, 2007).

Glycerol ethers are amphiphilic compounds that possess a central hydrophilic glycerol back-bone, conferring them a certain degree of polarity, and apolar alkyl side-chains (GARCÍA *et al.*, 2010; QUESTÉ *et al.*, 2006b, 2007). Besides glycerol being abundantly available (as a by-product of biodiesel production, for example (LEONETI *et al.*, 2012)), glycerol ethers are synthesized from it via a green pathway (SUTTER *et al.*, 2015). Moreover, glycerol ethers may be viewed as *designer molecules*, since it is possible to tune their physicochemical properties by changing the number and size of their alkyl groups (GARCÍA *et al.*, 2010; LEAL-DUASO *et al.*, 2017b, 2017a, 2019; QUESTÉ *et al.*, 2006b). The name of these compounds is usually abbreviated as [x.y.z], where x, y and z, as shown in Figure 3.1, represent the number of carbon atoms of the alkyl chains linked to the oxygens in the three different positions of the glycerol backbone. A value of zero in any of these variables means that there is a proton linked to the oxygen instead of an alkyl chain, i.e. there is a hydroxyl group in that position.

Figure 3.1. Nomenclature for the alkyl glycerol ethers studied in this work.



So far only a few works have been reported regarding the use of monoglycerol ethers as hydrotropes. Moity *et al.* (MOITY *et al.*, 2013) prepared three pentyl and three aryl 1-O-monoglyceryl ethers via esterification from glycerol, all presenting low volatility (vapor pressure below 0.01 kPa), and investigated their potential as hydrotropes. The results obtained show great solubility enhancement of a hydrophobic dye (Disperse

Red 13), especially when using aryl monoglycerol ethers. Lebeuf *et al.* (LEBEUF *et al.*, 2016) have also studied the hydrotropic potential of mono, di and tri-alkyl glycerol ethers for a hydrophobic dye (Disperse Red 13). Some of the compounds studied, such as [2.1.1], [3.1.1], [4.1.1] and [2.2.2], have a solubility limit, not being fully miscible with water, and are also the most volatile. Among the hydrotropes studied, [5.0.0] presents the greater solubilization power at low concentrations (up to 30 wt.%) and possesses the highest boiling point (535.2 K), making it one of the best candidates to be used as an hydrotrope in that case.

The present work investigates the effect of the alkyl side chain of glycerol ethers on the solubility enhancement of two phenolic acids, the poorly water-soluble gallic and syringic acids. They were selected as model compounds for this study due to their relevant bioactivities such as strong antioxidant properties (CHOUBEY *et al.*, 2015; SRINIVASULU *et al.*, 2018), their presence in a wide variety of natural organic matrices and industrial applications (BADHANI; SHARMA; KAKKAR, 2015; SRINIVASULU *et al.*, 2018), and their different levels of polarity, with syringic acid being more hydrophobic than gallic acid (suggested by their different octanol/water partition coefficients (“ChemSpider database. <http://www.chemspider.com>”, [s.d.])). Moreover, gallic and syringic acids are monomers of lignin and excellent model molecules for its solubility in hydrotropic systems, an active area of research (ALZAGAMEEM *et al.*, 2018; CAI *et al.*, 2020; SOARES *et al.*, 2017, 2019). The experimental data obtained was used to better understand the mechanism of hydrotropy, through the calculation of Setschenow constants (SETSCHENOW, 1989). Additionally, a recently proposed thermodynamics model of cooperative hydrotropy, developed by Shimizu and Matubayasi (SHIMIZU; MATUBAYASI, 2016), was used to fit the solubility data, enabling an analysis for the estimation of the recovery ease of the solutes from the hydrotropic systems by using water as anti-solvent.

3.1.3. Material and Methods

3.1.3.1. Chemicals

The chemicals used in this work are displayed in Table 3.1, along with their source and mass purity. Water was double distilled, passed across a reverse osmosis system, and further treated with a Milli-Q plus 185 water purification apparatus. The alcohols were dried and distilled over calcium hydride previous to being used.

Table 3.1. List of substances used in this work along with their CAS number, source and purity.

Substance	CAS Number	Source	Purity (wt%)
Glycidol	556-52-5	Sigma-Aldrich	96
Epichlorohydrin	106-89-8	Sigma-Aldrich	99
Methanol	67-56-1	Scharlab	>99
Ethanol	64-17-5	Scharlab	>99
Sodium Hydroxide	1310-73-2	Scharlab	98
Hydrochloric Acid (37%)	7647-01-0	Fisher	
Propan-1-ol	71-23-8	Alfa Aesar	99.5
Butan-1-ol	71-36-3	Alfa Aesar	>99
Pentan-1-ol	71-41-0	Alfa Aesar	>99
Hexan-1-ol	111-27-3	Alfa Aesar	>99
Calcium Hydride	7789-78-8	Acros Organics	>93.0
Gallic Acid	149-91-7	Merck	>99.5
Syringic Acid	530-57-4	Acros Organics	>98.0
Glycerol	56-81-5	Fisher Chemical	>99.8
[1.0.0]	623-39-2	This work	>98 ^{b)}
[2.0.0]	1874-62-0	This work	>99 ^{b)}
[3.0.0]	61940-71-4	This work	>99 ^{b)}
[4.0.0]	624-52-2	This work	>99 ^{b)}
[5.0.0]	22636-32-4	This work	>99 ^{b)}
[6.0.0]	a)	This work	>99 ^{b)}
[1.0.1]	623-69-8	This work	>99 ^{b)}
[2.0.2]	4043-59-8	This work	>99 ^{b)}

a) Compound has no CAS number available. **b)** Estimated by NMR.

3.1.3.2. Synthesis of Glycerol Ethers

A scale up to 3 mol of the previously described glycerol ether synthesis was carried out (LEAL-DUASO *et al.*, 2017b, 2017a). Thus, in order to synthesize glycerol monoethers, 45 mol of alcohol and sodium hydroxide (20 mol% with respect to glycidol) were placed into a round bottomed flask. The reaction mixture was stirred and heated at 338 K, under argon, until total dissolution of the catalyst (NaOH). Then, glycidol (3 mmol) was added dropwise. The reaction was monitored at different times by extracting samples that were neutralized with 0.3 M HCl previous to injection in a Hewlett Packard 7890 series II GC (Gas Chromatography), as described in section S1 of **Appendix A**. After the total consumption of glycidol, the reaction was quenched with 0.3 M HCl and

the salt formed (sodium chloride) was filtered off. Finally, the excess of the starting alcohol was eliminated by reduced pressure distillation and glycerol monoethers were purified by vacuum distillation.

To synthesize symmetric glycerol diethers, 45 mol of the corresponding alcohol and the catalyst NaOH (4.2 mol), were placed into a round bottomed flask. The mixture was stirred and heated at 338 K under argon until total dissolution of the base. Then, epichlorohydrin (3 mmol) was added dropwise. Reactions were monitored by GC until total consumption of epichlorohydrin. Then, the reaction was also quenched with HCl 0.3 M and salts were filtered off. Finally, the excess of the starting alcohol was distilled and recovered and the resulting glycerol diether was purified by vacuum distillation.

Purity of the products was checked by proton and carbon nuclear magnetic resonance (^1H -NMR, ^{13}C -NMR), recording the spectra in a Bruker Avance 400 MHz device, and using DMSO-*d*6 (dimethyl sulfoxide-*d*6) as solvent (with chemical shifts δ in ppm). All the obtained spectra and the water content of each product (measured using Karl-Fischer titration) can be found in section S1 of the **Appendix A**.

3.1.3.3. Solubility Measurements

The solubility of the phenolic acids (gallic acid or syringic acid) was measured by the analytical isothermal shake-flask method, previously described in detail (MOTA *et al.*, 2008). The phenolic acid was added in excess to each hydrotrope aqueous solution. For the aqueous systems the samples were equilibrated in an air oven at (303.2 ± 0.5) K under constant stirring (950 rpm) and an equilibration time of 72 h, using an Eppendorf Thermomixer Comfort equipment. For the pure glycerol ethers, which are more viscous than their aqueous solutions, the samples were placed over plate stirrers inside a thermostatic water bath at (303.2 ± 0.1) K for 72 h. The equilibration conditions were previously optimized (CLÁUDIO *et al.*, 2015; SINTRA *et al.*, 2018).

After equilibrium was reached (72 h), all samples were centrifuged at (303.2 ± 0.5) K for 20 minutes using a Hettich Mikro 120 centrifuge operating at 4500 rpm, in order to separate the excess undissolved solute from the liquid phase. After centrifugation, all samples were placed in an air bath equipped with a Pt 100 probe and a PID controller at the temperature used in equilibrium assays during 2 h. Then, the samples of the liquid phase were carefully collected and diluted in ultra-pure water, and the amount of phenolic acid was quantified by UV-spectroscopy using a SHIMADZU UV-1700, Pharma-Spec

spectrometer at 262 and 267 nm for gallic and syringic acid, respectively. At least three individual samples were quantified for each system.

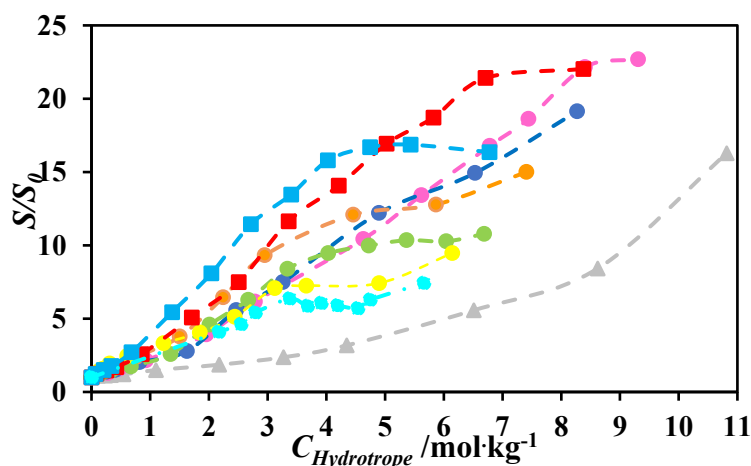
3.1.4. Results and discussion

3.1.4.1. Solubility Curves

The solubility of gallic acid and syringic acid (see Figure S9 for chemical structures) in aqueous solutions of glycerol ethers was measured in the entire concentration range, at 303.2 K, and is reported in section S2 of the **Appendix A** (Tables S2-S18). Since [6.0.0] forms a two-phase system with water (QUESTÉ *et al.*, 2006b) at concentrations below 38 wt% its hydrotropic capability was studied only for gallic acid in the single-phase region.

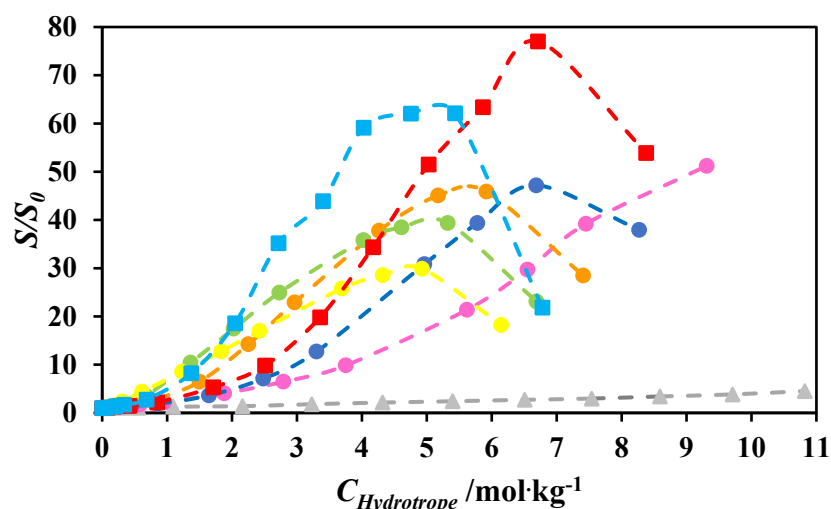
The solubility data for gallic acid is shown in Figure 3.2, where S and S_0 represent the solubility (mol/L) of gallic acid in the aqueous solutions of the hydrotrope and in pure water, respectively. Choosing attainable maximum solubility as the metric of interest, the ability of the linear glycerol ethers to enhance the solubility of gallic acid increases in the following order: [6.0.0] < [5.0.0] < [4.0.0] < [3.0.0] < [2.0.0] < [1.0.0]. Moreover, [1.0.1] is better than [2.0.2] while glycerol shows the least solubility enhancement. This initial analysis suggests that the shorter the alkyl chain of the hydrotrope, the better the solubility enhancement. Note that the maximum solubility of gallic acid in aqueous [1.0.0] or [1.0.1] is about the same, with the plateau being reached at a lower concentration for the [1.0.1] curve.

Figure 3.2. Effect of glycerol ether (hydrotrope) concentration on the solubility of gallic acid in aqueous solutions of [1.0.0] ●, [2.0.0] ●, [3.0.0] ●, [4.0.0] ●, [5.0.0] ●, [6.0.0] ●, [1.0.1] ■, [2.0.2] ■ and glycerol ▲, at 303.2 K. S/S_0 is the relative solubility (expressed in mol/L) of the solute and $C_{\text{Hydrotrope}}$ is the concentration of the hydrotrope in the solvent (solute-free basis). Dashed lines are visual guides.



The solubility data for syringic acid is depicted in Figure 3.3. Contrary to what is seen in Figure 3.2, most of the solubility curves depicted in Figure 3.3 pass through a maximum, with glycerol and [1.0.0] as the exceptions, suggesting an optimal concentration of hydrotrope. Again considering attainable maximum solubility as the metric of interest, the conclusions drawn from Figure 3.2 for gallic acid hold true for syringic acid as well. Hence, it appears that the smaller the hydrotrope the better the solubility enhancement. Interestingly, the increase in solubility of syringic acid is proportionally much more pronounced than that for gallic acid. For instance, it is possible to reach a 77-fold increase in the solubility of syringic acid using [1.0.1] while a 22-fold increase in solubility was achieved for gallic acid with the same hydrotrope.

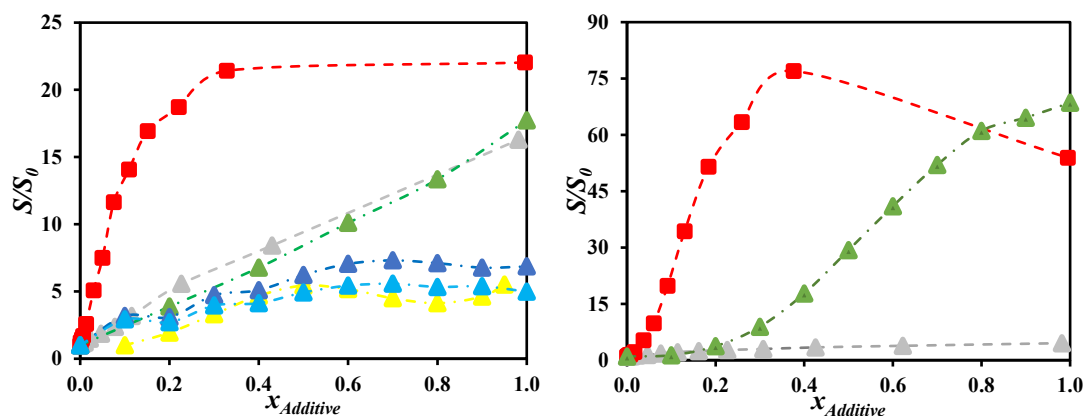
Figure 3.3. Effect of glycerol ether (hydrotrope) concentration on the solubility of syringic acid in aqueous solutions of [1.0.0] $\color{violet}\bullet$, [2.0.0] $\bullet$, [3.0.0] $\color{brown}\bullet$, [4.0.0] $\color{green}\bullet$, [5.0.0] $\color{yellow}\bullet$, [1.0.1] $\color{red}\blacksquare$, [2.0.2] $\color{blue}\blacksquare$ and glycerol $\blacktriangle$, at 303.2 K. S/S_0 is the relative solubility (expressed in mol/L) of the solute and $C_{\text{Hydrotrope}}$ is the concentration of the hydrotrope in the solvent (solute-free basis). Dashed lines are visual guides.



The solubility enhancement of gallic and syringic acids obtained using glycerol ethers as hydrotropes was compared against results using co-solvency with traditional solvents (Figure 3.4). As Figure 3.4 clearly demonstrates, glycerol ethers are much better solubilizing agents for gallic acid than traditional co-solvents such as methanol, acetonitrile and propan-2-ol. Even glycerol, the worst hydrotrope studied in this work, is better than traditional co-solvents. While methanol provides higher solubility values than glycerol in the solubilization of syringic acid, it is much inferior to [1.0.1] in most of the

concentration range. Methanol is, indeed, commonly added to enhance the solubility of hydrophobic substances, but possesses high volatility and toxicity (CHEMAT *et al.*, 2019), contrary to the glycerol ethers studied in this work (GARCÍA *et al.*, 2015; LEAL-DUASO *et al.*, 2017b, 2017a; PERALES *et al.*, 2017). It is interesting to note that the methanol solubility curve for gallic acid presents a linear shape, in contrast with its sigmoidal shape for syringic acid, similar to what is expected in hydrotropy.

Figure 3.4. Solubility enhancement of gallic acid (left panel) and syringic acid (right panel) using the hydrotropes [1.0.1] $\blacksquare$, and [0.0.0] $\blacktriangle$ (this work) and the co-solvents methanol $\blacktriangle$ (NOUBIGH *et al.*, 2013; NOUBIGH; AKERMI, 2017), ethanol $\blacktriangle$ (NOUBIGH *et al.*, 2012) acetonitrile $\blacktriangle$ (DALI *et al.*, 2016) and propan-2-ol $\blacktriangle$ (DALI *et al.*, 2016). S/S_0 is the relative solubility (expressed in mol/L) of the solute and the x-axis represents the mole fraction of the additive (hydrotrope or co-solvent) in the solvent (solute-free basis). Dashed lines are visual guides.



3.1.4.2. Dilute Hydrotrope Region

The pronounced effect of glycerol ethers as hydrotrope agents, when compared to traditional organic solvents, is of great interest for extraction processes, especially in the replacement of traditional volatile organic compounds. However, apart from studying the entire concentration range of hydrotrope, the careful analysis of the dilute hydrotrope region is also important for the following reasons. Firstly, from an application-wise perspective, using smaller quantities of additives (hydrotropes) is economically preferable. Secondly, from a fundamental perspective the identification of the molecular mechanism of hydrotropy is easier in the dilute region where effects can be isolated (contribution from the hydrotrope self-association in the bulk phase can, for instance, be neglected). To study the dilute region the Setschenow constant was used (SETSCHENOW, 1989). Originally this was proposed as an empirical approach to describe the effect of a salt, or co-solvent on the solubility of a compound in aqueous

solution, and has since been applied to describe the effect of hydrotropes on the aqueous solubility of solutes (CLÁUDIO *et al.*, 2015; SINTRA *et al.*, 2018). It has been shown, using statistical thermodynamics, that this approach has a sound physical basis in the dilute region (ABBOTT; BOOTH; SHIMIZU, 2016).

The Setschenow constant (SETSCHENOW, 1989) quantifies the change in the solubility of a solute due to the presence of an hydrotrope, in the dilute region. It is herein defined as:

$$\ln(S_s) = K_H \cdot C_H \quad (3.1)$$

where S_s is the molar solubility of the solute, K_H is the Setschenow constant and C_H is the molarity of the hydrotrope. Equation 3.1 is valid from a hydrotrope molarity of zero up to a value where the variation of the natural logarithm of the solubility of the solute remains linear with the increase in the molarity of the hydrotrope (about 5 wt% for the hydrotropes studied in this work).

Besides being useful to quantify the hydrotropic power of a substance (albeit in the dilute region), Setschenow constants can also be linked to statistical thermodynamics. The Setschenow constants calculated as per Equation 3.1 are related to Kirkwood–Buff Integrals (KBI) through the following expression (ABBOTT; BOOTH; SHIMIZU, 2016):

$$K_H = G_{S,H} - G_{S,W} \quad (3.2)$$

where $G_{S,H}$ is the KBI between solute and hydrotrope and $G_{S,W}$ is the KBI between solute and water. Equation 3.2 shows that the higher the Setschenow constant is, the higher the preference of the solute to interact with the hydrotrope instead of with water and, consequently, the higher the solubility enhancement of the solute.

The Setschenow constants were calculated (assuming density of the systems in the dilute region equal to that of water) for all solute-hydrotrope pairs reported in this work, except for [6.0.0] since solubility data in the dilute region is not available for this compound. These results are reported in Table 3.2. Interestingly, the values obtained are in contradiction with the initial qualitative analysis from Figures 3.2 and 3.3. That is, the

hydrotropic power of glycerol ethers in the dilute region increases with the increase in size of the alkyl side chain, in line with previous studies (LEBEUF *et al.*, 2016): [0.0.0] < [1.0.0] < [2.0.0] < [3.0.0] < [4.0.0] < [5.0.0].

Table 3.2. Setschenow constants for gallic and syringic acids in the glycerol ether hydrotrope systems studied in this work, along with the hydrotrope molarity range considered in their calculation.

	Hydrotrope	Setschenow Constant	Molarity Range /M
Gallic acid	[0.0.0]	0.149	0-0.3
	[1.0.0]	0.802	0-0.5
	[2.0.0]	0.976	0-0.3
	[3.0.0]	1.273	0-0.2
	[4.0.0]	1.528	0-0.2
	[5.0.0]	2.054	0-0.2
	[1.0.1]	1.171	0-0.3
	[2.0.2]	1.560	0-0.2
Syringic acid	[0.0.0]	0.129	0-0.6
	[1.0.0]	0.988	0-0.3
	[2.0.0]	0.759	0-0.3
	[3.0.0]	1.148	0-0.2
	[4.0.0]	1.604	0-0.2
	[5.0.0]	2.443	0-0.2
	[1.0.1]	1.401	0-0.3
	[2.0.2]	1.667	0-0.2

The results reported in Table 3.2 are in agreement with a previous study by Bauduin and co-workers (BAUDUIN *et al.*, 2005) that suggested the apolar volume of an hydrotrope to be directly connected with its capability to enhance the solubility of a solute. In fact, the progressive increase, through the addition of methyl groups, in apolar volume of the glycerol ethers seems to positively correlate with the Setschenow constants obtained, shedding light into the molecular mechanisms of hydrotropy.

Considering Equation 3.2, which has shown that Setschenow constants increase if the KBI of the solute-hydrotrope pair increases or the KBI of the solute-water pair decreases, it makes sense that apolarity or hydrophobicity plays a role in hydrotropy. An increase in the hydrophobicity of the solute should lead to a decrease of its interaction with water, leading to a decrease of the solute-water KBI and a consequent increase in the Setschenow constant. This is exactly what is seen in this work: for the same

hydrotrope, the Setschenow constant obtained for systems containing syringic acid are higher than that of gallic acid (syringic acid has a higher octanol/water partition coefficient than gallic acid, suggesting it is more hydrophobic than gallic acid (“ChemSpider database. <http://www.chemspider.com>”, [s.d.])). On the other hand, increasing the hydrophobicity of the hydrotrope weakens its interaction with water, promoting interaction with the hydrophobic solute, leading to an increase in the solute-hydrotrope KBI, increasing the Setschenow constant, in accordance to what is reported in Table 3.2.

It is not yet clear why there is an inversion on the behavior of the hydrotropic power of glycerol ethers above a certain concentration range, with more hydrophobic hydrotropes being better at low concentrations and more hydrophilic hydrotropes being better at higher concentrations. The Setschenow analysis above reveals that the size of the alkyl chain length appears to be the dominating factor at low concentrations. However, it is expectable that increasing hydrotrope concentration will also increase the activity coefficient of water, as supported by the immiscibility observed for [6.0.0]. That is, increasing the alkyl chain length of the hydrotrope increases its hydrophobicity, leading to a less favorable interaction to water for high hydrotrope concentration. Thus, above a certain concentration of these hydrotropes, the nefarious effect of being too hydrophobic prevails over favorable solute-hydrotrope interactions, leading to a drastic negative impact in the hydrotropic behavior.

3.1.4.3. Modelling

The solubility data obtained in this work was fitted using a statistical thermodynamics-based model developed by Shimizu and Matubayasi (SHIMIZU; MATUBAYASI, 2016). This model based on the cooperativity concept was developed not only to describe the usual sigmoidal solubility curves found in hydrotropy but also to give insight into the interactions between solute and hydrotrope molecules. The model can be expressed as:

$$\ln \left[\frac{1 - \frac{x_S}{x_{S,0}}}{\frac{x_S}{x_{S,0}} - \left(\frac{x_S}{x_{S,0}} \right)_{max}} \right] = m \cdot \ln(x_H) + b \quad (3.3)$$

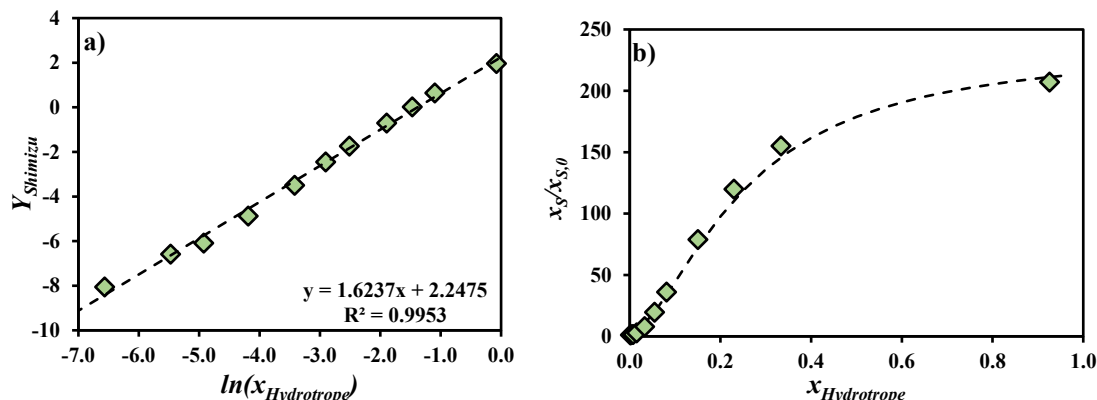
where x_S is the solute saturation mole fraction (solubility) in the hydrotropic system, $x_{S,0}$ is the solute saturation mole fraction in water and x_H is the mole fraction of the hydrotrope. Note that x_H is not the mole fraction of the hydrotrope in a solute-free basis but its mole fraction in the ternary water-hydrotrope-solute system; interconversion between them is done assuming the density of the mixture equal to that of water. From the definition of x_S and $x_{S,0}$ it becomes clear that the term $x_S/x_{S,0}$ represents the relative solubility in mole fraction basis. As such, $(x_S/x_{S,0})_{max}$ (henceforth *max*) is the maximum of the relative solubility caused by a given hydrotrope, i.e. the value of the plateau in the sigmoidal solubility curve. Finally, m and b are parameters that give insight into the molecular interactions between solute and hydrotrope. More specifically, m represents the number of hydrotrope molecules in the vicinity of the solute (SHIMIZU; MATUBAYASI, 2016).

Due to the difficulty of identifying a clear solubility plateau in the many of the systems studied in this work, the parameter *max* was treated as an adjustable parameter of the model. Note that m and b are not adjustable parameters of the model, since they are directly calculated from the experimental data and the *max* parameter. The modelling algorithm used goes as follow. A value is arbitrarily chosen for variable *max*. Then, the m and b parameters are extracted from the experimental data as the slope and intercept of the linearized curve defined as:

$$Y = \ln \left[\frac{1 - \frac{x_S}{x_{S,0}}}{\frac{x_S}{x_{S,0}} - \left(\frac{x_S}{x_{S,0}} \right)_{max}} \right] ; x = \ln(x_H) \quad (3.4)$$

Using the calculated m and b parameters, the experimental data is reproduced using the model and the quadratic error between predicted and experimental data is calculated. Variable *max* is then varied until the sum of the quadratic errors is minimized. This procedure for the application of the model is illustrated in Figure 3.5a) with Equation 3.4 and Figure 3.5b) with the actual fitting, both for the syringic acid-[3.0.0] solute-hydrotrope pair. Fitted curves for all systems herein studied are depicted in section S3 of the **Appendix A** (Figures S10 and S11).

Figure 3.5. Linearized plot of the cooperative hydrotrophy model based on Equation 3.4 (left panel, $\blacklozenge$ experimental data, - - - least squares fit) and fitted experimental data (right panel, $\blacklozenge$ experimental data, - - - cooperative hydrotrophy model) for the water-[3.0.0]-syringic acid system. The x-axis represents the mole fraction of the hydrotrope in the ternary system (as opposed to its mole fraction in the solvent free of solute).



As Figures S10 and S11 show (see section S3 of the **Appendix A**), the model reproduces the experimental data quite well. It is curious to note that the characteristic sigmoidal shape of hydrotropic solubility curves is much more patent in the systems with syringic acid than in the systems with gallic acid. Moreover, it is important to note that expressing the composition of hydrotrope in the actual ternary system instead of its composition in the solvent (solute-free basis) removes the maxima seen in the solubility curves depicted in Figures 3.2 and 3.3. No clear pattern can be identified on the model parameters, which are reported in Table S19 of the **Appendix A**.

The cooperative model employed in this work (Equation 3.3) can be applicable not only to cooperative (sigmoidal) solubility increases but also linear (non-cooperative cases), such as those seen for gallic acid. In the latter case, m becomes close to 1, leading to a very large max variable. Thus, the general applicability of the model is supported by its success in describing both linear and sigmoidal solubility curves.

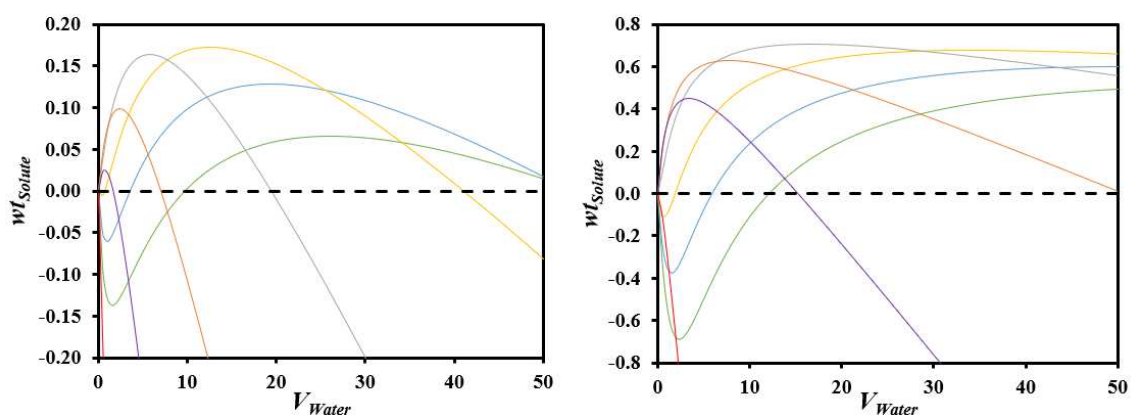
3.1.4.4. Solute Recovery

Besides quantifying their dissolution ability, it is fundamental to address the recovery of solute from hydrotropic solutions. For most organic solvents, a simple evaporation suffices. However, evaporating water from a hydrotropic solution would increase hydrotrope concentration which, generally, would increase the solubility of the solute. Moreover, the hydrotropes are often non, or poorly volatile. There is, however, a clever turnaround that allows for the easy recovery of solute from an hydrotrope solution

with similar energy cost when compared to traditional solvents: the use of water as anti-solvent. As proposed in previous works (DHAPTE; MEHTA, 2015; GAIKAR; SHARMA, 1993; LAI, 2005; MCKEE, 1946), addition of water to an hydrotropic solution may induce the precipitation of the solute due to the dilution of the hydrotrope, providing an easy and straightforward approach to recover the solute in high purity.

Whether this approach to solute recovery is feasible was here evaluated by calculating the recoverable fraction of dissolved solute, using the solubility curves modelling reported in the previous section. The calculation algorithm and detailed results are reported in section S4 of the **Appendix A**. Figure 3.6 illustrates the recovery curves (recovered solute fraction versus water volume fraction added) obtained using the hydrotrope [1.0.1] for gallic acid and syringic acid. Note that a negative solute fraction is possible, meaning that there is no precipitation and the system is no longer saturated, thus, being able to dissolve more solute.

Figure 3.6. Estimated fraction of gallic acid (left panel) and syringic acid (right panel) recovered from hydrotrope solution (w_{Solute}) by the addition of water (V_{Water} is the volumetric ratio between added water and initial system), with an initial hydrotrope mole fraction of 0.01 —, 0.05 —, 0.1 —, 0.2 —, 0.4 —, 0.6 — and 0.8 —. A negative value indicates that no precipitation happens, with the system being able to dissolve further solute.



Surprisingly, addition of water does not always lead to solute precipitation. Considering the examples depicted in Figure 3.6, both hydrotrope and its composition clearly play a role in determining the feasibility of recovering the solute by using water as anti-solvent. For instance, in this case (gallic acid-[1.0.1] system), the solute can only be recovered if the hydrotrope mole fraction is in the 0.05-0.4 range, with a maximum recovery of 17% achieved in the 0.2-0.4 hydrotrope mole fraction range. If the hydrotrope

mole fraction is higher than 0.4, the solute may still be recoverable but only after the addition of a large quantity of water. Below a mole fraction of 0.05, solute recovery is unfeasible. These conclusions are similar for the syringic acid-[1.0.1] system. In this case, the solute can be recovered in a narrower mole fraction window, but up to 70% can be recovered.

Despite the increased volume of water when water is added to a hydrotropic system, the hydrotrope becomes less concentrated, which makes hydrotropy less effective. Whether any amount of solute precipitates from a hydrotrope solution after adding water is determined by the trade-off between these two factors. Thus, the recovery of solute is more favorable as the slope of the solute solubility curve increases, which corresponds to a bigger change in solubility due to a smaller change in hydrotrope concentration. It is also important to note that, since syringic acid is much less soluble in water than gallic acid, it is easier to recover it since the first factor (solute dissolution in the new water volume) loses importance. Moreover, the slope of the solubility curves tends to be higher in mid composition ranges of hydrotrope (sigmoidal shape), explaining the recovery windows positioned in mid hydrotrope mole fractions.

Figure S12 reveals that, in terms of gallic acid recovery, it is better to use a hydrotrope mole fraction of 0.2-0.8 for the most hydrophilic hydrotropes ([1.0.0] and [2.0.0]), whilst a 0.05-0.2 window is better for the least hydrophilic ones. Interestingly, it is impossible (using the addition of water) to recover gallic acid dissolved in aqueous [5.0.0] solutions. The same conclusions hold true for syringic acid, as Figure S13 shows, albeit in narrower mole fraction windows, similarly to what was concluded through the analysis of Figure 3.6. The biggest difference is the fraction of solute recovered, which is much higher for syringic acid than for gallic acid, due to the almost 10-fold difference between their solubility in pure water.

3.1.5. Conclusions

The solubility of gallic acid and syringic acid was measured in aqueous solutions of glycerol ethers, a recently proposed new class of hydrotropes, demonstrating their excellent hydrotropic ability. Their solubilization capacity is more prominent for the smaller, more hydrophilic hydrotropes and is superior to traditional organic co-solvents.

The Setschenow constants for the hydrotropic systems shed light into the hydrotropy mechanism of these systems. The results show that the hydrophobicity of the

hydrotrope plays a major role in hydrotrophy, being dominant in the dilute region. Furthermore, the hydrophobicity of the solute is also important, since its relative solubility enhancement is directly linked to it.

The experimental data herein obtained was fitted using the cooperative hydrotrophy model. It provided appropriate fitting and allowed for the analysis of the feasibility of recovering solute from hydrotropic solutions by the addition of water. It was herein shown that solute recovery is not always possible. As such, the choice of hydrotrope and operating concentration for a given application should consider not only the solubility enhancement provided by the hydrotrope but also the ease of solute recovery from the system.

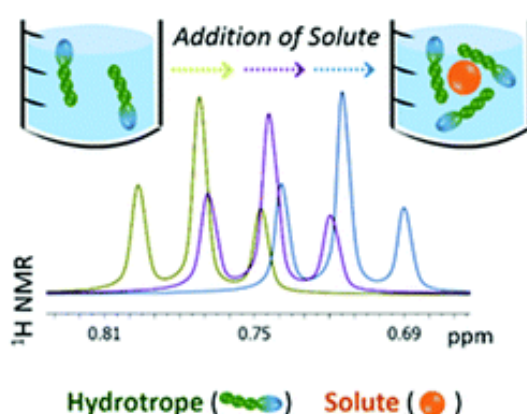
Acknowledgments

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3.2. UNVEILING THE MECHANISM OF HYDROTROPY: EVIDENCE FOR WATER-MEDIATED AGGREGATION OF HYDROTROPES AROUND THE SOLUTE

Chapter based on the published manuscript:

Dinis O. Abranches, Jordana Benfica, Bruna P. Soares, Alejandro Leal-Duaso, Tânia E. Sintra, Elísabet Pires, Simão P. Pinho, Seishi Shimizu and João A. P. Coutinho, Unveiling the mechanism of hydrotropy: evidence for water-mediated aggregation of hydrotropes around the solute *Chem. Commun.*, 2020,**56**, 7143-7146.



Synopsis: experimental evidence by ^1H -NMR to quantify the apolarity effect on hydrotropy.

3.2.1. Abstract

A recent proposal attributes the origin of hydrotropy to the water mediated aggregation of hydrotrope molecules around the solute. Experimental evidence for this phenomenon is reported for the first time in this work, using ^1H -NMR. A new computational technique to quantify apolarity is introduced and is used to show that apolarity of both solute and hydrotrope is the driving force of hydrotropy.

keywords: hydrotropy, apolarity, water mediated aggregation, chemical shift deviations, mechanism of hydrotrope.

3.2.2. Introduction

Hydrotropes, in their ability to increase the solubility of hydrophobic substances in water, can expand the applicability of the greenest and most abundant of all solvents.

Broadening the repertoire of safer solvents is in line with the principles of green chemistry (ANASTAS; EGHBALI, 2010) and is essential for a sustainable future (ANASTAS; ZIMMERMAN, 2019; CVJETKO BUBALO *et al.*, 2015; JESSOP, 2011). Besides, the addition of water suffices to force the precipitation of a solute dissolved in a hydrotropic solution, facilitating its purification (CLÁUDIO *et al.*, 2015; SOARES *et al.*, 2020).

The mechanism of hydrotropy is still not clearly understood, despite a century of research (NEUBERG, 1916) and a large debate in the literature (BAUDUIN *et al.*, 2005; BOOTH *et al.*, 2015; BOOTH; ABBOTT; SHIMIZU, 2012; BUCHECKER *et al.*, 2017; COFFMAN; KILDSIG, 1996; CUI, 2010; FRANK; FRANKS, 1968; KIM *et al.*, 2010; KUNZ; HOLMBERG; ZEMB, 2016; SANGHVI; EVANS; YALKOWSKY, 2007). Traditional speculations regarding the mechanism of hydrotropy revolved around (i) bulk-phase self-aggregation (or pre-clustering) of hydrotropes analogous to micellar solubilization (BAUDUIN *et al.*, 2005; BUCHECKER *et al.*, 2017; KIM *et al.*, 2010), (ii) “water structure” disruption by the hydrotrope that would behave like chaotropic agents weakening the hydrophobic effect (COFFMAN; KILDSIG, 1996; FRANK; FRANKS, 1968) and (iii) specific stoichiometric association between solute and hydrotrope (CUI, 2010; SANGHVI; EVANS; YALKOWSKY, 2007). However, none of these hypotheses are supported by statistical thermodynamics descriptions of hydrotropy (BOOTH *et al.*, 2015; BOOTH; ABBOTT; SHIMIZU, 2012) which suggest that hydrotrope accumulation around the solute is driven by a strong water-mediated (or hydrophobic) interaction between hydrotrope and solute. Because the apolar (or hydrophobic) moiety of a molecule interacts with water much weaker than a water-water hydrogen bond, it is driven out to associate with another hydrophobic moiety, resulting in strong agglomeration of hydrotrope around the solute.

Despite stemming from the principles of statistical thermodynamics and its consequent superiority, in terms of theoretical grounds, to the previous hypothesis, no direct experimental evidence has been to this day reported for the water-mediated accumulation mechanism proposed. In this work it will be shown that (1) the hydrophobic interaction between a hydrotrope and a solute is the driving force for the accumulation and that (2) such interaction can be quantified via a measure for apolarity derived using COSMO-RS. Both results are crucial to understand the mechanism of hydrotropy and support the hypothesis of strong water-mediated solute-hydrotrope apolar interactions.

Proton nuclear magnetic resonance (^1H -NMR) is herein employed to provide experimental evidence for hydrotrope-solute aggregation. The strategy is based on the well-known principle that the chemical shifts of the protons of a molecule dissolved in water may change due to the presence of another substance (CLÁUDIO *et al.*, 2015; STARK *et al.*, 2012). More precisely, a chemical shift that diminishes in the presence of another substance infers a higher shielding of that proton or a less probable contact between it and water. Thus, the chemical shifts of the protons associated to apolar moieties (namely methyl groups) of a hydrotrope dissolved in water can be measured in the presence and absence of a solute. If the solute induces the aggregation of hydrotropes around itself through water-mediated apolar interactions, as is predicted by statistical thermodynamics, the chemical shift of the protons associated to the apolar moieties of the hydrotrope should decrease.

Gallic acid and syringic acid were chosen as solutes and a series of monoalkylglycerol ethers, as hydrotropes. Note that the apolarity of monoalkylglycerol ethers can be made to vary smoothly through the progressive increase in the length of their alkyl chain. Likewise, gallic acid and syringic acid are structurally similar but present different hydrophobicities. Furthermore, Kunz and co-authors pointed out the necessity to study this class of hydrotropes (KUNZ; HOLMBERG; ZEMB, 2016). The solubilities of both acids in monoalkylglycerol ether aqueous solutions have been reported elsewhere (SOARES *et al.*, 2020).

3.2.3. Material and Methods

3.2.3.1. Chemicals

The substances experimentally used in this work are reported below in Table 3.3, along with their CAS number, supplier and purity, and their name abbreviation used throughout this work. The water used was doubly distilled, passed through a reverse osmosis system and later treated with Milli-Q (Millipore, model A10, Billerica, MA, USA). The glycerol ethers used were part of a batch synthesized in a previous work (SOARES *et al.*, 2020).

Table 3.3. Name, CAS Number, supplier, purity and name abbreviation of the substances used in this work.

Substance	CAS number	Supplier	Purity	Abbreviation
3-Methoxypropane-1,2-diol	623-39-2	Synthesized ^[a]	>98%	[1.0.0]
3-Ethoxypropane-1,2-diol	1874-62-0	Synthesized ^[a]	>99%	[2.0.0]
3-Propoxypropane-1,2-diol	61940-71-4	Synthesized ^[a]	>99%	[3.0.0]
3-Butoxypropane-1,2-diol	624-52-2	Synthesized ^[a]	>99%	[4.0.0]
3-Pentoxypropane-1,2-diol	22636-32-4	Synthesized ^[a]	>99%	[5.0.0]
Gallic Acid	149-91-7	Merck	>99.5%	GA
Syringic Acid	530-57-4	Acros	>98.0%	SA
Deuterium Oxide	7789-20-0	Aldrich	99.9% ^[b]	D ₂ O
3-(trimethylsilyl)propionic-2,2,3,3-d₄ acid sodium salt	24493-21-8	Eurisotop	99% ^[b]	TMSP-D ₄

[a] Compound belongs to a batch synthesized in a previous work (SOARES *et al.*, 2020). [b] atom %.

3.2.3.2. ¹H-NMR

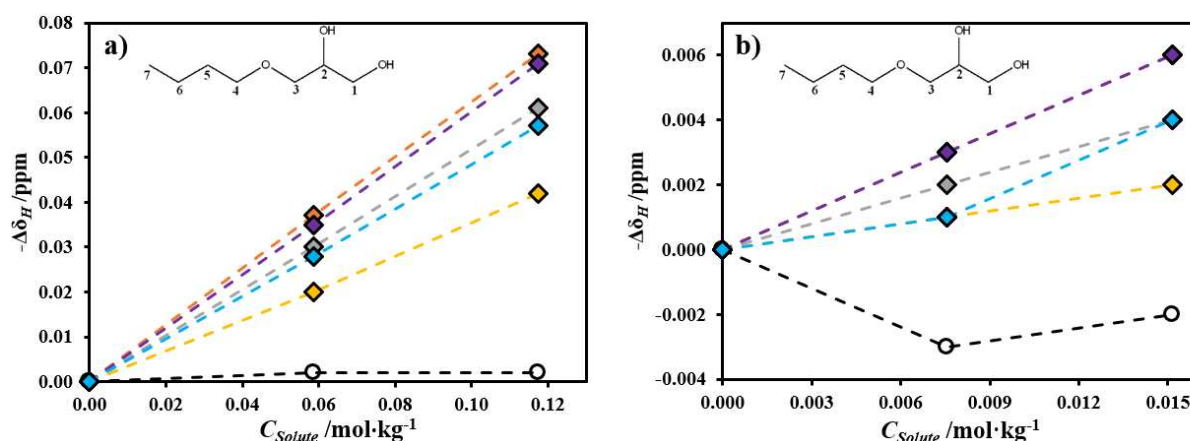
¹H-NMR spectra were acquired, at 298 K, using a Bruker Avance 300 spectrometer operating at 300.13 MHz (with chemical shifts, $\Delta\delta$, in ppm). All systems were prepared with a hydrotrope molar concentration of 0.4 M in a 1:1 ratio of water and deuterium oxide (using TMSP-D₄ as the internal reference). Five systems were prepared for each hydrotrope, varying the solute and its concentration, as described in the **Appendix B** Table S1. The aqueous solutions were prepared by adding the solute to the hydrotropic solution. All systems remained in constant agitation (1050 rpm) for 72 h using Thermomixer Eppendorf Comfort equipment, in order to ensure complete homogenization. All samples were filtered using syringe filters (0.45 μ m) to remove possible suspended solid particles. The NMR peaks considered, clearly marked in Figure S1 of **Appendix B**, were those of the protons of the alkyl side chain of the monoalkylglycerol ether along with the sole proton of the second carbon of the glycerol head. The protons of the first and third carbon of the glycerol head were not analysed due to the difficulty of distinguishing their peaks in the NMR spectra.

3.2.4. Results and discussion

The results obtained using gallic acid as a solute (Figure 3.7a as an illustrative example and Figure S2) show that the chemical shift of the protons diminishes as the concentration of solute increases. This means that the apolar moieties of the hydrotropes are statistically less prone to interact with water, providing evidence for the idea of

association between these moieties and the apolar moieties of the solute. Moreover, the decrease in chemical shift seen in Figures 3.7a and S2 is proportional to the concentration of solute. This is expected since if more solute is present in the system, more hydrotrope is needed to interact with it. Because syringic acid is much less soluble in water, its concentration was one order of magnitude lower than that of gallic acid in the NMR experiments, leading to smaller changes in the chemical shifts of the hydrotrope. In some cases, the changes produced are comparable to the experimental uncertainty of the technique (*ca.* 0.002 ppm). Nevertheless, the conclusions taken from the NMR results of gallic acid hold true for syringic acid as well, as depicted in Figures 3.7b and S3.

Figure 3.7. Change in chemical shift of the protons ($-\Delta\delta_H$) associated to water and several methyl groups of [4.0.0] (structure as inset) dissolved in water (0.4 mol/kg) as a function of a) gallic acid or b) syringic acid concentration. Legend: $-\circ-$ water; $-\diamond-$ 2nd carbon; $-\diamond-$ 4th carbon; $-\diamond-$ 5th carbon; $-\diamond-$ 6th carbon; $-\diamond-$ 7th carbon.



The NMR results question the idea that pre-clustering of the hydrotrope is fundamental in hydrotropy. Hydrotropes unquestionably do possess a degree of aggregation (clustering) with themselves. However, the hydrotrope aggregation (whether it is present depends on the system) clearly changes with the addition of the solute, providing clear evidence that the solute is not merely entering a “micelle”-like bulk-phase pre-clustering of the hydrotrope. If this were the case, there should be no change in the chemical shifts of the hydrotrope protons, since alternating from a previously hydrotrope-hydrotrope contact to a hydrotrope-solute contact would not make the hydrotrope less prone to interact with water, hence would not lead to a decrease in the chemical shifts.

Insight is given by Figures 3.7, S2 and S3 not only into its existence but also into the geometry of aggregation. In fact, the peak assigned to the protons of the second carbon of the hydrotrope always shifts less than the remaining peaks. This means that the second carbon is less prone to aggregation, which is explained by its higher degree of polarity, brought about by the presence of hydroxyl groups in its vicinity. Moreover, for all systems, the peak assigned to the protons in the last methyl group of the side alkyl chain is consistently the second less-shifting peak. That is, the second less-shifting peak for [2.0.0] is that of carbon 5, for [3.0.0] is that of carbon 6, for [4.0.0] is that of carbon 7 and for [5.0.0] is that of carbon 8 (see insets of Figure S1 for clarification). This can be interpreted in terms of interaction geometry; a parallel contact between hydrotrope and solute covers more apolar area, thus being more energetically favourable to water, than a hydrotrope tail-solute contact.

It has been argued that, for certain types of apolar yet slightly hydrophilic solutes, hydrotropy could occur due to hydrotrope-solute interactions through their polar functional groups (BUCHECKER *et al.*, 2017). This is clearly not the case for the solutes and hydrotropes studied in this work, since NMR spectroscopy revealed that the aggregation of hydrotrope around the solute happens through apolar contacts, even giving information about the geometry of these contacts.

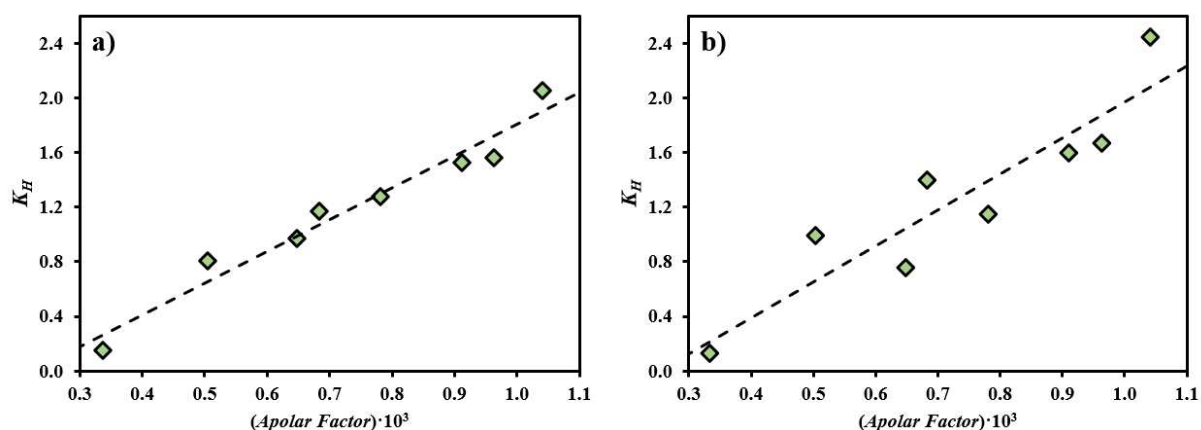
Having provided direct, experimental evidence for the hydrotrope-solute aggregation, we now address its driving force. As a quantitative measure for the apolarity of a molecule, the unnormalized σ -profile framework of COSMO-RS has been adopted (KLAMT, 2005). This is a histogram representing the amount of molecular surface with a given polarization charge-density, σ . The unnormalized σ -profile framework should prove advantageous for the study of hydrotropy since it can quantify the apolarity of both hydrotrope and solute. Moreover, the geometry and polarity of molecules optimized within the COSMO solvation model should more closely resemble that which is found in a real aqueous solution than molecules optimized in the gas phase. Details for the optimization, using TURBOMOLE V7.1, of the solute and hydrotrope molecules herein studied are given in **Appendix B**.

To study the relationship between apolarity and hydrotropy using the σ -profile framework, an apolar factor was defined:

$$\int_{-0.0082}^{0.0082} p(\sigma) \cdot (0.0082 - |\sigma|) \cdot d\sigma \quad (3.5)$$

This factor is a measure of the area under the curve of the apolar region, with the apolar/polar limit being defined as $\sigma = 0.0082 \text{ e/\AA}^2$ and $\sigma = 0.0082 \text{ e/\AA}^2$, in line with previous studies (KLAMT, 2005; PALOMAR *et al.*, 2010). The amount of apolar surface area, $p(\sigma)$, is weighted by the actual polarity of the surface, with regions near the apolar/polar limit being progressively penalized by the term $(0.0082-|\sigma|)$. The Setschenow constant (the ratio between solubility enhancement and hydrotrope concentration in the dilute region) of several hydrotropic systems previously reported in the literature was correlated against this apolar factor. Table S2 of the **Appendix B** contains the apolar factors of these hydrotropes while the correlations obtained are reported in Figure 3.8 for glycerol ether systems and Figure S4 for systems taken from the literature where the Setschenow constants were reported for different hydrotropes but the same solute. The results obtained show that, for the same family of hydrotropes, the solubility enhancement of the solute positively correlates with the apolar factor of the hydrotrope. This supports the view of water-mediated aggregation of hydrotrope around the solute and supports the idea of hydrotrope apolarity as the driving force of hydrotropy.

Figure 3.8. Setschenow constants (KUNZ; HOLMBERG; ZEMB, 2016) for a) gallic acid or b) syringic acid in glycerol ether-based hydrotropic solutions as a function of the apolar factor of the hydrotrope. The dashed line is the straight line fitted to the data using the least squares method.



Shimizu and Matubayasi (SHIMIZU; MATUBAYASI, 2016) derived a hydrotropy model based on cooperative water-mediated hydrotrope-solute aggregation using statistical thermodynamics. When regressed against experimental solubility curves, this model returns the average number of hydrotrope molecules in the vicinity of the

solute (parameter m). Since this model was previously applied to the systems herein studied (SOARES *et al.*, 2020), Figure 3.9 depicts this parameter plotted against the apolar factor of the corresponding hydrotrope. The resulting plot shows that m reaches a maximum for both gallic acid and syringic acid. Surprisingly, this maximum is located at the apolar factor of the solute. As described above, solute-hydrotrope interactions are established between their apolar moieties, resulting in strong and favourable interactions only due to the presence of water. However, there is no distinction between apolar moieties of solute and hydrotrope. Consequently, a hydrotrope that is more apolar than the solute will tend to agglomerate with itself more promptly than with the solute. Put differently, in terms of apolar contacts, it is as if there are three different forms of hydrotrope present in the system: free hydrotrope, hydrotrope associated with solute and hydrotrope associated with itself. This, again, disputes the pre-clustering hypothesis, since more self-aggregation of the hydrotrope leads to less aggregation around the solute.

Note that links between hydrophobicity of the hydrotrope and the extent of hydrotropy have been proposed before (BAUDUIN *et al.*, 2005; BUCHECKER *et al.*, 2017; KUNZ; HOLMBERG; ZEMB, 2016; SOARES *et al.*, 2020). However, this is the first time that these parameters (hydrotropy extent and apolarity of the hydrotrope) are quantified and shown to correlate remarkably well with each other. Furthermore, it is shown that the apolarity of the hydrotrope is not the only factor influencing hydrotropy. In fact, the driving force for aggregation is the balance between apolarity of both solute and hydrotrope, as demonstrated by Figure 3.9.

Figure 3.9. Dependency of the number of hydrotrope molecules in the vicinity of the solute (m) on the apolar factor of the hydrotrope, estimated using the Shimizu and Matubayasi¹⁹ model for a) gallic acid and b) syringic acid in aqueous solutions of glycerol ethers¹⁶. The black dashed line is a visual guide whilst the red dashed line represents the apolar factor of the solute.

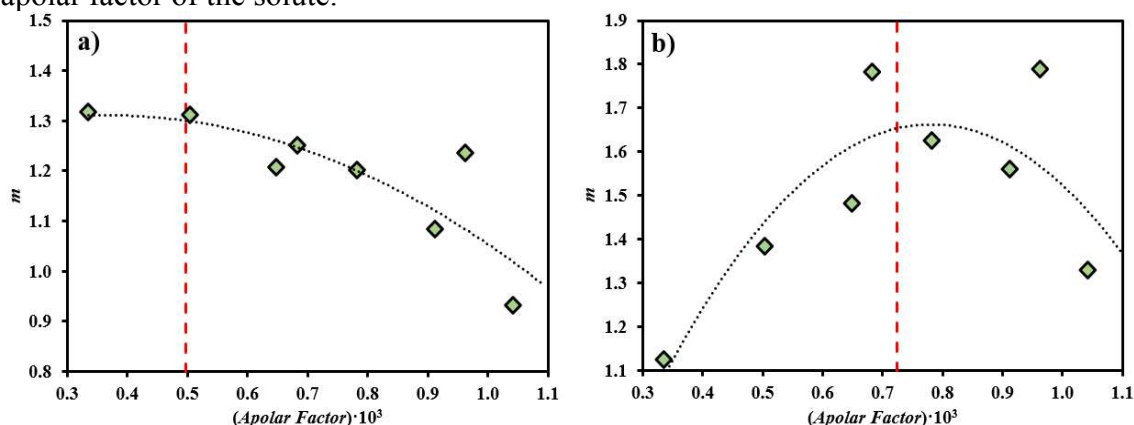


Figure 3.9 shows that hydrotropes that are more apolar than the solute will tend to aggregate less around it. This does not translate, however, into a maximum on the solubility enhancement (Figure 3.8) for the case of glycerol ethers. This is rationalized by taking into account that even though the most apolar hydrotropes may statistically possess less molecules around the solute, they are able to cover more of its apolar area due to their larger size and chain linearity, and, thus, increase its solubility.

3.2.5. Conclusions

In conclusion, experimental evidence based on ^1H -NMR chemical shifts is here reported for the first time showing that hydrotrope molecules aggregate around the solute, which supports the cooperativity theory of hydrotropy. Moreover, it was shown that apolarity of both hydrotrope and solute is the driving force of hydrotropy, with strong solute-hydrotrope interactions arising in the presence of water. These interactions are statistical and are established between apolar moieties of both solute and hydrotrope, instead of interactions between their polar functional groups. These water-mediated interactions are, however, not exclusive to solute-hydrotrope pairs and the number of hydrotropes aggregated around the solute is maximum when the apolarity of hydrotrope and solute is the same. The results reported in this work are, thus, of the utmost importance in the understanding of the water-mediated hydrotrope-solute interactions hypothesis and provide the necessary background to design new hydrotrope molecules for specific applications. A schematic illustration of the mechanism of hydrotropy, in light of these findings, is provided in Figure S5.

Acknowledgments

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desde Aragón” is acknowledged. B.P.S. acknowledges FCT for her Ph.D. grant SFRH/BD/138439/2018.

3.3. THE OILING-OUT IN HYDROTROPIC DISSOLUTION

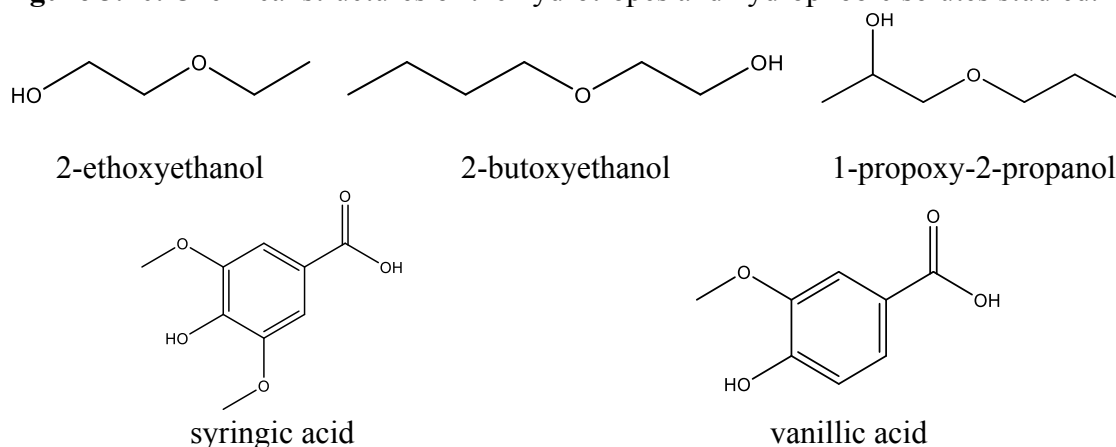
During the hydrotropic solubility studies of some compounds such as vanillic acid, a phenomenon known as oiling-out in crystallization was observed (DE ALBUQUERQUE; MAZZOTTI, 2014; DU *et al.*, 2016; SVÄRD; GRACIN; RASMUSON, 2007). This chapter presents the results obtained using glycol ethers as hydrotropes in the solubilization of vanillic and syringic acids. In addition, the oiling-out effect observed for the vanillic acid is discussed.

3.3.1. Material and Methods

3.3.1.1. Solubility Curves

Two solutes were selected, namely syringic and vanillic acids, and three glycol ethers were tested: 2-ethoxyethanol, 2-butoxyethanol, and 1-propoxy-2-propanol. Their chemical structures are depicted in Figure 3.10.

Figure 3.10. Chemical structures of the hydrotropes and hydrophobic solutes studied.



The information on the chemicals used is listed in Table 3.4. These were used without further purification. The solubility of syringic and vanillic acids in each water/hydrotrope solution was measured at 303.15 K using the analytical isothermal shake-flask methodology (HEFTER; TOMKINS, 2003), as previously reported in **Chapter 3.1**.

Table 3.4. List of substances experimentally used in this work, along with their CAS number, supplier, and mass purity.

Substance	CAS number	Source	Purity (wt%)
1-propoxy-2-propanol	1569-01-3	Sigma Aldrich	> 98.5
2-ethoxyethanol	110-80-5	Acros Organics	> 99.0
2-butoxyethanol	111-76-2	Alfa Aesar	> 99.0
syringic acid	530-57-4	Acros Organics	> 97.0
vanillic acid	121-34-6	Acros Organics	> 97.0

The solubility measurements followed the methodology described in previous chapters, and the solubility data were correlated using the Cooperative Hydrotrophy model developed by Shimizu and Matubayasi (SHIMIZU; MATUBAYASI, 2016), described in detail in **Chapter 3.1**.

3.3.1.2. COSMO-RS predictions

An SLE diagram relates the phase transitions of a mixture of two or more compounds with its composition. Assuming i) a pure solid phase and ii) neglecting the heat capacity term, which requires data that is often unavailable, and negligible when compared to other terms of the SLE equation, if the difference between T and T_m is lower than 100 K, the melting curves can be described by (GMEHLING *et al.*, 2019; PRAUSNITZ; LICHTENTHALER; AZEVEDO, 1998):

$$\ln(x_i\gamma_i) = \frac{\Delta_m h}{R} \left(\frac{1}{T_m} - \frac{1}{T} \right) \quad (3.6)$$

where γ_i is the activity coefficient of compound i , at a mole fraction x_i , in the liquid phase, T_m is its melting temperature, $\Delta_m h$ is its enthalpy of melting (at temperature T_m), R is the ideal gas constant, and T is the absolute temperature of the system. Equation 3.6 was used in this work, altogether with the melting properties ($T_m = 483.3\text{K}$ and $\Delta_m h = 34.63 \text{ kJ/mol}$) from Vilas-Boas *et al.* (VILAS-BOAS *et al.*, 2019) and COSMO-RS to predict the solid-liquid phase diagram of the vanillic acid/water system. Therefore, Equation 3.6 is solved in an iterative, self-consistent procedure by varying the temperature and using COSMO-RS to compute the activity coefficient (γ_i) of each component at a given composition, and at that iteration temperature (x_i, T). The algorithm is halted once the left- and right-hand sides of Equation 3.6 match within a given

tolerance. For comparison purposes, the calculated ideal curves taking $\gamma_i = 1$, were considered.

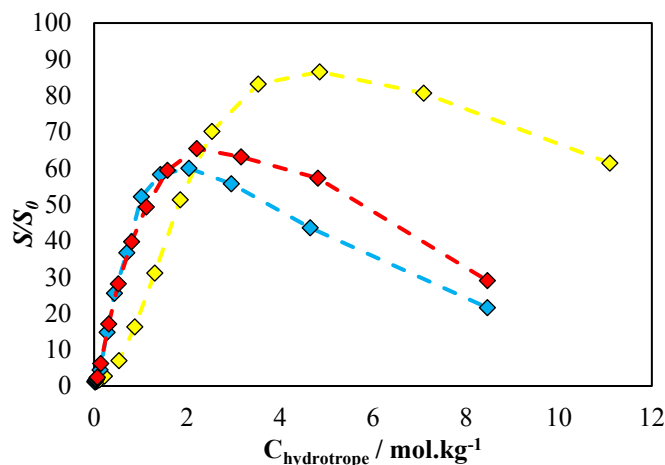
The geometries of the compounds and corresponding σ -surfaces were optimized in the software package TURBOMOLE V7.4 2019 using DFT with the BP-86 functional, the triple- ζ valence polarized basis set (TZVP), and the COSMO solvation model with infinite permittivity. Finally, the COSMOtherm software package with the BP_TZVPD_FINE_21 parametrization was used to perform all COSMO-RS-related calculations, including the aforementioned iterative algorithm.

3.3.2. Results and Discussion

3.3.2.1. Syringic acid solubilization

The results obtained for syringic acid, without surprise, given everything that has already been discussed in previous chapters, are shown in Figure 3.11 and in detail in Table S1 of **Appendix C**.

Figure 3.11. Solubility enhancement of syringic acid in aqueous solutions of glycol ethers, namely 2-ethoxyethanol $\blacklozenge$; 2-butoxyethanol $\blacklozenge$ and 1-propoxy-2-propanol $\blacklozenge$ at 303.15 K. Dashed lines are guides to the eyes.



While at low concentrations of the hydrotrope, 2-ethoxyethanol showed the lowest solubility enhancement, at high concentrations, it gives the highest S/S_0 . This same tendency was observed for the previously studied glycerol ethers, in which, although in the diluted region the hydrotropes obey a sequence of increased solubility with the chain length increase (confirmed by the Setschenow constant values), this behavior is different in the concentrated hydrotrope region, where less hydrophobic compounds have higher

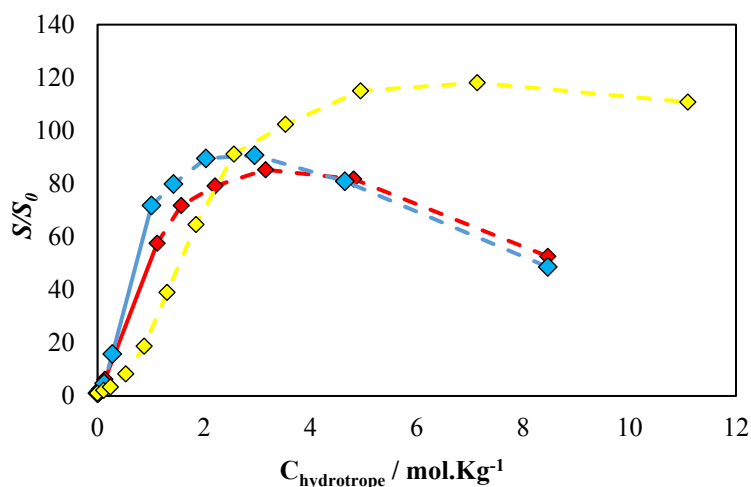
S/S_0 , as in the case of $[3.0.0] > [4.0.0] > [5.0.0]$. This apparent inconsistency was reconciled in **Chapters 3.1 and 3.2** due to the nonpolar effect of both hydrotrope and solute, in which apolarity matches cause maximum solubility enhancements.

Comparing the two hydrotropes of the same molar mass, 2-butoxyethanol and 1-propoxy-2-propanol (118.17 g/mol), despite showing similar solubility enhancements in the diluted hydrotrope region, 2-butoxyethanol performed better than 1-propoxy-2-propanol in the concentrated hydrotrope region. These differences will be discussed further in detail in **Chapter 4**, studying alkanediols as hydrotropes. It can be anticipated they are mainly related to the non-polar volume of the chain and their different levels of solute-hydrotrope aggregation and self-aggregation.

3.3.2.2. Vanillic acid solubilization and the oiling-out effect

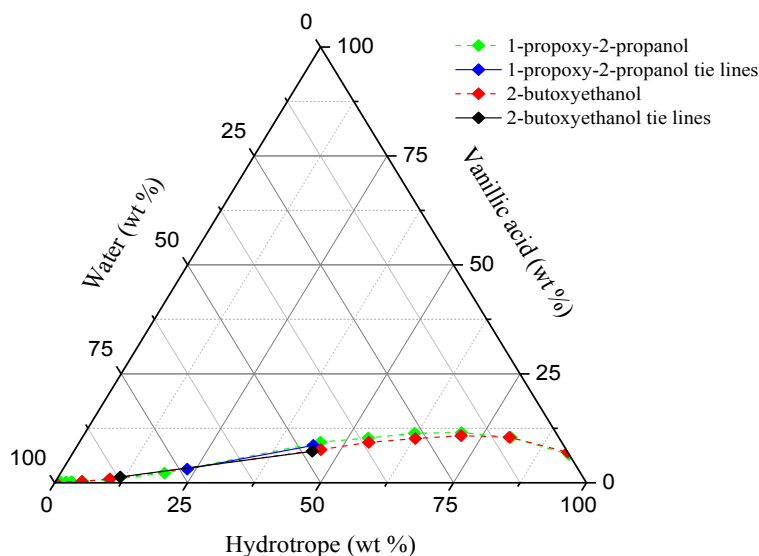
While for syringic acid, the data obtained are consistent with those reported in the previous chapters of this thesis, the studies carried out for vanillic acid were more challenging due to their unexpected behavior. In the case of vanillic acid solubilization, liquid-liquid phase separations occurred for both 2-butoxyethanol and 1-propoxy-2-propanol, at different concentrations of hydrotrope, as shown in Figure 3.12, and on the solubility data reported in detail on Tables S2-S4 of **Appendix C**. The quantification of each one of the liquid-liquid phases, determining the concentration of hydrotrope and water in both phases, was attempted. Unfortunately, the protocols developed to quantify the water content by Karl-Fisher titration, or nuclear magnetic resonance (NMR), have failed. The mass balances did not close up to a certain tolerance degree. An effort was also made trying to evaporate the water from the systems, at a controlled temperature, for a certain period, but in the end, all the solvent had evaporated, leaving back only dry vanillic acid crystals. Unfortunately, due to time constraints, it was not possible to explore more these measurements.

Figure 3.12. Solubility enhancement of vanillic acid in aqueous solutions of glycol ethers, namely 2-ethoxyethanol $\blacklozenge$; 2-butoxyethanol $\blacklozenge$ and 1-propoxy-2-propanol $\blacklozenge$ at 303.15 K. Solid lines represent the liquid-liquid phase separation region. Dashed lines are guides to the eyes.



The formation of this second phase is known in the literature as the oiling-out effect (SVÄRD; GRACIN; RASMUSON, 2007; XU *et al.*, 2021). It is usually described as the emergence of a second liquid phase during an attempted crystallization and has been reported in systems containing vanillin (DE ALBUQUERQUE; MAZZOTTI, 2014; DU *et al.*, 2016; SVÄRD; GRACIN; RASMUSON, 2007) and its derivatives, such as ethyl vanillin (ZHANG *et al.*, 2022), both in pure water and in mixed solvents. From a thermodynamic point of view, oiling-out is supposed to be caused by a superposition of liquid-liquid phase separation and solid-liquid phase equilibrium (ZHAO *et al.*, 2012). As mentioned, one of the main difficulties in the study of these systems was related to the measurements of the hydrotrope and water content in each of the phases. In such a situation, using the solubility data, the tie-lines were obtained by connecting the determined solute compositions in the ternary phase diagram, as shown in Figure 3.13.

Figure 3.13. Ternary oiling-out phase diagram with tie lines of 2-butoxyethanol and 1-propoxy-2-propanol at 303.15 K.



Comparing syringic and vanillic acid solubilization systems by glycol ethers, vanillic acid is prone to liquid-liquid separations, while for syringic acid that phenomenon does not occur. This can be due to the greater hydrophobicity of vanillic acid. Interestingly, vanillic acid shows an opposite behavior to that observed for syringic acid with the two glycol ethers. Although at high dilution both compounds present similar solubility enhancement (see Figure 3.12), starting from the liquid-liquid phase separation region, 1-propoxy-2-propanol performs better than 2-butoxyethanol, until higher hydrotrope concentrations, when similar enhancements are obtained again. This behavior is directly related to the higher solubility capacity of 2-butoxyethanol to dissolve vanillic acid, making it more prone to liquid-liquid phase separation (in the water-domain region) than 1-propoxy-2-propanol, confirmed by little higher m values obtained for 2-butoxyethanol (see Table S5 in **Appendix C**).

The liquid-liquid phase separation region for 2-butoxyethanol is larger, 20-40% wt. *versus* 30-40% wt. for 1-propoxy-2-propanol. The solid-liquid-liquid phase separation boundary increases in size when the strength of the solute-hydrotrope interaction increases. In the case of 2-ethoxyethanol, this strength of interaction was not enough to induce the liquid-liquid phase separation, also achieving the highest S/S_0 for vanillic acid and the higher m values ($m = 2.0$), affirming the superior level of aggregation between

hydrotrope-solute molecules for that compound, compared to the other glycol ethers, that induced the liquid-liquid phase separation, with m values around 1.6.

In the vanillic acid-hydrotrope-water system, when the hydrotrope concentration is low, the solution is dominated by water with little vanillic acid dissolved. Then, when the hydrotrope proportion is below about 20% and 30% in solvent regarding the proportion of 2-butoxyethanol and 1-propoxy-2-propanol, respectively, there is no liquid-liquid phase separation. Increasing the hydrotrope concentration more vanillic acid is dissolved in the solution until the liquid-liquid phase separation occurs. When the two liquid phases are formed, increasing the global hydrotrope concentration provokes the decrease of the water-rich phase, and the hydrotrope-rich phase increases, until the solution is dominated by the hydrotrope, leading to a homogenous solution, when the liquid-liquid phase separation disappears. The evolution along the hydrotrope concentration change is depicted in Figure 3.14.

Figure 3.14. The liquid-liquid phase separation layers for different mass percentages of 2-butoxyethanol: A) 10 wt. % (homogeneous system), B) 20 wt. % (water-rich layer), C) 30 wt.% (water-rich layer and hydrotrope rich-layer), D) 40 wt.% (hydrotrope-rich layer), E) 50 wt.% (homogeneous system).

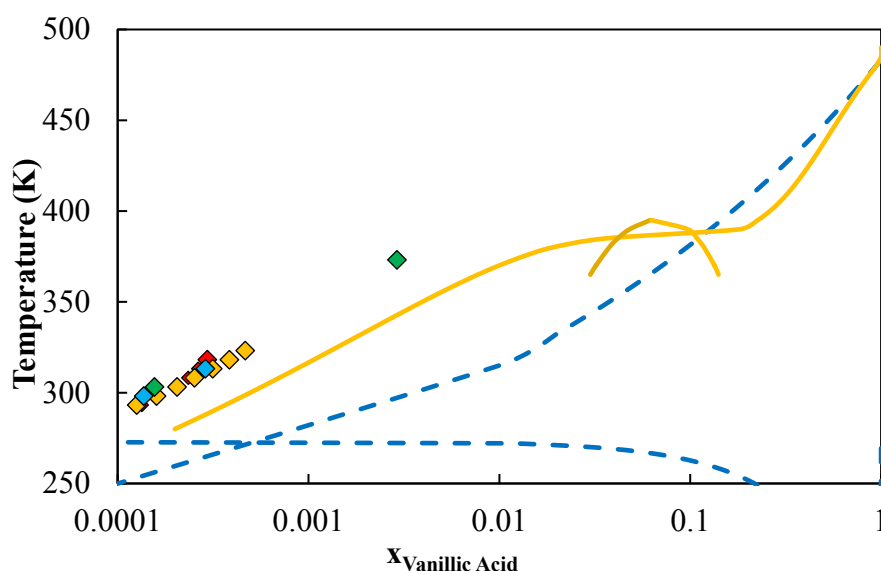


3.3.2.3. Vanillic acid/water equilibrium

It is significant to mention that the liquid-liquid separation in binary solute-water systems has been previously reported for vanillin (at 324.15 K) (SVÄRD; GRACIN; RASMUSON, 2007) and even for different parabens, namely, butyl paraben (at 322.55 K), propyl paraben (at 348.75 K), and ethyl paraben (at 357.65 K) (YANG; RASMUSON, 2014). In these cases, the temperatures for liquid-liquid phase separation occurring in

water solely are correlated to the melting points of the correspondent solute and the process of oiling-out in water can be considered as melting liquid solutes (vanillin/parabens) is “supercooled” below the melting point, and then mixed with pure water (YANG; RASMUSON, 2014). For vanillin, ethyl vanillin, and the parabens, their pure melting point ranges from 343 to 388 K however, for vanillic acid, the melting point is much higher, between 480 and 485 K (VILAS-BOAS *et al.*, 2019). Lower melting enthalpies would compensate for the high melting point to form the liquid-liquid separation, but this is also not the case for vanillic acid since the melting enthalpy is in the range of 25-35 kJ·mol⁻¹ (VILAS-BOAS *et al.*, 2019), of the same order of magnitude as those found for parabens, vanillin, and ethyl vanillin (around 23-26 kJ·mol⁻¹) (YANG; RASMUSON, 2014; ZHANG *et al.*, 2022). Interestingly, COSMO-RS predictions of the phase diagram of vanillic acid/water show the formation of a liquid-liquid equilibrium, as can be seen in Figure 3.15.

Figure 3.15. Solid-liquid phase diagram of vanillic acid/water. Lines represent the COSMO-RS predictions (—) and ideal melting curves of the system (---). Diamonds represent the literature data: (NOUBIGH *et al.*, 2008) ◆; (YALKOWSKY; HE; JAIN, 2016) ◆; (ZHANG *et al.*, 2016) ◆ and (VILAS-BOAS *et al.*, 2019) ◆.



From Figure 3.15, it is possible to see two different regions: i) the first dominated by water, with positive deviations from the ideality (concerning γ_i values), which means weak interactions between water and vanillic acid, favoring the oiling-out effect, and ii) the second region dominated by the presence of vanillic acid, with negative deviations

from the ideality, with stronger interactions between water and vanillic acid than between the vanillic acid molecules, and the absence of a second phase. This diagram shows, once again, that the appearance of liquid-liquid separation in water/solute systems is primarily caused by the poor interaction of water with the solute in the water-dominated region, due to the high hydrophobicity of the solute.

It is relevant to highlight that despite the solid-liquid diagram from COSMO-RS provides a good approximation of literature data, the liquid-liquid equilibrium for vanillic acid has not yet been observed experimentally. In this work, the solubility was measured at 303.15 K, but an extensive study concerning a wide temperature range would be necessary to define at what point the oiling-out would be observed and the impact of the temperature in the liquid-liquid phase separation that occurs for hydrotrope/water mixtures. The oiling-out predicted by COSMO-RS could be present at temperatures below the SLE, and therefore, it may not be thermodynamically stable. Despite this, the COSMO-RS predictions suggest that the immiscibility of vanillic acid in water is the main factor behind the oiling-out observed in the ternary systems. For hydrotrope/water mixtures, however, the mechanism of oiling-out is more evident since hydrotropes increase the solubility of vanillic acid, facilitating the appearance of the liquid-liquid phase separation.

Lastly, as noticed by the discussion above, the oiling out effect may be a disadvantage for the solubilization of hydrophobic solutes like vanillin derivatives as well as for solute crystallization. Thus, the study of how the solutes are partitioned in the two liquid phases is crucial to design and control crystallization and purification process.

3.3.3. Conclusions and future perspectives

This short work shows that liquid–liquid phase separation, known as oiling-out, might happen under some conditions in hydrotrope/water systems for vanillic acid. To understand the mechanism of the liquid–liquid split phase diagram was extrapolated from the solubility data of vanillic acid in 2-butoxyethanol and 1-propoxy-2-propanol/ water mixtures. It was concluded that the oiling-out effect occurred only at specific hydrotrope concentrations resulting from the interplay between the hydrotrope-solute interaction and solute hydrophobicity. At the temperature studied syringic acid did not show an oiling-out effect due to its lower hydrophobicity, which is not enough to induce liquid-liquid separation. The oiling-out for vanillic acid is somewhat different from that for other

solutes like vanillin derivatives and parabens with lower melting points, but COSMO-RS suggests that despite its high melting point, the oiling out is probably driven by the poor mutual solubility of water and vanillic acid.

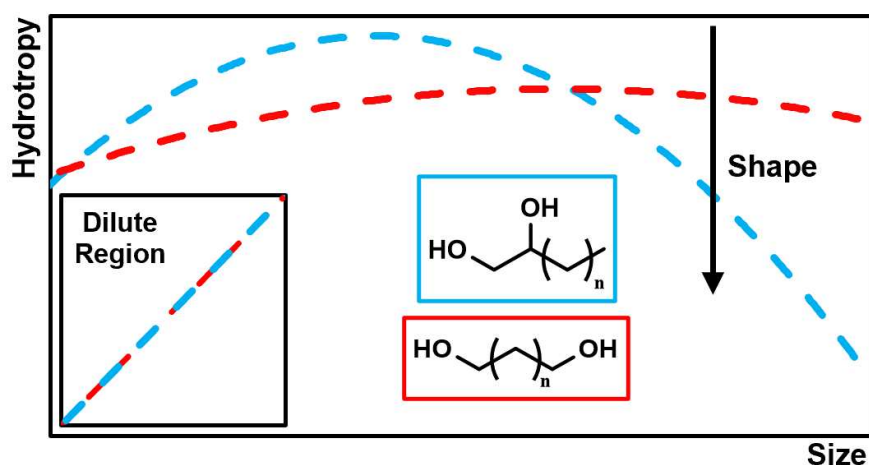
To better understand the oiling-out phenomena affecting hydrotropes, studies concerning the temperature effect are essential since miscibility is a function of temperature. Phase diagrams must be built more accurately using specific experimental methodologies for determining liquid-liquid equilibrium, and their respective tie lines, in the ternary diagrams. In the end the liquid-liquid phase separation of vanillic acid, and its crystallization, can be circumvented by liquid-liquid and solid-liquid equilibrium studies.

4. CHAPTER IV - ALKANEDIOLS AS HYDROTROPES

4.1. THE IMPACT OF SIZE AND SHAPE IN THE PERFORMANCE OF HYDROTROPES: A CASE-STUDY OF ALKANEDIOLS

Chapter based on the published manuscript:

Dinis O. Abranches[‡], Bruna P. Soares[‡], Tânia E. Sintra, Ana M. Ferreira, Seishi Shimizu, Simão P. Pinho and João A. P. Coutinho, The impact of size and shape in the performance of hydrotropes: a case-study of alkanediols, *Phys. Chem. Chem. Phys.* v. 24, p. 7624-7634, 2022.



Synopsis: studying the impact of the size and shape of hydrotropes in their ability to enhance the aqueous solubility of hydrophobic compounds.

4.1.1. Abstract

Inspired by the recently proposed cooperative mechanism of hydrotropy, where water molecules mediate the aggregation of hydrotrope around the solute, this work studies the impact of apolar volume and polar group position on the performance of hydrotropes. To do so, the ability of two different families of alkanediols (1,2-alkanediols and 1,n-alkanediols) to increase the aqueous solubility of syringic acid is initially investigated. Interestingly, it is observed that in the dilute region (low hydrotrope concentration), the relative position of the hydroxyl groups of the alkanediols does not impact their performance. Instead, their ability to increase the solubility of syringic acid correlates remarkably well with the size of their alkyl chains. However, this is not the case for larger hydrotrope concentrations, where 1,2-alkanediols are found to perform, in general, better than 1,n-alkanediols. These seemingly contradictory findings are

reconciled using theoretical and experimental techniques, namely the cooperative model of hydrotrophy and chemical environment probes (Kamlet-Taft and pyrene polarity scales). It is found that the number of hydrotropes aggregated around a solute molecule does not increase linearly with the apolar volume of the former, reaching a maximum instead. This maximum is discussed in terms of competing solute-hydrotrope and hydrotrope-hydrotrope interactions. The results suggest that hydrotrope self-aggregation is more prevalent in 1,n-alkanediols, which negatively impacts their performance as hydrotropes. The results reported in this work support the cooperative model of hydrotrophy and, from an application perspective, show that hydrotropes should be designed taking into consideration not only their apolar volume but also their ability to stabilize their self-aggregation in water, which negatively impacts their performance as solubility enhancers.

keywords: alkanediols, hydrotrophy, apolar volume, self-aggregation, mechanism of action of hydrotropes.

4.1.2. Introduction

Following the principles of Green Chemistry (ANASTAS; EGHBALI, 2010), water is undoubtedly the most sustainable solvent: it is readily available, non-flammable, non-toxic, cheap, and environmentally benign. Unfortunately, many compounds that possess relevant properties and bioactivities display poor aqueous solubility. Rather than using different solvents to process these compounds, hydrotropes can be used to overcome this limitation. Hydrotropes are amphiphilic organic substances that significantly increase the aqueous solubility of hydrophobic compounds (KUNZ; HOLMBERG; ZEMB, 2016; PAUL; CHATTARAJ; PAUL, 2021; SHIMIZU, 2020). Since the concept was first proposed (NEUBERG, 1916), a variety of substances have been studied as hydrotropes, namely organic sulfonate salts (HODGDON; KALER, 2007), glycerol ethers and esters (LEBEUF *et al.*, 2016; MOITY *et al.*, 2013; SOARES *et al.*, 2020), ionic liquids (ABRANCHES *et al.*, 2021; CLÁUDIO *et al.*, 2015; SINTRA *et al.*, 2021), and even simple organic molecules such as ethanol and urea (COFFMAN; KILDSIG, 1996).

In contrast with surfactants, hydrotropes do not form well-structured aggregates such as micelles, and their mechanism of action is still not fully understood. After much

speculation in the literature, the statistical thermodynamics-based cooperative model of hydrotropy (SHIMIZU; MATUBAYASI, 2016) was recently proposed and experimentally confirmed by proton nuclear magnetic resonance (^1H -NMR) (ABRANCHES *et al.*, 2020a). Within the framework of this model, a hydrotrope increases the solubility of a hydrophobic substance by aggregating around it in solution through its apolar moieties (hydrophobic effect). This aggregation is water-mediated, in the sense that aggregation between the apolar moieties of both hydrotrope and solute maximizes hydrogen bond-type interactions between water molecules or between water and hydrotrope. As such, the hydrotropic effect can be heightened by carefully selecting hydrotropes with large apolar volumes that still retain aqueous solubility.

Given the dependence of hydrotropy on the apolar volumes of solute and hydrotrope, families of compounds that possess easily tuned apolarity are a powerful tool to understand the overall mechanism of this phenomenon and rationally design better green hydrotropes for specific applications. For instance, while investigating the ability of glycerol and glycerol-derivatives to enhance the aqueous solubility of syringic acid, increasing the size of the alkyl side chain of monoglycerol ethers provided key insight into the mechanism of hydrotropy (ABRANCHES *et al.*, 2020a; SOARES *et al.*, 2020), revealing that the magnitude of the solubility enhancement of a given solute correlates well with the apolarity of the hydrotrope and reaches a maximum when the apolar surface area of the hydrotrope and solute match. However, whether this is a general trend for all hydrotrope-solute pairs or a phenomenon specific to glycerol ethers is still unclear.

Alkanediols are a family of compounds that possess similar structures and amphiphilicity to monoglycerol ethers yet are structurally simpler and offer an additional array of tuning opportunities, namely the size of their alkyl chain and the position of their hydroxyl groups. As such and considering the recent success of glycerol ethers highlighted in the previous paragraph, alkanediols offer the most valuable opportunity to systematically study the combined effects of hydrophobic and hydrophilic interactions in hydrotropy. Furthermore, alkanediols present a certain degree of sustainability, with many being FDA-approved and widely used in the cosmetic, food, and pharmaceutical industries (BEHR *et al.*, 2008). They have also been used to prepare sustainable deep eutectic solvents that extract and recover bioactive compounds from natural sources with high overall efficiency (OZTURK; GONZALEZ-MIQUEL, 2019).

To assess the impact of the alkyl chain length (size) and relative position of polar groups (shape) of hydrotropes on the solubility enhancement of hydrophobic solutes, the ability of two sets of alkanediols (1,2-alkanediols and 1,n-alkanediols with alkyl chain sizes ranging from one to seven carbons) to increase the aqueous solubility of syringic acid is investigated in this work. Alkanediols were chosen as model hydrotropes due to their tuning potential and sustainable character described above. Syringic acid was selected as the model solute due to its hydrophobicity, therapeutic activity, and presence in bio-relevant natural sources where sustainable extraction processes are desirable (SRINIVASULU *et al.*, 2018). In fact, syringic acid is an antioxidant and an anti-inflammatory, while also functioning as a therapeutic agent for a wide range of diseases, such as cancer and diabetes. It can be found in several plants, ranging from grapes and olives to natural products such as honey, and functions as a surrogate of lignin (of which it is a monomer) in solubility studies. The solubility results obtained are modeled and interpreted in light of the Setschenow correlation and cooperative model of hydrotropy. The Kamlet-Taft (KAMLET; ABBOUD; TAFT, 1977) and *Py* (DONG; WINNIK, 1982) polarity scales are measured and used to rationalize the results obtained. To better understand the impact of the hydrophobicity of the solute in its interactions with the hydrotrope, two additional solutes with large apolar volumes are also studied (Reichardt's dye and pyrene).

4.1.3. Experimental Section

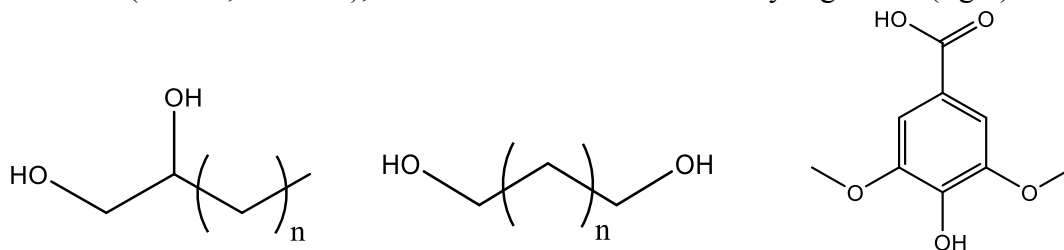
4.1.3.1. Chemicals

The chemical substances used in this work are listed in Table 4.1, along with their CAS number, source, and mass purity. The generic chemical structures of 1,2-alkanediols and 1,n-alkanediols, and the chemical structure of syringic acid are depicted in Figure 4.1 (see Figure S1 in **Appendix D** for a full list of chemical structures). The water used in all solubility essays was double distilled, passed across a reverse osmosis system, and treated with a Milli-Q plus 185 water purification apparatus.

Table 4.1. List of substances experimentally used in this work, along with their CAS number, supplier, and mass purity.

Substance	CAS	Source	Purity (wt %)
1,2-Ethanediol	107-21-1	Fisher	>99.0
1,2-Propanediol	57-55-6	Sigma-Aldrich	99.5
1,3-Propanediol	504-63-2	Sigma-Aldrich	>98.0
1,2-Butanediol	584-03-2	Sigma-Aldrich	>98.0
1,4-Butanediol	110-63-4	Alfa Aesar	>99.0
1,2-Pentanediol	5343-92-0	TCI	97.0
1,5-Pentanediol	111-29-5	Alfa Aesar	97.0
1,2-Hexanediol	6920-22-5	Alfa Aesar	97.0
1,6-Hexanediol	629-11-8	Acros Organics	97.0
1,7-Heptanediol	629-30-1	TCI	>98.0
Syringic acid	530-57-4	Acros Organics	>98.0
Reichardt's dye	10081-39-7	Sigma-Aldrich	90.0
Pyrene	129-00-0	Sigma-Aldrich	>99.0
N,N-diethyl-4-nitroaniline	2216-15-1	Fluorochem	>99.0

Figure 4.1. Generic chemical structures of 1,2-alkanediols (left, $n=0$ to 3) and 1, n -alkanediols (middle, $n=0$ to 5), and the chemical structure of syringic acid (right).



4.1.3.2. Solubility Curves

The solubility of syringic acid in each water/hydrotrope solution was measured using the analytical isothermal shake-flask methodology.^(HEFTER; TOMKINS, 2003) Syringic acid was added in slight excess to each aqueous solution of hydrotrope or pure hydrotrope. The samples were equilibrated in an air oven at (303.2 ± 0.5) K under constant stirring (1050 rpm) for 72 h, using an Eppendorf Thermomixer Comfort equipment. After 72 h, all samples were centrifuged at (303.2 ± 0.5) K and 4500 rpm for 20 minutes in order to separate the excess undissolved solute from the liquid phase. Then, liquid-phase samples were carefully collected, filtered using syringe filters, and diluted in ultra-pure water. The amount of syringic acid was quantified by UV-spectroscopy

using a SHIMADZU UV-1700 Pharma-Spec spectrometer at 267 nm. The solubility of syringic acid in pure water was found to be (1.462 ± 0.004) g/L, in line with previous results reported in the literature (ABRANCHES *et al.*, 2020c, 2021).

The procedure described above was also used to measure the solubility of Reichardt's dye and pyrene in aqueous solutions of 1,2-pentanediol or 1,5-pentanediol. Due to the high hydrophobicity of the solutes, the collected and filtered samples were diluted in water/ethanol 50% (v/v) and then quantified by UV-spectroscopy at 304 nm and 335 nm, respectively. The solubility of Reichardt's dye in pure water was found to be (30.8 ± 0.5) mg/L, while the solubility of pyrene in pure water (0.17 mg/L) was taken from the literature (MAY *et al.*, 1983).

4.1.3.3. Cooperative Model of Hydrotropy

The solubility data for syringic acid obtained in this work was fitted using the cooperative model of hydrotropy, a statistical thermodynamics-based model developed by Shimizu and Matubayasi.(SHIMIZU; MATUBAYASI, 2016) This model is based on solute-hydrotrope aggregation in solution and was developed for sigmoidal-shaped solubility curves. It can be summarized as:

$$\ln \left[\frac{1 - \frac{S}{S_0}}{\frac{S}{S_0} - \left(\frac{S}{S_0}\right)_{max}} \right] = m \ln(x_H) + b \quad (4.1)$$

where S and S_0 are the molar solubilities (mol/L) of the solute in the aqueous hydrotrope solution, or pure water, respectively, $(S/S_0)_{max}$ is the maximum attainable relative solubility caused by a given hydrotrope, x_H is the mole fraction of the hydrotrope in the ternary water/hydrotrope/solute solution, and m and b are fitted parameters of the model.

Equation 4.1 is often fitted against solubility data by leaving $(S/S_0)_{max}$ as an adjustable parameter as well (ABRANCHES *et al.*, 2020c). This is done because most experimental solubility curves are not perfectly sigmoidal, with some presenting an absolute maximum, casting doubt on the ability to determine a value for $(S/S_0)_{max}$ correctly. To avoid this problem and given that some of the solubility curves obtained in this work are not sigmoidal, this procedure is adopted here.

4.1.3.4. Solvent Polarity Scales

The Kamlet-Taft solvatochromic parameter π^* (solvent dipolarity/polarizability) (KAMLET; ABOUD; TAFT, 1977) and the *Py* scale parameter I_1/I_3 (ratio between the first and third bands of the fluorescence spectrum of pyrene) (DONG; WINNIK, 1982) were experimentally measured in this work for aqueous solutions of 1,2-pentanediol or 1,5-pentanediol.

The solvatochromic parameter π^* was measured using the dye N,N-diethyl-4-nitroaniline. After vigorous stirring for the complete dissolution of the dye, samples were scanned using a UV-Vis spectrophotometer (BioTeck Synergy HT microplate reader). The longest wavelength absorption band was used to determine the parameter using the following equation:

$$\pi^* = \frac{\nu_{alkanediol} - \nu_{ciclohexane}}{\nu_{DMSO} - \nu_{ciclohexane}} \quad (4.2)$$

where ν is the experimental wave number and the subscripts *alkanediol*, *ciclohexane*, and *DMSO* correspond to the solvent used.

To measure the polarity index (I_1/I_3), fluorescence measurements using pyrene were carried out, as previously described in the literature (DONG; WINNIK, 1982; KUSUMOTO *et al.*, 1997; VASILESCU *et al.*, 1997; ZANA; ELJEBARI, 1993). The fluorescence emission spectra were obtained with aerated solutions at very low pyrene concentration ($3 \cdot 10^{-7}$ mol/L), which ensured the absence of pyrene excimers. Adequate volumes of pyrene in ethanol solutions were transferred to empty Eppendorf vessels and the solvent was carefully evaporated. The aqueous solutions of alkanediols (1,2 and 1,5-pentanediol) were then added. In order to ensure equilibrium, the spectra were measured after 48 h of constant stirring (2000 rpm) using an Eppendorf Thermomixer Comfort equipment. The pyrene spectra were quantified using a JASCO FP-8300 spectrofluorometer at an excitation wavelength of 318 nm. The spectra were used to determine the ratio of the intensities of the first and third vibronic peaks of monomeric pyrene (polarity index, I_1/I_3).

4.1.4. Results and Discussion

4.1.4.1. Solubility Curves of Syringic Acid

The solubility curves of syringic acid in aqueous solutions of 1,2-alkanediols and 1,n-alkanediols, measured in this work, are depicted in Figure 4.2. Detailed experimental results are reported in **Appendix D** (Tables S1 and S2). The solubility of syringic acid was measured in the entire experimentally available concentration range of the hydrotrope, from pure water to pure hydrotrope or its aqueous solubility limit.

Figure 4.2. Solubility enhancement of syringic acid in aqueous solutions of 1,2-ethanediol $\blacklozenge$ or $\blacktriangle$, 1,2-propanediol $\blacklozenge$, 1,2-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, 1,2-hexanediol $\blacklozenge$, 1,3-propanediol $\blacktriangle$, 1,4-butanediol $\blacktriangle$, 1,5-pentanediol $\blacktriangle$, 1,6-hexanediol $\blacktriangle$, and 1,7-heptanediol $\blacktriangle$ as a function of hydrotrope concentration, at 303.2 K. S/S_0 is the relative solubility of syringic acid and C_H is the concentration of hydrotrope per kg of solution (solute-free basis). Dashed lines are visual guides.

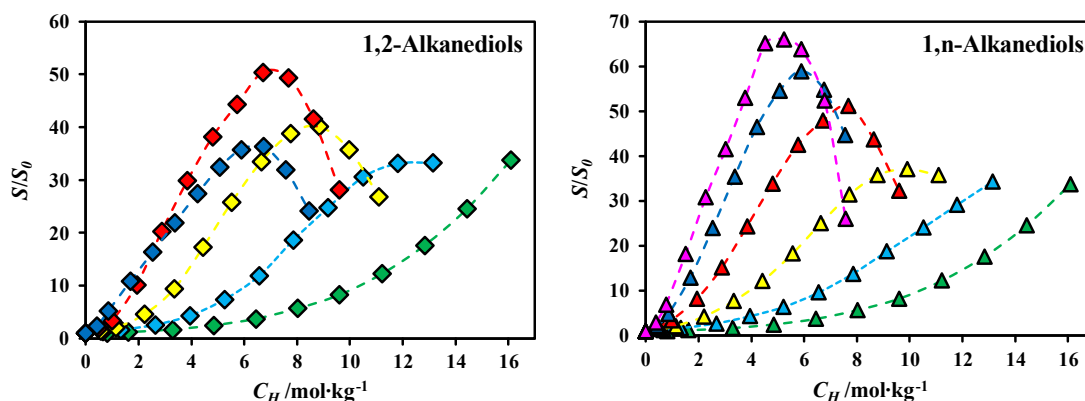


Figure 4.2 reveals that the aqueous solubility of syringic acid can be increased up to 60-fold using alkanediols as hydrotropes. This value is similar to that obtained with other nonionic hydrotropes previously studied, such as glycerol ethers (up to 77-fold for 1,3-dimethoxy-2-propanol) (SOARES *et al.*, 2020) and Cyrene (up to 45-fold) (ABRANCHES *et al.*, 2020c). Furthermore, the performance of alkanediols is superior to ionic hydrotropes with densely charged counterions, such as the prototypical 1-butyl-3-methylimidazolium chloride (which enhances the aqueous solubility of syringic acid up to 40-fold), but inferior to ionic hydrotropes containing amphiphilic counter ions, such as 1-butyl-3-methylimidazolium p-toluenesulfonate that reached a 170-fold solubility improvement (ABRANCHES *et al.*, 2021).

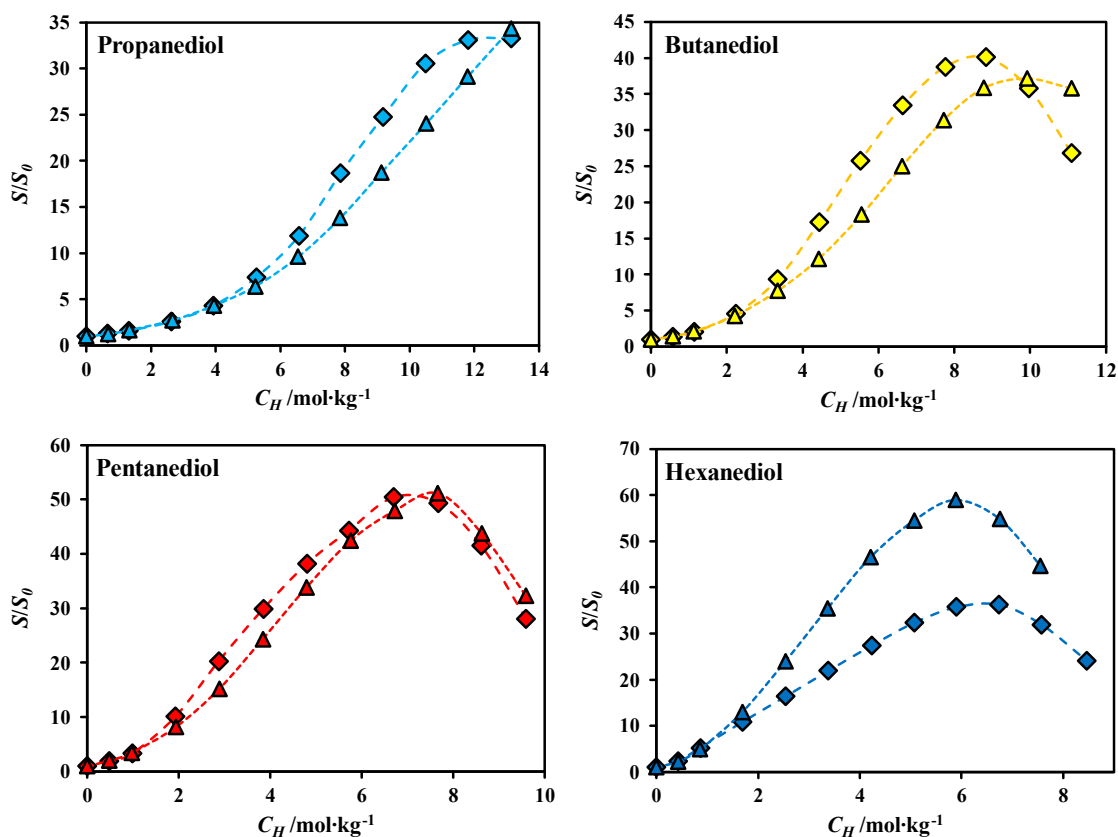
Except for 1,2-hexanediol, the results depicted in Figure 4.2 reveal that the solubility enhancement of syringic acid generically increases with the size of the alkyl

chain of the hydrotrope, up to a hydrotrope concentration of 6 mol/kg. This trend is in line with the recently proposed molecular mechanism of hydrotropy, where the apolar volume of the hydrotrope is one of the main driving forces of hydrotropy (ABRANCHES *et al.*, 2020a; SHIMIZU; MATUBAYASI, 2016). However, between hydrotrope concentrations of 6 and 9 mol/kg, the solubility of syringic acid reaches a maximum for hydrotropes with larger alkyl chains, decreasing afterward. This is most likely due to a change in the solvation regime, as will be discussed in the following sections.

Although both families of hydrotropes display the same apolar volume trend, as discussed in the previous paragraph, the position of the hydroxyl group appears to impact their performance. To better showcase this, the data reported in Figure 4.2 are replotted in Figure 4.3 to highlight the differences between the performances of 1,2-alkanediols and 1,n-alkanediols.

The results depicted in Figure 4.3 reveal a curious trend. For shorter alkyl chains, 1,2-alkanediols outperform 1,n-alkanediols, with both 1,2-propanediol and 1,2-butanediol displaying higher solubility enhancements than 1,3-propanediol and 1,4-butanediol, respectively. However, the trend is inverted for larger alkyl chains. While there is no noticeable difference between the performances of 1,2-pentanediol and 1,5-pentanediol, 1,6-hexanediol can achieve a greater syringic acid solubility enhancement (up to 60-fold) than 1,2-hexanediol (up to 40-fold). This clearly shows that the relative position of polar groups in hydrotropes impacts their ability to enhance the aqueous solubility of hydrophobic solutes, although the reason for this phenomenon is not yet clear.

Figure 4.3. Solubility enhancement of syringic acid in aqueous solutions of propanediol (1,2-propanediol $\blacklozenge$ and 1,3-propanediol $\blacktriangle$), butanediol (1,2-butanediol $\blacklozenge$ and 1,4-butanediol $\blacktriangle$), pentanediol (1,2-pentanediol $\blacklozenge$ and 1,5-pentanediol $\blacktriangle$), and hexanediol (1,2-hexanediol $\blacklozenge$ and 1,6-hexanediol $\blacktriangle$), as a function of hydrotrope concentration, at 303.2 K. S/S_0 is the relative solubility of the solute and C_H is the concentration of hydrotrope per kg of solution (solute-free basis). Dashed lines are visual guides.



4.1.4.2. The Dilute Region

The interpretation of the effect that the relative position of hydroxyl groups in alkanediols has on hydrotropy is made difficult by the diversity of possible interactions that can be established between water, hydrotrope, and solute, each playing favorable or unfavorable roles towards the solubility enhancement of the solute. While this is certainly true for larger concentrations of hydrotrope, in the dilute region (i.e., low hydrotrope concentration) the contribution to solubilization from hydrotrope-hydrotrope and solute-solute interactions has been shown to be greatly decreased (SHIMIZU, 2020). Thus, studying the dilute region should provide important clues to understand solute-hydrotrope interactions.

The Setschenow constant (K_S) quantifies the solubility enhancement of a substance due to the addition of a third component (e.g. hydrotrope, cosolvent, salt, etc.)

in the dilute region (NI; YALKOWSKY, 2003; SETSCHENOW, 1989). In this work, it is defined as:

$$\ln(S/S_0) = K_S \cdot C_H \quad (4.3)$$

where S and S_0 are the molar solubilities (mol/L) of the solute in the aqueous hydrotrope solution and pure water, respectively, and C_H is the concentration (mol/kg_{solution}) of the hydrotrope in a solute-free basis.

Equation 4.3 was initially proposed by Setschenow (SETSCHENOW, 1989) to quantify salting-out and salting-in effects but has since proved useful in the field of hydrotropy (ABRANCHES *et al.*, 2020c; SHIMIZU, 2020; SOARES *et al.*, 2020). Note that this equation is, in essence, a linear correlation between $\ln(S/S_0)$ and C_H , and is only valid when these variables possess a linear relationship (i.e., the dilute region). In this work, this was found to be true up to about 20 wt%, depending on the hydrotrope. As such, each solubility curve reported in the previous section was fitted using Equation 4.3 in the dilute region, and the results obtained are depicted in Figure 4.4.

Figure 4.4. Natural logarithm of the relative solubility enhancement of syringic acid, at 303.2 K, as a function of the concentration of hydrotrope for 1,2-alkanediols (left) and 1,n-alkanediols (right), in particular 1,2-ethanediol $\blacklozenge$, 1,2-propanediol $\blacklozenge$, 1,2-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, 1,2-hexanediol $\blacklozenge$, 1,3-propanediol $\blacktriangle$, 1,4-butanediol $\blacktriangle$, 1,5-pentanediol $\blacktriangle$, 1,6-hexanediol $\blacktriangle$ and 1,7-heptanediol $\blacktriangle$. Dashed lines represent the best linear fitting for each data set obtained using the least square method and setting the intercept to zero.

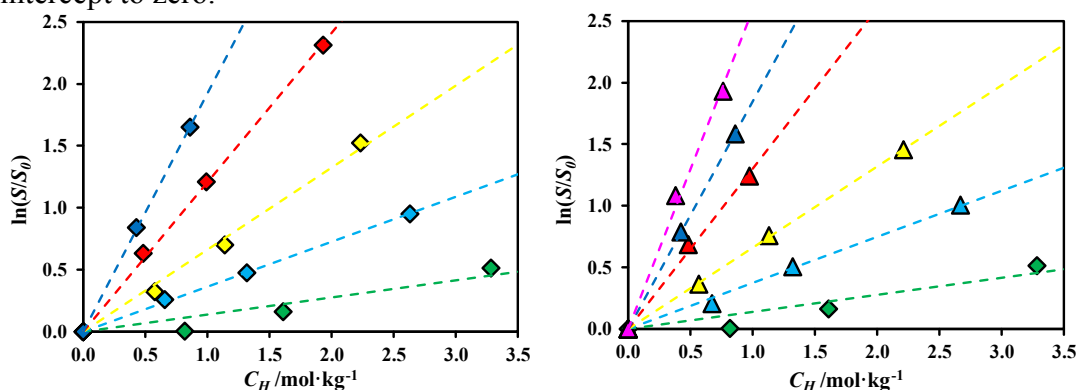
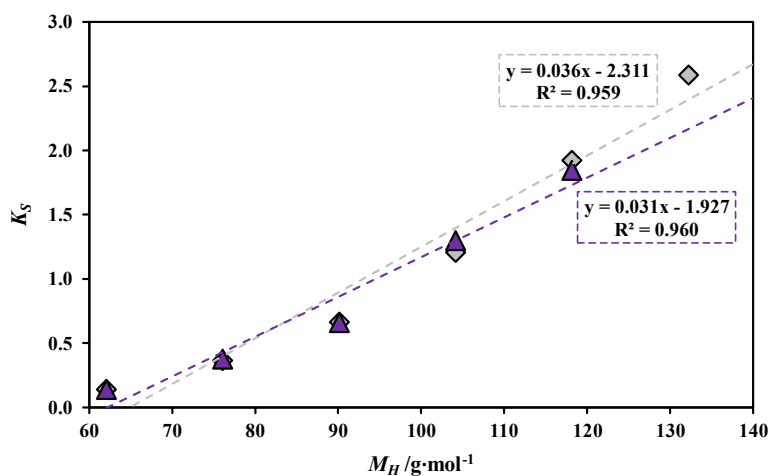


Figure 4.4 reinforces the conclusions presented in the previous section: the ability of each hydrotrope to increase the solubility of syringic acid correlates positively with the size of their alkyl chain. In other words, the larger the apolar volume of the

hydrotrope, the larger the solubility enhancement in the dilute region. Thus, while 1,2-pentanediol is, surprisingly, better than 1,2-hexanediol at higher concentrations, both hydrotropes follow the general apolar trend in the dilute region.

Besides the impact of the apolar volume, the results discussed in the previous section suggest that the relative position of the hydroxyl groups has an impact on the performance of alkanediols as hydrotropes. To evaluate whether this phenomenon is present in the dilute region, the Setschenow constant (slope of each curve depicted in Figure 4.5) is now correlated against a metric that quantifies the apolar volume of the hydrotrope. Given the structural similarity of the alkanediols studied in this work, their molar mass (or their number of carbon atoms) is a good surrogate for their apolar volume. In this sense, Figure 4.5 depicts the Setschenow constants as a function of the molar mass of the hydrotrope, for the two hydrotrope series. The numerical values are reported in Table S5 of the **Appendix D**.

Figure 4.5. Setschenow constant (K_S) as a function of the hydrotrope molar mass (M_H) for 1,2-alkanediols ($\diamond$) and 1,n-alkanediols ($\blacktriangle$). Dashed lines represent the best linear fitting obtained using the least square method.



As expected from the results reported in Figures 4.2 and 4.4, Figure 4.5 reinforces the positive correlation between the performance of alkanediols as hydrotropes (here quantified by the Setschenow constant) and their apolar volume (here quantified by their molar mass). More interestingly, Figure 4.5 reveals that in the dilute region, from where the Setschenow constants were estimated, there are no significant differences in the hydrotropic effect of the two series of alkanediols as the lines practically overlap. In fact, the standard deviations associated with the slopes shown in Figure 4.5 are 0.0043

and 0.0034 for the 1,2 and 1,n-alkanediol sets, respectively. These very small standard deviations, when compared to the difference between the two slopes, show them to be statistically the same at a 5% significance level (significance = 0.512 > 0.05). This is in direct conflict with the observations of the previous section, where the relative position of the hydroxyl groups was found to have an impact on hydrotropy, with 1,2-alkanediols performing better than 1,n-alkanediols for shorter alkyl chains but worse for longer alkyl chains.

The fact that the hydroxyl position appears to impact hydrotropy only for larger concentrations of hydrotrope but not in the dilute region suggests that its effect may stem from interactions that are heightened by the presence of the hydrotrope. In other words, this suggests that hydrotrope-hydrotrope interactions, whose presence is expected to be more significant at higher hydrotrope concentrations, are the reason behind the performance differences between the dilute and non-dilute regions.

It is worth noting that the Setschenow constant, as defined in Equation 4.3, is proportional to the difference between the Kirkwood-Buff (KB) integrals of solute and hydrotrope ($G_{S,H}$) and of solute and water ($G_{S,W}$) (ABBOTT; BOOTH; SHIMIZU, 2016):

$$K_S \propto G_{S,H} - G_{S,W} \quad (4.4)$$

The KB integrals quantify the excess of a component in the local vicinity of another component. As such, $G_{S,H}$ quantifies the excess of hydrotrope around the solute and $G_{S,W}$ quantifies the excess of water around the solute. Put differently, the difference between these KB integrals, which is proportional to K_S , measures the preference of the solute to be surrounded by hydrotrope rather than water. The results depicted in Figure 4.3 show that the Setschenow constant increases with the apolar volume of the hydrotrope. This, once more, supports the current understanding of the mechanism of hydrotropy (ABRANCHES *et al.*, 2020a; SHIMIZU; MATUBAYASI, 2016): as the apolar volume of the hydrotrope increases, so does the driving force for aggregation around the solute. Thus, $G_{S,H}$ increases, leading to larger hydrotrope-solute clusters and better solubility enhancements of the solute.

4.1.4.3. Cooperative Model of Hydrotropy

The results discussed in the previous two sections reveal interesting and somewhat contradictory phenomena:

In the dilute region, the performance of alkanediols correlates with their apolar volume, and the relative position of their hydroxyl groups has no impact;

In the non-dilute region, the performance of alkanediols somewhat correlates with their apolar volume (except for 1,2-hexanediol), yet the relative position of their hydroxyl groups has an impact on their performance, which depends on the size of their alkyl chains (the performance of 1,2-hexanediol is inferior to the performances of 1,2-butanediol and 1,2-pentanediol, and the performance of 1,6-hexanediol is largely superior to that of 1,2-hexanediol).

To shed light on the origins of these discrepancies, a more holistic approach in the form of the cooperative model of hydrotropy is used in this section. This model, which has strong statistical thermodynamics foundations and has been experimentally validated (ABRANCHES *et al.*, 2020a; SHIMIZU; MATUBAYASI, 2016), is used in this section to fit all the syringic acid solubility curves measured in this work. Note that hydrotrope concentrations (solute-free basis) were converted to ternary mole fractions and that the fitting was carried out using the solubility data up to the solubility maxima. These results are depicted in Figure 4.6, and the fittings are reported in Figures S2-S3 and Table S6 of **Appendix D**.

Figure 4.6. Relative solubility enhancement (S/S_0) of syringic acid, at 303.2 K, as a function of ternary hydrotrope mole fraction, for 1,2-alkanediols (left) and 1,n-alkanediols (right), in particular 1,2-ethanediol $\blacklozenge$, 1,2-propanediol $\blacklozenge$, 1,2-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, 1,2-hexanediol $\blacklozenge$, 1,3-propanediol $\blacktriangle$, 1,4-butanediol $\blacktriangle$, 1,5-pentanediol $\blacktriangle$, 1,6-hexanediol $\blacktriangle$ and 1,7-heptanediol $\blacktriangle$. Dashed lines represent the fitted curves obtained using the cooperative model of hydrotropy.

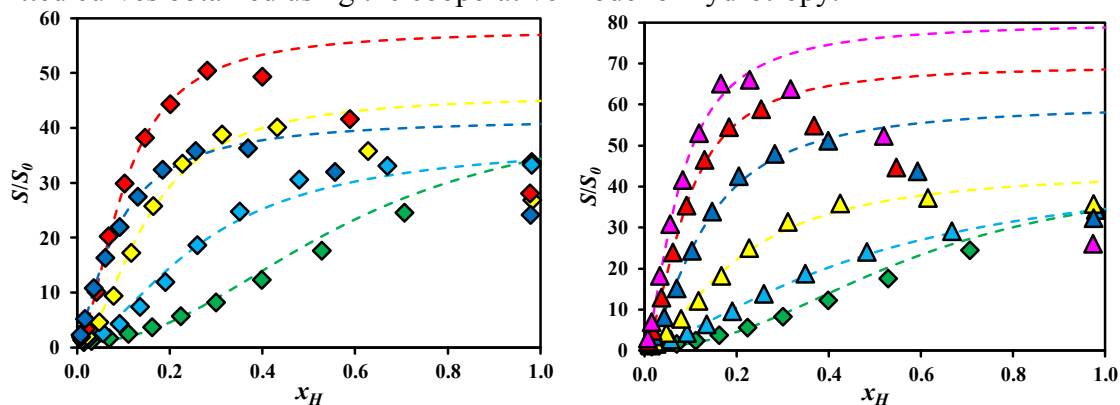


Figure 4.6 shows that the cooperative model of hydrotropy can accurately fit the solubility curves of all solute-hydrotrope pairs, until the solubility maximum of each curve is reached. It should be noted that, within the framework (and limitation) of the model, hydrotropic solubility curves cannot pass through local maxima. Thus, and according to the current understanding of the mechanism of hydrotropy, the maxima displayed by some of the solubility curves reported in this work is related to a change in the solvation environment of the solute. At high concentrations of hydrotrope, water is no longer the major solvent of the system, and water-mediated hydrotrope-solute interactions lose importance, with the system transitioning from a hydrotropy to a co-solvency regime (ABRANCHES *et al.*, 2020b).

The parameter m of the cooperative model of hydrotropy (see Equation 4.1) is related to the average number of hydrotropes that aggregate around a given molecule of solute. For syringic acid and glycerol ethers, this parameter was shown to be non-monotonous with respect to the apolarity of the hydrotrope, reaching a maximum when the apolarities of the solute and hydrotrope match (ABRANCHES *et al.*, 2020a). This was interpreted in light of competing interactions, as follows: from the perspective of water, there is no difference between the apolar moieties of the hydrotrope and the apolar moieties of the solute; as such, when the apolar volume of the hydrotrope is larger than the apolar volume of the solute, the driving force (hydrophobic effect) for hydrotrope-hydrotrope aggregation is larger than the driving force for solute-hydrotrope aggregation. This decreases the average number of available hydrotropes to aggregate around the solute (parameter m). However, this does not necessarily translate into a decrease in hydrotropic efficiency, as even though a more apolar hydrotrope may be statistically less aggregated around a solute, they can cover more of the apolar area of the solute given their larger apolar volume.

To check whether the phenomenon discussed in the previous paragraph is also present in the systems studied in this work, the m parameters here obtained (see Table S6 of **Appendix D**) are now correlated against the apolarity of the alkanediols (in line with Figure 4.5, the apolarity is quantified by the molar mass of the hydrotrope). These results are depicted in Figure 4.7.

Figure 4.7. Parameter m of the cooperative model of hydrotrophy as a function of the hydrotrope molar mass (M_H) for 1,2-alkanediols ($\diamond$), 1,n-alkanediols ($\blacktriangle$), and 1,2-ethanediol ($\bullet$). The dashed lines represent quadratic fittings obtained using the method of least squares.

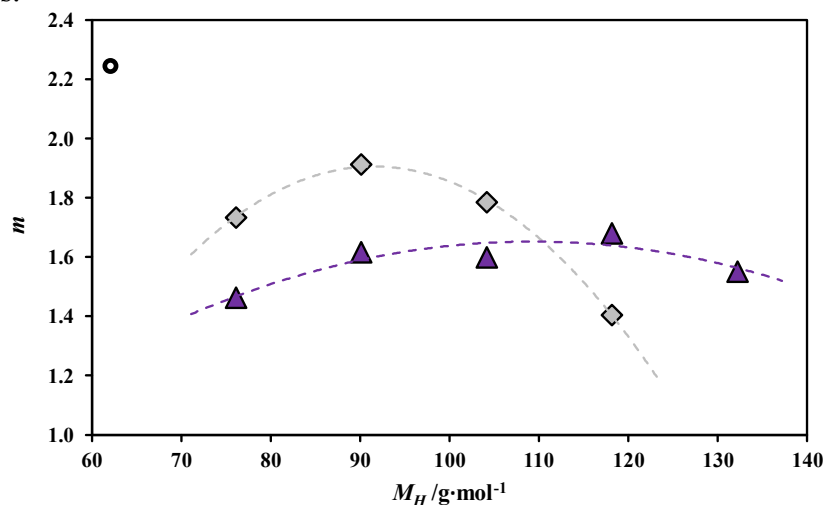


Figure 4.7 reveals several interesting phenomena. To start, parameter m of the cooperative model of hydrotrophy clearly reaches a maximum as a function of the apolar volume of the hydrotrope for both families of alkanediols. This phenomenon has also been observed for monoglycerol ethers (ABRANCHES *et al.*, 2020a), which suggests that this is a general trend of hydrotrophy and not a peculiarity of alkanediols. However, the position and shape of the maxima are different for both series of alkanediols. The maximum for 1,2-alkanediols occurs at a smaller hydrotrope apolar volume when compared to the maximum of the 1,n-alkanediols. Surprisingly, 1,2-ethanediol clearly does not fit the quadratic trend observed in Figure 4.7, which suggests that its ability to enhance the solubility of syringic acid arises from a mechanism other than cooperative hydrotrophy. The nature of this mechanism and how it relates to hydrotrophy will be the object of future work.

The observation made in the previous sections that 1,2-butanediol and 1,2-pentanediol outperform 1,2-hexanediol in the non-dilute region, despite possessing smaller apolar volumes, can now be interpreted considering the results depicted in Figure 4.7. Because m reaches a maximum for 1,2-butanediol in the 1,2-alkanediol series, both 1,2-pentanediol and 1,2-hexanediol are statistically less aggregated around the solute than their less apolar counterparts. If the maximum of parameter m is rationalized in terms of competing interactions as discussed above (the driving force for hydrotrope-hydrotrope aggregation becomes larger than the driving force for solute-hydrotrope aggregation), it

means that the driving force for hydrotrope-hydrotrope aggregation is greater in aqueous solutions of 1,2-pentanediol and 1,2-hexanediol. While for 1,2-pentanediol this is not enough to be noticeable in its performance as a hydrotrope for syringic acid, it is enough to greatly decrease the amount of 1,2-hexanediol aggregated around the solute, leading to lower hydrotropic performance. This also explains why this effect is not seen in the dilute region (Figures 4.3 and 4.4): Since 1,2-hexanediol is outperformed due to its tendency to self-aggregate, this effect is only noticeable when hydrotrope-hydrotrope interactions become more likely (i.e., non-dilute region), and does not impact the dilute region performance.

Having interpreted the discrepancy between the dilute and non-dilute regions regarding the relationship between apolar volume and hydrotrope performance, now the impact of the relative position of the hydroxyl groups is explored. As shown in Figure 4.7, m is generally smaller for 1, n -alkanediols than 1,2-alkanediols. The position of the maximum of this parameter, which is connected to the apolarity of the solute as discussed above (ABRANCHES *et al.*, 2020a), is also shifted, occurring at larger molar masses for the 1, n -alkanediol family. Again considering that the extent of solute-hydrotrope aggregation (quantified by m) is a balance between the driving forces for solute-hydrotrope and hydrotrope-hydrotrope aggregation, these observations suggest that, compared to 1,2-alkanediols, 1, n -alkanediols are either more prone to bulk-phase self-aggregation or less prone to aggregate around the solute.

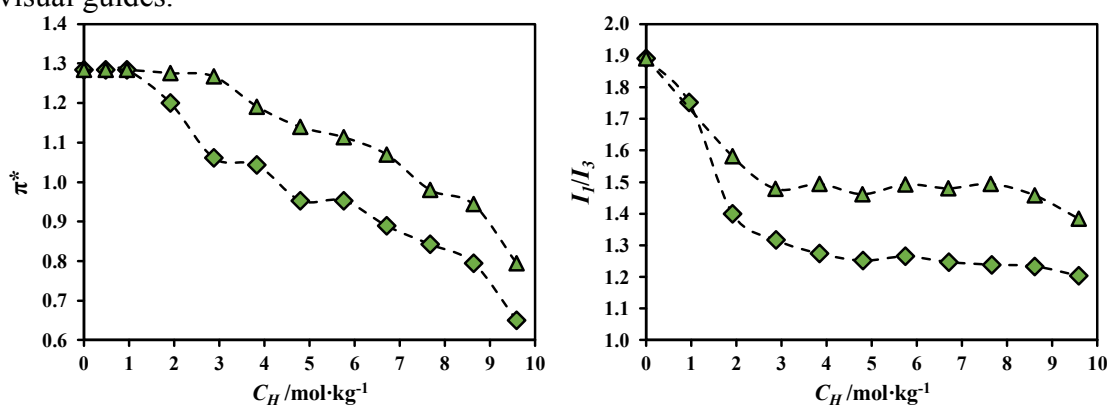
Due to the relative position of their hydroxyl groups, the most energetically favorable dimer of 1, n -alkanediols is an antiparallel conformation, while 1,2-alkanediols are more prone to head-to-head dimerization (YOO; KIM; KANG, 2015). From the standpoint of water, parallel aggregation of large alkyl chains is more favorable than head-to-head because it covers a larger apolar area (hydrophobic effect), maximizing the number of water-water contacts. This, together with the fact that the enthalpies of vaporization of 1, n -alkanediols are superior to those of 1,2-alkanediols (UMNAHANANT *et al.*, 2006; VEREVKIN, 2004), suggests that 1, n -alkanediols are more prone to self-aggregation in water than 1,2-alkanediols. In turn, this suggests that the m discrepancy between both families of alkanediols is related to hydrotrope self-aggregation, not solute-hydrotrope interactions.

4.1.4.4. Polarity Probes and Local Environment

The previous section reconciled the relationship between the apolar volume of the hydrotrope and the solubility enhancement of the solute in the dilute and non-dilute regions while providing a hypothesis to explain the different performances of 1,2-alkanediols and 1,n-alkanediols. Now, two experimental polarity scales are explored to provide further insight on how alkanediols interact with the solute and to validate the conclusions drawn above: the Kamlet-Taft solvatochromic parameter π^* (solvent dipolarity/polarizability) (KAMLET; ABOUD; TAFT, 1977) and the *Py* scale parameter I_1/I_3 (ratio between the first and third bands of the fluorescence spectrum of pyrene) (DONG; WINNIK, 1982).

The polarity scales π^* and I_1/I_3 were experimentally measured in this work, as described in the experimental section, for aqueous solutions of 1,2-pentanediol and of 1,5-pentanediol, in the entire experimentally available concentration range of the hydrotrope, from pure water to pure hydrotrope. These results are depicted in Figure 4.8 and reported in Tables S7 and S8 of **Appendix D**. These two hydrotropes were chosen because they represent a compromise between the position of the maxima of parameter *m* for both alkanediol families and show the lower performance deviation for syringic acid (Figure 4.3).

Figure 4.8. Kamlet-Taft solvatochromic parameter π^* (left) and the *Py* scale parameter I_1/I_3 (right) for aqueous solutions of 1,2-pentanediol (◆) or 1,5-pentanediol (▲). C_H is the concentration of hydrotrope per kg of solution (solute-free basis). Dashed lines are visual guides.



Noting that both the Kamlet-Taft solvatochromic parameter π^* and the *Py* scale parameter I_1/I_3 are proportional to the polarity of the solvent (larger values of π^* or I_1/I_3 represent more polar or less apolar environments), Figure 4.8 unambiguously shows that

aqueous solutions of 1,2-pentanediol are more apolar than aqueous solutions of 1,5-pentanediol, from the perspective of the molecular probe. In other words, the local environment of the probes used to measure these parameters maintains more apolar contacts in aqueous solutions of 1,2-pentanediol than in aqueous solutions of 1,5-pentanediol. This is in line with the hypothesis raised in the previous section to explain the impact of the relative position of hydroxyl groups in the performance of alkanediols as hydrotropes: because 1,5-pentanediol is more prone to self-aggregation in water, its apolar moieties are not as available to aggregate around hydrophobic molecules as those of 1,2-pentanediol. Note that the reduction of solubilization efficacy due to hydrotrope self-aggregation in water has been shown statistical thermodynamically using the Kirkwood-Buff integrals (SHIMIZU, 2020). As such, from a designing standpoint, this shows that hydrotropes should be chosen not only taking into consideration their apolar volume but also their ability to stabilize their self-aggregation in water, which negatively impacts their performance as solubility enhancers.

Acree *et al.* (ACREE *et al.*, 1994; ACREE; TUCKER; WILKINS, 1993) developed a method to estimate the local composition of pyrene in aqueous solutions using the *Py* scale parameter I_1/I_3 , which has been tested for aqueous solutions of alcohols (KUSUMOTO *et al.*, 1997). Briefly, the method assumes that I_1 and I_3 can be estimated by taking the composition average of these values in the pure solvents (water and hydrotrope). In other words:

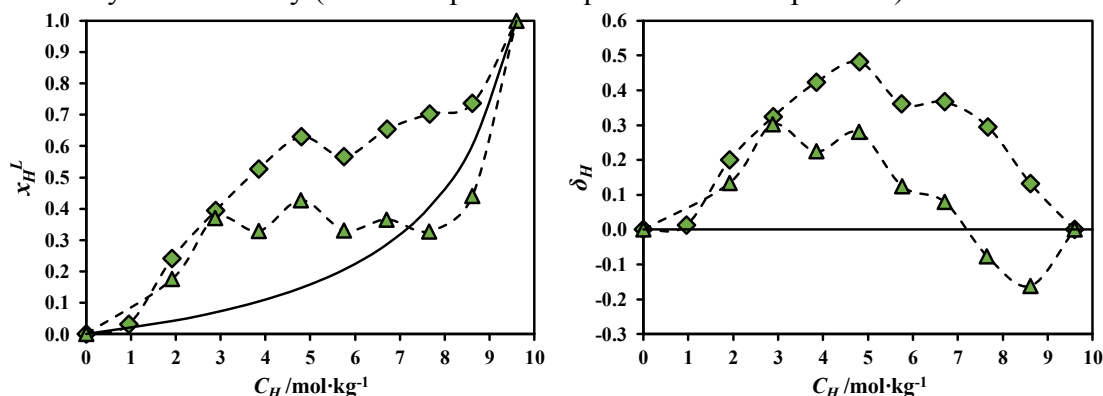
$$\frac{I_1}{I_3} = \frac{x_H^L \cdot I_{1,H} + (1 - x_H^L) \cdot I_{1,W}}{x_H^L \cdot I_{3,H} + (1 - x_H^L) \cdot I_{3,W}} \quad (4.5)$$

where $I_{1,H}$ and $I_{3,H}$ are the intensities of the first and third bands of the fluorescence spectrum of pyrene dissolved in pure hydrotrope, respectively, $I_{1,W}$ and $I_{3,W}$ are the intensities of the first and third bands of the fluorescence spectrum of pyrene dissolved in pure water, respectively, and x_H^L is the local mole fraction of hydrotrope around pyrene in the ternary mixture (water/hydrotrope/pyrene). Using x_H^L , a metric that quantifies the excess of hydrotrope around the solute can be defined in terms of the difference between local and bulk hydrotrope compositions:

$$\delta_H = x_H^L - x_H \quad (4.6)$$

Equations 4.5 and 4.6 were used to calculate x_H^L and δ_H , respectively, and the results are depicted in Figure 4.9 and reported in Tables S10 and S11 of **Appendix D**.

Figure 4.9. Local composition of hydrotrope around the pyrene probe (left) and difference between local and bulk compositions (right) for aqueous solutions of 1,2-pentanediol (◆) or 1,5-pentanediol (▲). C_H is the concentration of hydrotrope per kg of solution (solute-free basis). Dashed lines are visual guides and full lines represent thermodynamic ideality (local composition equal to bulk composition).



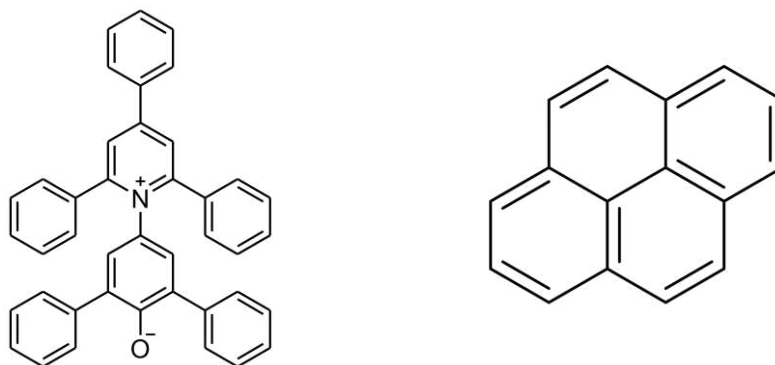
Even though the method used to estimate the local composition of hydrotrope around pyrene is quite sensitive to the uncertainty of the experimental I_1/I_3 data, Figure 9 supports all the conclusions drawn in this work. In particular, the results depicted in Figure 4.9 show the preferential aggregation of hydrotrope around the apolar probe (pyrene): the local composition of hydrotrope is larger than its bulk composition in most of its concentration range. Furthermore, the local composition of 1,2-pentanediol is superior to that of 1,5-pentanediol, reinforcing that 1,n-alkanediols are more prone to self-aggregation and, thus, less available to aggregate around hydrophobic solutes. Finally, the fact that the differences in local composition are mostly seen for larger hydrotrope concentrations reinforces the discrepancies observed between the non-dilute and dilute regions.

4.1.4.5. Highly hydrophobic solutes

Although pyrene was used in the previous section in trace quantities to act as an apolarity probe, the results obtained clearly match the mechanism of hydrotropy (aggregation of hydrotrope around a hydrophobic molecule). As such, it is reasonable to speculate that alkanediols can act as hydrotropes and enhance the aqueous solubility of pyrene. In the final section of this work, the solubility of pyrene in aqueous solutions of 1,2- and 1,5-pentanediols is investigated. Seeing as pyrene is quite more hydrophobic

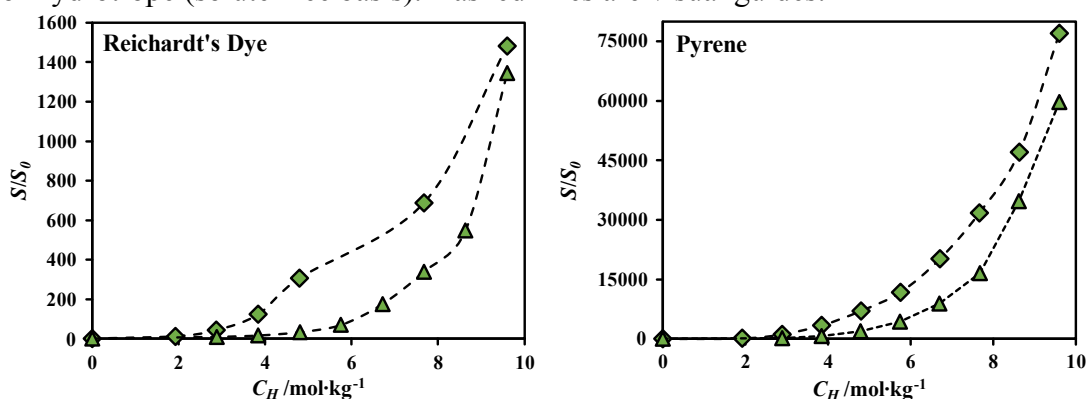
than syringic acid, this presents an opportunity to assess the impact of the hydrophobicity of the solute on the performance of hydrotropes. Taking this into consideration, another solute, Reichardt's dye, was also tested. This solute was chosen due to the ease of its quantification using UV techniques despite its very low water solubility. The chemical structures of these two additional solutes are depicted in Figure 4.10.

Figure 4.10. Chemical structure of Reichardt's dye (left) and pyrene (right).



To start this discussion, the solubility curves of Reichardt's dye and pyrene in aqueous solutions of 1,2-pentanediol and of 1,5-pentanediol, experimentally measured in this work, are depicted in Figure 4.11 and reported in Tables S3 and S4 of **Appendix D**.

Figure 4.11. Solubility enhancement of Reichardt's Dye (left) and pyrene (right) in aqueous solutions of 1,2-pentanediol (◆) or 1,5-pentanediol (▲). C_H is the concentration of hydrotrope (solute-free basis). Dashed lines are visual guides.



As expected, Figure 4.11 shows that alkanediols are able to enhance the aqueous solubility of pyrene and Reichardt's dye. It is noteworthy that, for these more hydrophobic solutes, 1,2-pentanediol clearly performs better than 1,5-pentanediol, in contrast with the results obtained for syringic acid, where the differences are much less noticeable. This can be interpreted by considering the results depicted in Figure 4.7. As previously shown

for glycerol ethers, parameter m of the cooperative model of hydrotrophy reaches a maximum when the apolarities of the solute and hydrotrope match (ABRANCHES *et al.*, 2020a). Because for syringic acid the maximum of m is located at around the apolarities of 1,2- and 1,5-pentanediols, the maxima for these more hydrophobic solutes is expected to only be reached for larger hydrotropes. Thus, the differences in the performance of the pentanediols for these solutes should be similar to those observed in the smaller alkanediols for syringic acid.

4.1.5. Conclusions

To assess the impact of the apolar volume and polar group position of hydrotropes in their ability to enhance the aqueous solubility of hydrophobic substances, two families of alkanediols were used in this work to increase the aqueous solubility of syringic acid. All alkanediols tested were able to act as hydrotropes, and the solubility of syringic acid could be enhanced up to 65-fold for the best-performing hydrotrope (1,7-heptanediol).

Using the empirical Setschenow model in the dilute hydrotrope concentration range, it was found that the ability of a hydrotrope to enhance the aqueous solubility of a hydrophobic substance correlates with its apolar volume, with larger alkanediols being able to achieve higher performances. The relative position of their hydroxyl groups was found to have no impact on their performance in the dilute region. This is in line with the cooperative mechanism of hydrotrophy, which states that water, being highly polar and establishing strong hydrogen bonding with other water molecules, drives the apolar moieties of the hydrotrope to aggregate around hydrophobic solute molecules. On the other hand, in the non-dilute region, the performance of 1,2-hexanediol was found to be inferior to that of 1,2-butanediol and 1,2-pentanediol, and the performance of 1,6-hexanediol was largely superior to that of 1,2-hexanediol. This is intriguing, as the apolar volume of 1,2-hexanediol is larger than the apolar volumes of 1,2-butanediol and 1,2-pentanediol and equal to the apolar volume 1,6-hexanediol.

The contradictory observations made for 1,2-hexanediol were reconciled using the cooperative mechanism of hydrotrophy that showed that the number of hydrotropes aggregated around the solute did not increase linearly with the apolar volume of the hydrotrope, reaching a maximum instead. This maximum was interpreted in light of competing solute-hydrotrope and hydrotrope-hydrotrope interactions. Furthermore, the

model revealed smaller solute-hydrotrope aggregation values for 1,n-alkanediols, which were rationalized in terms of hydrotrope self-aggregation.

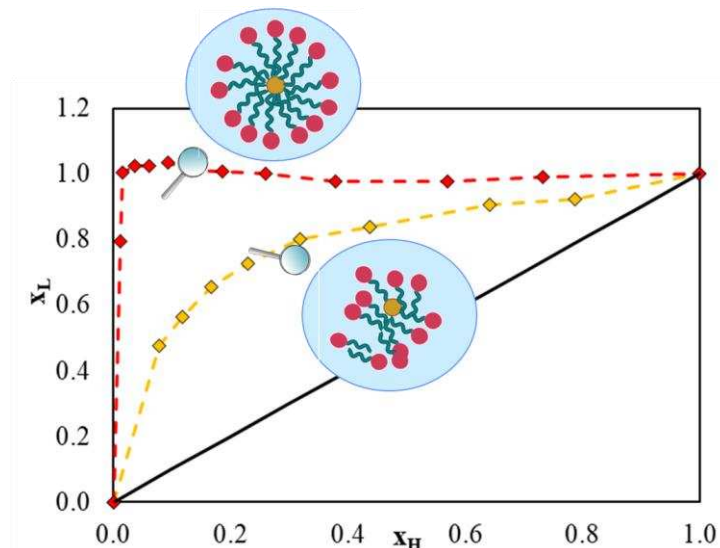
Finally, two chemical environment probes were used to confirm the different degrees of self-aggregation for 1,2-alkanediols and 1,n-alkanediols. These results showed that the extent of solute-hydrotrope aggregation is larger in 1,2-alkanediols than in 1,n-alkanediols, which is most noteworthy for high hydrotrope concentration.

Acknowledgements

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4.2. POLARITY SCALES AS TOOLS TO UNDERSTAND THE HYDROTROPY EFFECT INDUCED BY ALKANEDIOLS

This chapter is unpublished.



Synopsis: polarity scales as tools to access the interactions contributions and aggregation level on the solubilization by alkanediols.

4.2.1. Abstract

The Kamlet–Taft solvatochromic parameter π^* and the P_y polarity scales were measured and correlated in this work with the enhanced solubility. The aim is to assess the contributions of the various interactions (dipolarity/polarizability, hydrogen bonding, etc.) that promote the solute solubilization using alkanediols as hydrotropes. A detailed experimental protocol for the accurate quantification of the P_y scale is described and from that, the local composition was determined and correlated with the degree and nature of the aggregation of alkanediols in aqueous solutions. Good correlations ($R^2 > 0.96$) were found between π^* and P_y for 1,n-alkanediols series due to the higher contribution of dipolarity/polarizability to the solvent-solute solvation in hydrotropy. A transition point, from hydrotropic to micellar aggregation, was identified for 1,2-hexanediol aqueous solutions, which presented local compositions higher than the unit and low correlation between π^* and P_y scales, unlike observed for the remaining systems.

keywords: hydrotrope aggregation; micellar aggregation; P_y scale; solvatochromic parameters; alkanediols; pyrene.

4.2.2. Introduction

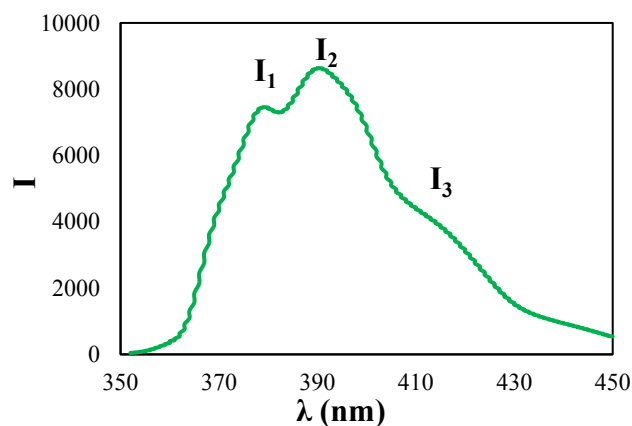
The term “polarity” is defined as the sum of all specific and non-specific interactions between a solvent and a solute in the solvation process without considering the interactions associated with solute chemical transformations (REICHARDT; WELTON, 2011a, 2011b). It is unattainable to describe the multiple solvent-solute solvation interactions using a single parameter, and Kamlet and Taft proposed a multiparametric approach based on a set of solvatochromic probes, allowing the assessment of different interactions specific to each solvent (KAMLET, 1975; KAMLET; ABBOUD; TAFT, 1977; KAMLET; TAFT, 1976). Kamlet–Taft solvatochromic parameters have been studied as a set of the most comprehensive and directly quantitative measures of polarity (JESSOP *et al.*, 2012), namely the parameters α (hydrogen-bond donating ability) (KAMLET; TAFT, 1976), β (hydrogen-bond acceptor ability) (KAMLET, 1975) and π^* (solvent dipolarity/polarizability) (KAMLET; ABBOUD; TAFT, 1977). These three parameters allowed the development of correlations for the description of a variety of properties by the Linear Solvation Energy Relationship (LSER) model, described as:

$$(XYZ) = (XYZ)_0 + s\pi^* + a\alpha + b\beta \quad (4.7)$$

where (XYZ) is the value for a particular solvent-dependent property, (XYZ)₀ is the value for the reference system, and s , a , and b are the solute-dependent coefficients characterizing the respective influence of π^* , α , and β terms on the (XYZ) property under study (KAMLET, 1975; KAMLET; ABBOUD; TAFT, 1977; KAMLET; TAFT, 1976). The polarity of a specific solvent governs its interactions with itself and other compounds, and it is also one of the simplest indicators of solvent strength (FLETCHER *et al.*, 2003), being a key parameter to understand the hydrotrophy phenomenon. In this scenario, solvatochromic probes have been used to establish relations between the solute solubility enhancement and the solvent characteristics, as in the case of some ionic liquids investigated by Sales *et al.* (SALES *et al.*, 2021), in the hydrotropic solubilization of artemisinin. In their work, the authors combined the solvatochromic parameters with solubility measurements, computational simulations, and thermodynamic models and identified the polarity of the medium as the key to artemisinin solubility enhancements.

The *Py* polarity scale, based on pyrene, has been also widely used to study the polarity of the medium and the “preferential solvation” in pure solvents,(DONG; WINNIK, 1982; NAKAJIMA, 1971) surfactants (ALMGREN *et al.*, 1992; NANDI *et al.*, 2013; VASILESCU *et al.*, 1997), lipophilic environments (BELLETTINI *et al.*, 2012; GLUSHKO; THALER; KARP, 1981; MAY *et al.*, 1983), and also water-solvent mixtures (KUSUMOTO *et al.*, 1997; WAKISAKA; KOMATSU; USUI, 2001; ZANA; ELJEBARI, 1993). The *Py* values can be calculated by dividing the intensity of the first peak (I_1) by that of the third vibronic peak (I_3), i.e., $Py = I_1/I_3$ (as depicted in Figure 4.12) and is a function of solvent dielectric constant (ϵ) and refractive index (n) via the dielectric cross term [$f(\epsilon, n^2)$] (FLETCHER *et al.*, 2003).

Figure 4.12. Example (from this work) of fluorescence emission spectra of $4.5 \cdot 10^{-7}$ M pyrene in hydrotrope/water solution with the first three vibronic picks (I_1 , I_2 and I_3).



An increase in the intensity of I_1 occurs in the presence of polar solvents, while only a slight effect occurs in the case of I_3 . Polarity changes in the microenvironment of the probe by, for example, solvent concentration, are easily reflected in a variation in the I_1/I_3 ratio (NANDI *et al.*, 2013). As mentioned in detail in **Chapter 4.1**, Acree *et al.* (ACREE *et al.*, 1994; ACREE; TUCKER; WILKINS, 1993) developed a method to estimate the local composition of pyrene in aqueous solutions using the *Py* scale parameter I_1/I_3 , which has been tested for aqueous solutions of alcohols (KUSUMOTO *et al.*, 1997) and it is an important tool to estimate preferential aggregations of hydrotropes around pyrene (ABRANCHES *et al.*, 2022). Despite being a useful tool, due to the high sensitivity of the methodology, some difficulties in determining precise *Py* values have been reported in the literature, with significant variations of *Py* values obtained by different laboratories (REICHARDT; WELTON, 2011b).

In the previous chapter (**Chapter 4.1**) the solubility results obtained using alkanediols as hydrotropes were modeled and interpreted in light of the Setschenow correlation and cooperative model of hydrotrophy. To provide further insight into how alkanediols interact with the solute, the Kamlet-Taft, (KAMLET; ABBOUD; TAFT, 1977) and *Py* (DONG; WINNIK, 1982) polarity scales were measured for 1,2 and 1,5-pentanediols and used to rationalize the results obtained. Aiming to better understand the impact of the hydrophobicity of the solute in its interactions with the hydrotrope, the solubility of two solutes (aside from syringic acid) with large apolar volumes were also measured for 1,2 and 1,5-pentanediols, namely Reichardt's dye and pyrene.

Aiming to assess the contributions of intermolecular interactions on the solvation environment generated by alkanediols, in this chapter, the studies of the two solvent polarity scales (π^* and *Py*) were extended to all alkanediols studied previously, as well as pyrene solubility measurements. Furthermore, during the implementation of the *Py* polarity scale methodology, improvements were made in the experimental procedure, increasing the reliability of the data and decreasing the sensitivity of the parameters calculated from the experimental measurements. Some modifications were proposed to the experimental measurement of the solvatochromic parameter π^* that does not directly affect the data, only facilitates its measurement.

4.2.3. Material and methods

4.2.3.1. Chemicals

The chemical substances used in this work are listed in Table 4.2, along with their CAS number, source, and mass purity. The water used in all essays was double distilled, passed across a reverse osmosis system, and treated with a Milli-Q plus 185 water purification apparatus.

Table 4.2. List of substances experimentally used in this work, along with their CAS number, supplier, and mass purity.

Substance	CAS number	Source	Purity (wt %)
1,2-ethanediol	107-21-1	Fisher	>99.0
1,2-propanediol	57-55-6	Sigma-Aldrich	99.5
1,3-propanediol	504-63-2	Sigma-Aldrich	>98.0
1,2-butanediol	584-03-2	Sigma-Aldrich	>98.0
1,4-butanediol	110-63-4	Alfa Aesar	>99.0
1,2-pentanediol	5343-92-0	TCI	97.0
1,5-pentanediol	111-29-5	Alfa Aesar	97.0
1,2-hexanediol	6920-22-5	Alfa Aesar	97.0
1,6-hexanediol	629-11-8	Acros Organics	97.0
1,7-heptanediol	629-30-1	TCI	>98.0
ethanol	64-17-5	Fisher Scientific	>99.8
pyrene	129-00-0	Sigma-Aldrich	>99.0
N,N-diethyl-4-nitroaniline	2216-15-1	Fluorochem	>99.0

4.2.3.2. Solubility Curves

The solubility of pyrene in each water/hydrotrope solution was measured using the analytical isothermal shake-flask methodology (HEFTER; TOMKINS, 2003). Pyrene was added in slight excess to each aqueous solution of hydrotrope or pure hydrotrope. The samples were equilibrated in an air oven at (303.2 ± 0.5) K under constant stirring (1050 rpm) for 72 h, using an Eppendorf Thermomixer Comfort equipment. After that, the samples of the liquid phase were carefully collected, filtered using syringe filters, and diluted in water/ethanol 50% (v/v). The amount of pyrene was quantified by UV-spectroscopy using a SHIMADZU UV-1700, Pharma-Spec spectrometer at 335 nm using a pre-determined calibration curve (Figure S1 in **Appendix E**) prepared in water/ethanol at 50% (v/v). Due to the high hydrophobicity of pyrene, its solubility in pure water at 303.2 K was measured and the final average value obtained by eight replicas was 0.184 ± 0.008 mg/L, in accordance with the literature (0.17 mg/L) (MAY *et al.*, 1983).

4.2.3.3. Solvent Polarity Scale

The Kamlet-Taft solvatochromic parameter π^* (solvent dipolarity/polarizability) (KAMLET; ABOUD; TAFT, 1977) and the P_y scale parameter I_1/I_3 (ratio between the first and third bands of the fluorescence spectrum of pyrene) (DONG; WINNIK, 1982) were experimentally measured in this work for all alkanediol/water solutions, excepting

the *Py* scale for 1,7-heptanediol, which at room temperature forms two-phase systems with water, making the measurement impossible.

The solvatochromic parameter π^* was measured using the dye N,N-diethyl-4-nitroaniline at low concentration ($1.7 \cdot 10^{-4}$ mol/L), which guarantee (for alkanediols solutions) the spectra peak reading between the detection limits of absorbance of the UV-Vis equipment. Adequate volumes of N,N-diethyl-4-nitroaniline in ethanol solutions were transferred to empty Eppendorf vessels, and the solvent gently evaporated. The aqueous solutions of alkanediols were then added, and after vigorous stirring, for the complete dissolution of the dye, samples were scanned by the UV-Vis spectrophotometer (BioTeck Synergy HT microplate reader). The longest wavelength absorption band was used to determine the parameter using the following equation:

$$\pi^* = \frac{\nu_{alkanediol} - \nu_{ciclohexane}}{\nu_{DMSO} - \nu_{ciclohexane}} \quad (4.8)$$

where ν is the experimental wave number and the subscripts alkanediol, ciclohexane, and DMSO correspond to the solvent used.

To measure the polarity index (I_1/I_3), fluorescence measurements using pyrene were carried out, as previously described in the literature (DONG; WINNIK, 1982; KUSUMOTO *et al.*, 1997; VASILESCU *et al.*, 1997; ZANA; ELJEBARI, 1993). Since the methodology was described by the first authors some years ago, and the information about the details of the methods was scarce, it was necessary to refine some steps to ensure similar replicas. The fluorescence emission spectra were obtained with pyrene solutions at very low pyrene concentrations ($4.5 \cdot 10^{-7}$ mol/L), which ensured the absence of pyrene excimers. Before preparing the solutions, empty glass vials with screw caps were cleaned in a 10% (v/v) nitric acid bath, passed through distilled water, and oven-dried to ensure contamination-free. After adequate volumes of pyrene in ethanol solution were transferred to the cleaned empty glass vials and the solvent was carefully evaporated at 323.2 K in a water bath. The aqueous solutions of alkanediols were then added, and the glass vials were closed by screw caps. To ensure equilibrium, the spectra were measured after 24 h of orbital/vibro rotation (25 rpm) using a Tryster equipment (Multi Bio RS-24, Biosan), followed by 24 h of constant stirring (2000 rpm) using an Eppendorf

Thermomixer Comfort equipment. The pyrene spectra were quantified using a JASCO FP-8300 spectrofluorometer, collected at 318 nm/min of excitation from 350 to 450 nm width and an emission bandpass of 0.5 nm. The spectra were used to determine the ratio of the intensities of the first and third vibronic peaks of monomeric pyrene (polarity index, I_1/I_3) and the three corresponding vibronic peaks were deconvoluted by ORIGIN 2018.

4.2.4. Results and discussion

4.2.4.1. Solubility Curves

The solubility enhancement of syringic acid in aqueous alkanediols was previously studied in **Chapter 4.1**. Aiming to assess the hydrophobicity solute impact on the performance of hydrotropes the pyrene solubilization in all the selected alkanediols was studied in this chapter, complementing the previous studies. Pyrene was chosen due to its high hydrophobicity, limited variability of interactions, and also to the facility of its quantification using UV techniques. The solubility curves of pyrene in aqueous solutions of alkanediols, experimentally measured in this work, are depicted in Figure 4.13 and reported in Tables S1 and S2 of **Appendix E**.

Figure 4.13. Solubility enhancement of pyrene in aqueous solutions of alkanediols at 303.2 K., namely 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacklozenge$; 1,4-butanediol $\blacklozenge$; 1,5-pentanediol $\blacklozenge$; 1,6-hexanediol $\blacklozenge$ and 1,7-heptanediol $\blacklozenge$. Dashed lines are guides to the eyes.

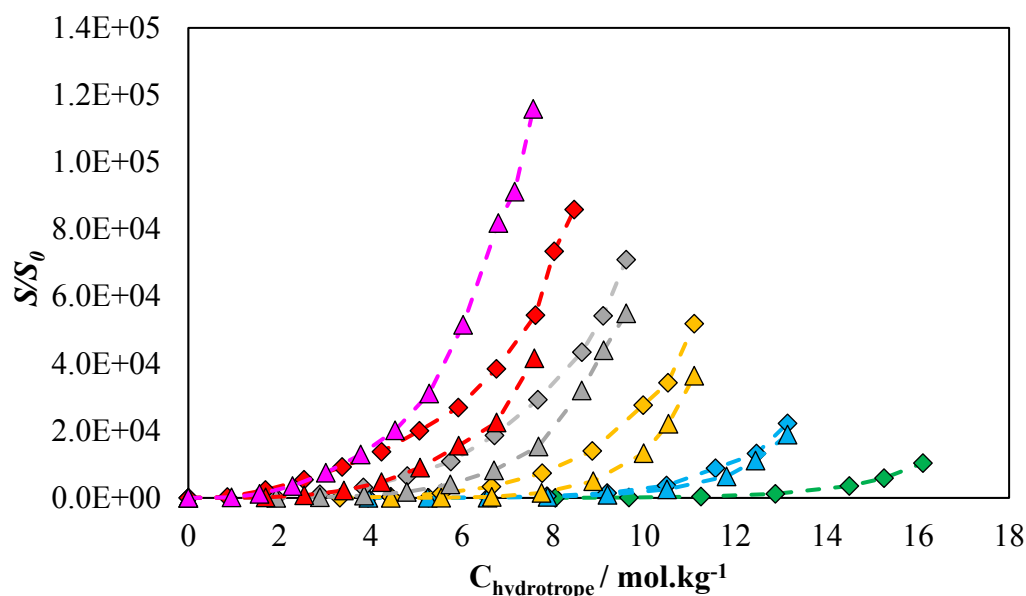
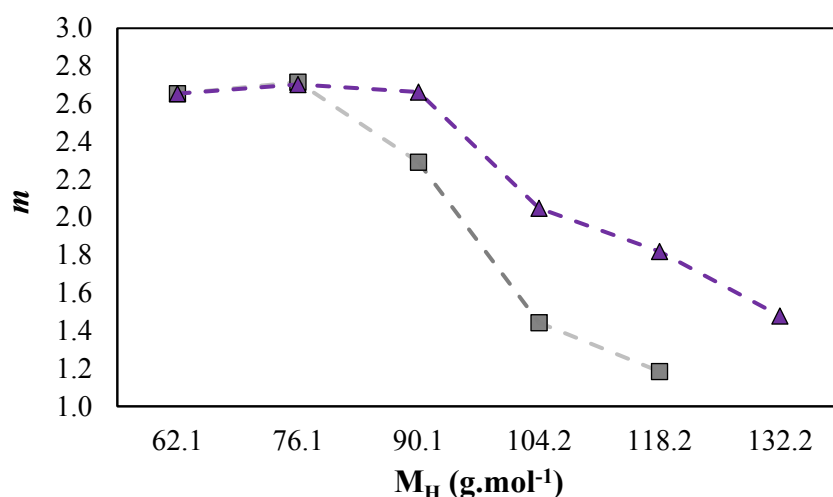


Figure 4.13 shows that alkanediols can enhance the aqueous solubility of pyrene thousands-fold. Increases up to 125,000-fold were found in 1,7-heptanediol, the best studied enhancer. For all alkanediols tested, the solubility enhancement induced by 1,2-alkanediols was higher than those by 1,n-alkanediols. The solubility data obtained in this work was also fitted using the Cooperative Hydrotrope model developed by Shimizu and Matubayasi (SHIMIZU; MATUBAYASI, 2016), described in detail in **Chapter 4.1**. Different from what was expected in the previous chapter, for this highly hydrophobic compound, the parameter m for pyrene does not reach a maximum increasing the apolarity of the alkyl chain (quantified by the molar mass of the hydrotrope), as can be seen in Figure 4.14 and in detail in Table S3 of **Appendix E**.

Contrary to what would be expected based on previous findings, the m value decreases with increasing alkyl chain length, and 1,n-alkanediols showed higher m values than 1,2-alkanediols. In this case, m is rationalized in terms of the ratio of hydrotropes participating in the hydrotrope-pyrene aggregation, then, increasing the apolarity of the alkyl chain, decreases the number of hydrotrope molecules required to solubilize the more hydrophobic solute. In this sense, more molecules of 1,n-alkanediols would be necessary to obtain the maximum attainable solubilization than their correspondents 1,2-alkanediols counterparts.

Figure 4.14. Parameter m of the cooperative model of hydrotropy as a function of the hydrotrope molar mass (M_H) for 1,2-alkanediols (■) and 1,n-alkanediols (▲). Dashed lines are guides to the eyes.



The main differences observed in pyrene and syringic acid solubilization (**Chapter 4.1**) concerning m value, is that the size and shape of pyrene (with four aromatic

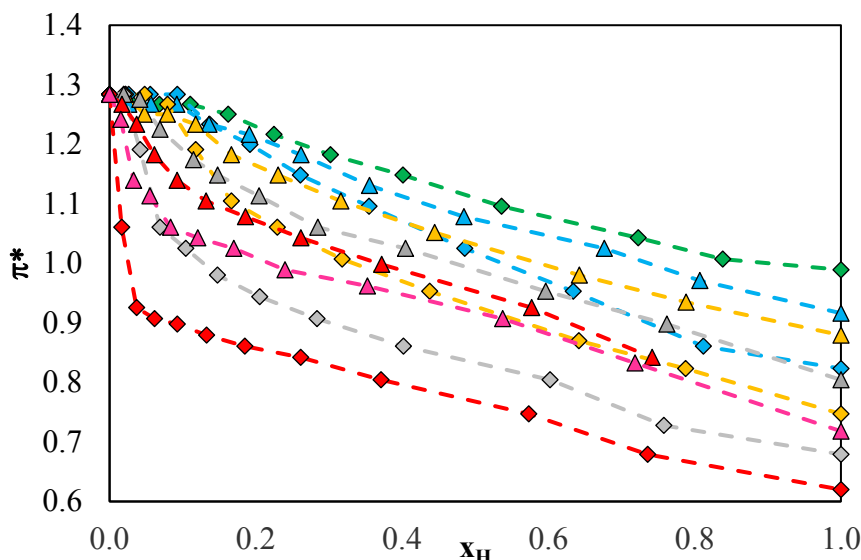
rings), and consequently its higher apolarity, allows only hydrophobic interactions, while syringic acid, containing polar and non-polar groups, both hydrophobic and polar interactions are possible. Besides that, for pyrene, the self-aggregation between pyrene molecules is higher than those for syringic acid molecules. The way the solute is covered by the hydrotrope influences its solubilization (and consequently the m values) since alkanediols can cover all the syringic acid molecule, while with pyrene only the apolar part of the alkyl chain covers the solute. To confirm the aggregation and self-aggregation of the molecules involved in the solubilization medium, other tools such as dynamic light scattering (DLS) or molecular dynamic simulations (MD) would be required.

Therefore, the ability of the hydrotrope to solubilize any hydrophobic solute is a function of its apolarity. In this sense, from the pyrene solubility data is possible to order the alkanediols in a relative hydrophobicity scale from the less apolar to the more apolar, that follows: 1,2-ethanediol < 1,3-propanediol < 1,2-propanediol < 1,4-butanediol < 1,2-butanediol < 1,5-pentanediol < 1,2-pentanediol < 1,6-hexanediol < 1,2-hexanediol < 1,7-heptanediol. From this relative polarity scale, we can see that 1,2-alkanediols would always achieve higher solubility enhancements than the correspondents 1,n-alkanediols, and that the increment of one carbon in the alkyl chain of the 1,n-alkanediols will always exceed the solubility enhancement of the 1,2-alkanediols with one less carbon alkyl chain. Hereupon, the polarity scale is based on the solubility correlation, and the next sections are based on the polarity scales of alkanediols and their contribution to understanding the solvation process.

4.2.4.2. The Polarity Scales

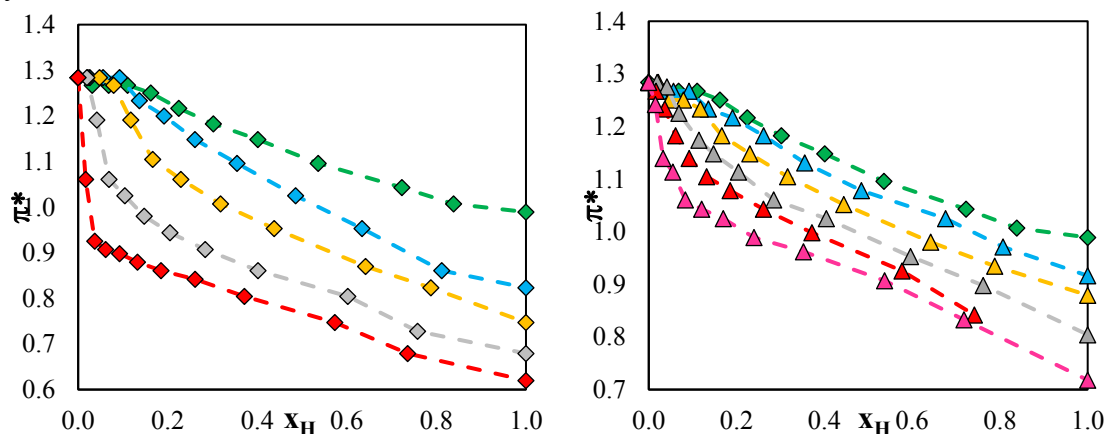
The Kamlet–Taft solvatochromic π^* parameter was also experimentally measured in this work, for aqueous solutions of all alkanediols, in the same concentrations for the solubility measurements, and from pure water to pure hydrotrope, with the exception of pure 1,6-hexanediol (solid at room temperature). The results are depicted in Figure 4.15 and reported in detail in Tables S4 and S5 of **Appendix E**.

Figure 4.15. Kamlet-Taft solvatochromic parameter π^* for aqueous solutions of alkanediols at 298.2 K, namely 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$; 1,6-hexanediol $\blacktriangle$ and 1,7-heptanediol $\blacktriangle$. Dashed lines are guides to the eyes.



Since smaller values of π^* represent more apolar environments, Figure 4.15 shows that 1,2-hexanediol is the more apolar compound and 1,2-ethanediol the less. In fact, 1,2-hexanediol have π^* values lower than 1,7-heptanediol, which contains the larger alkyl chain, this is due to the polarity differences between 1,2 and 1,n-alkanediol families that can be seen in detail in Figure 4.16.

Figure 4.16. Kamlet-Taft solvatochromic parameter π^* for aqueous solutions of alkanediols at 298.2 K, namely 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$; 1,6-hexanediol $\blacktriangle$ and 1,7-heptanediol $\blacktriangle$. Dashed lines are guides to the eyes.

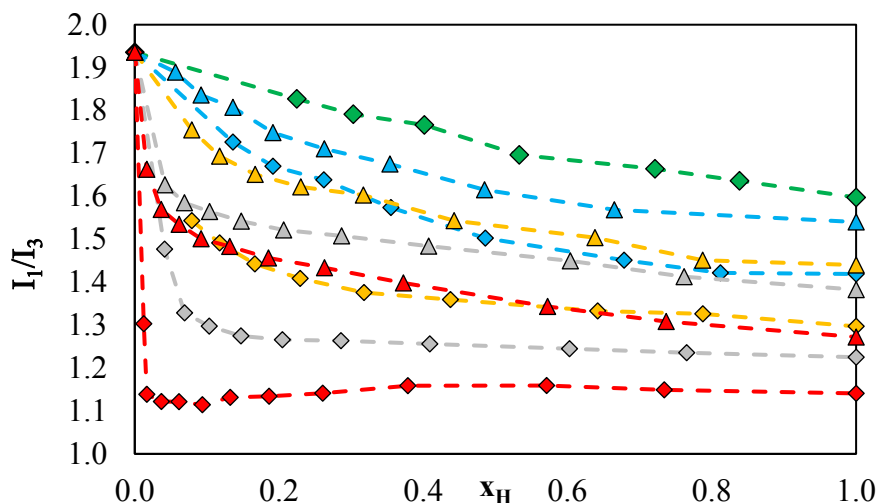


The polarity of a solvent is directly related to its capability for solute dissolution. From the perspective of the molecular π^* probe, aqueous solutions of 1,2-alkanediols are more apolar than aqueous solutions of their corresponding 1,n-alkanediols. Besides that, increasing the concentration of alkanediol increases the apolarity of the medium for all solvents.

It has been stated the nature of the molecular-microscopic probe reflects its emission characteristics like the position and intensity of emission maxima, the vibrational-fine structure (emission of light by an atom in the transition of energy states), the quantum yields, the excited-state lifetime, and the polarization of the fluorescence (REICHARDT; WELTON, 2011c). In the case of the π^* polarity scale, the dye selection can have a dramatic influence on the resulting measurement, since various dyes can be used to determine π^* , increasing the variability of the reported values, as the solvent–dye interaction is unique to each dye (EYCKENS *et al.*, 2016). In this work N,N-diethyl-4-nitroaniline was selected because this dye has found the most widespread use in previously reported determinations of π^* values. Besides that, other dyes such as 4-nitroaniline were tested, but their sparing solubility in pure water prevented measuring the π^* scale in diluted hydrotrope ranges.

Furthermore, hydrotropes possess an amphiphilic character that offers a certain nonpolar interior micro-environment, where the solute is incorporated. The π^* parameter itself, despite ordering the solvents on a polarity scale, does not provide further information regarding the micro-aggregates formed by the solvent-probe solvation. The polarity features of such micro-environments can be readily monitored using fluorescent probes such as pyrene and its Py scale (I_1/I_3). Figure 4.17 shows the variation in the Py scale as a function of the concentration of alkanediols. The data were measured for all alkanediols, except for 1,7-heptanediol/water mixtures (cloudy solutions formed at room temperature), and the data are reported in detail in Tables S6 and S7 of **Appendix E**.

Figure 4.17. *Py* scale (I_1/I_3) for aqueous solutions of alkanediols at 298.2 K, namely 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$ and 1,6-hexanediol $\blacktriangle$. Dashed lines are visual guides.

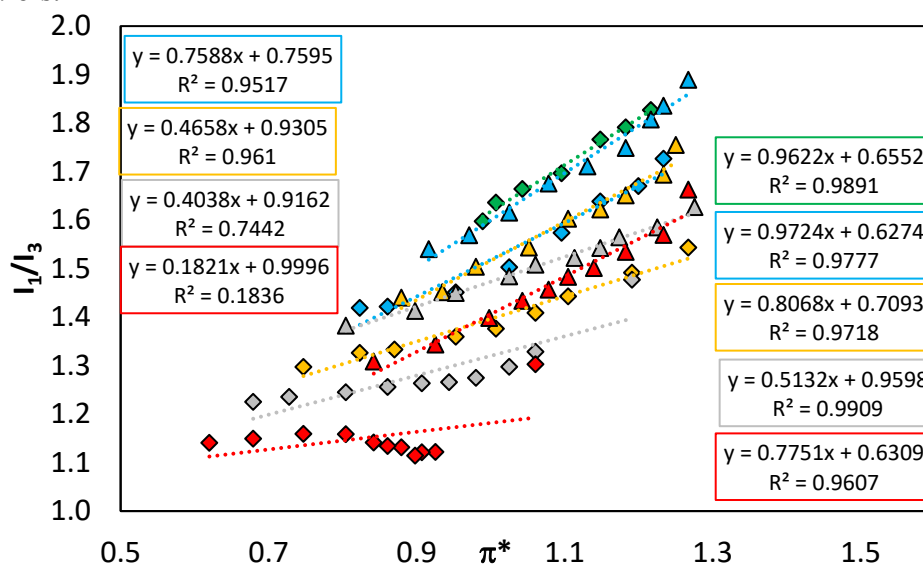


The I_1/I_3 , as happens for π^* , presents higher values for less apolar environments with aqueous 1,2-hexanediol showing the lowest I_1/I_3 values for all concentrations. Figure 4.17 shows that the value of I_1/I_3 decreases with the concentration of alkanediol, suggesting the formation of hydrophobic micro-structures around pyrene.

Different from what was observed for π^* , I_1/I_3 approaches a plateau more quickly for more nonpolar compounds, as is the case of 1,2-pentanediol and 1,2-hexanediol. These results suggest that larger and more compact aggregates are rapidly formed by increasing the alkyl chain length of alkanediols (YIN; CHEN; SHEN, 2019), which will be discussed in detail using the local compositions derived from the *Py* scale. In fact, the self-aggregation of 1,n-alkanediols is higher than in 1,2-alkanediols, negatively impacting their performance as solubility enhancers and offering a micro-environment of relatively lower apolarity. Besides that, 1,n-alkanediols form less compacted aggregates due to their spatial geometry than their 1,2-alkanediol counterparts.

Interestingly, although the π^* scale does not reach the same plateau as the *Py* scale, both show the same polarity behavior, describing alkanediols apolarity in the following order: 1,2-ethanediol < 1,3-propanediol < 1,2-propanediol ~ 1,4-butanediol < 1,5-pentanediol < 1,2-butanediol ~ 1,6-hexanediol < 1,2-pentanediol < 1,2-hexanediol. In fact, for both scales, the polarity of 1,2-propanediol and 1,4-butanediol are similar to the polarity of the pair 1,6-hexanediol and 1,2-butanediol. These similarities can be reconciled by the plot of π^* versus I_1/I_3 , shown in Figure 4.18.

Figure 4.18. Correlation between the *Py* scale (I_1/I_3) and the Kamlet–Taft solvatochromic parameter π^* for aqueous solutions of alkanediols at 298.2 K, namely 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$ and 1,6-hexanediol $\blacktriangle$. Left linear correlations corresponds to 1,2-alkanediols and right linear correlations to 1,n-alkanediols.



It is notorious that *Py* correlates poorly with π^* for some of the alkanediols, namely for 1,2-pentanediol ($R^2=0.74$) and 1,2-hexanediol ($R^2=0.18$), which presented a well-defined plateau for *Py* scale, as mentioned before, due to the high compatibility of the aggregates formed by these solvents in the microenvironment of pyrene.

On the other hand, for 1,n-alkanediols, a strong linear correlation between I_1/I_3 and π^* is observed ($R^2 > 0.96$), demonstrating the dipolarity/polarizability contribution to the solvent-solute interaction. Despite *Py* scale is not sensitive to the hydrogen bonding aspect (DONG; WINNIK, 1982), a residual sensibility is observed (DONG; WINNIK, 1984), since larger values of *Py* represent solvents that are good hydrogen bond donors (e.g. water and 1,2-ethanediol), instead of lower values of *Py* is characteristic of solvents dominated by dispersive interactions (e.g. 1,2-pentanediol and 1,2-hexanediol).

4.2.4.3. The Local Composition

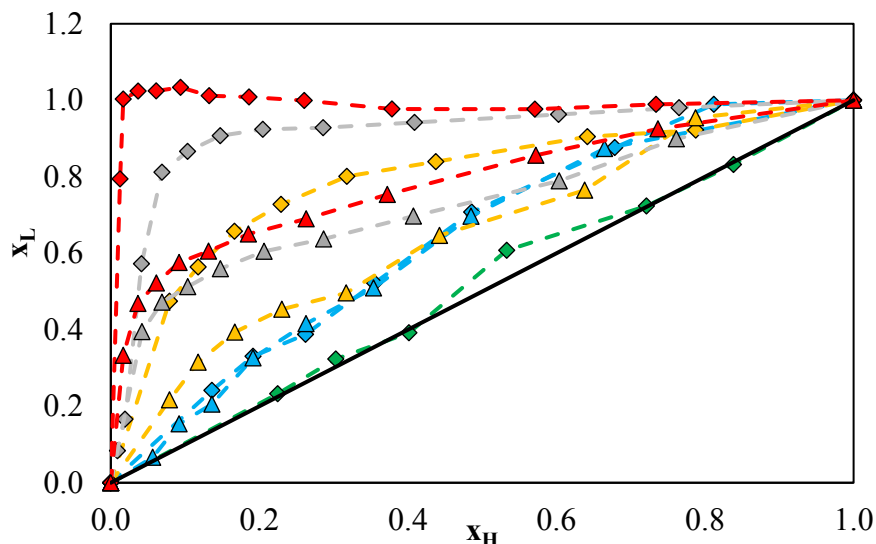
So far, it has been seen that polarity scales provide important information about the intermolecular forces related to the solvation process, but also about the degree of aggregation of molecules around a hydrophobic solute, such as pyrene. However, one of the most intriguing questions in hydrotopy is: *how to identify the switch from hydrotopy*

to micellar behavior? In the case of this work, how long should the alkyl chain be in order for alkanediols to form micellar aggregates?

This intriguing question was also raised by other authors, as in the case of the classical hydrotropes alkylbenzenesulfonate series investigated by Srinivas and Balasubramanian (SRINIVAS; BALASUBRAMANIAN, 1998), and the classical n-alkylbenzoate series studied by Hatzopoulos and co-authors (HOPKINS HATZOPOULOS *et al.*, 2011). Based on their works, despite the thermodynamic properties and related data being useful tools to identify switches in aggregation, no clear transition was identified between hydrotropes and surfactants as a function of substituent alkyl chain length (HOPKINS HATZOPOULOS *et al.*, 2011; SRINIVAS; BALASUBRAMANIAN, 1998).

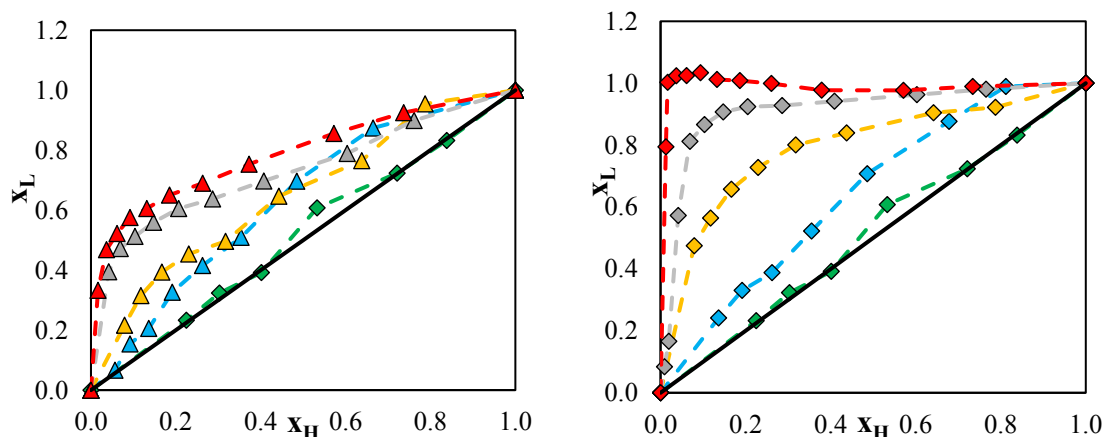
This section presents a tool addressing this question, despite not answering it totally. From the *Py* scale parameter I_1/I_3 , Acree *et al.* (ACREE *et al.*, 1994; ACREE; TUCKER; WILKINS, 1993) developed a method to estimate the local composition of pyrene in aqueous solutions, described in **Chapter 4.1**. For the selected alkanediols studied in this work, the local compositions were calculated and are depicted in Figures 4.19 and 4.20 and reported in Tables S8 and S9 of **Appendix E**.

Figure 4.19. Local composition of hydrotrope (x_L) around the pyrene probe as a function of the bulk alkanediol composition (x_H) at 298.2 K: 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$ and 1,6-hexanediol $\blacktriangle$. Dashed lines are visual guides and the full line represent equal local and bulk compositions.



The local composition of pyrene in aqueous solutions of hydrotrope from the *Py* scale shows the preferential aggregation of hydrotrope around the apolar probe (pyrene) since the local composition of hydrotrope is higher than its bulk composition for all hydrotropes tested, except for 1,2-ethanediol, which the local and bulk compositions are close. Furthermore, as stated before, the local composition of 1,2-alkanediols is superior to that of 1,n-alkanediols, reinforcing that 1,n-alkanediols are more prone to self-aggregation and, thus, less available to aggregate around hydrophobic solutes.

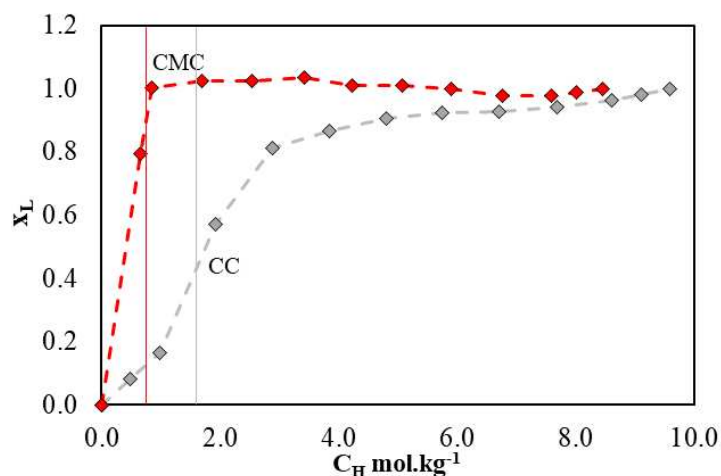
Figure 4.20. Local composition of hydrotrope (x_L) around the pyrene probe as a function of the bulk alkanediol composition (x_H) at 298.2 K: 1,2-ethanediol $\blacklozenge$; 1,2-propanediol $\blacklozenge$; 1,2-butanediol $\blacklozenge$; 1,2-pentanediol $\blacklozenge$; 1,2-hexanediol $\blacklozenge$; 1,3-propanediol $\blacktriangle$; 1,4-butanediol $\blacktriangle$; 1,5-pentanediol $\blacktriangle$ and 1,6-hexanediol $\blacktriangle$. Dashed lines are visual guides and full lines represent thermodynamic ideality (local composition equal to bulk composition).



Remarkably, for 1,2-hexanediol the local composition for most concentrations is around the unit ($x_L \sim 1$), which indicates that the aggregate is a typical micelle-like structure. 1,2-hexanediol presented high I_1/I_3 values, common for traditional surfactant solutions (YIN; CHEN; SHEN, 2019). Moreover, 1,2-hexanediol presents a critical micellar concentration (CMC) in the literature, around 0.82 mol/kg, while 1,2-pentanediol presents a critical concentration (CC) around 1.60 mol/kg (HAJJI, 2003), as depicted in Figure 4.21. The CC for 1,2-pentanediol indicates a transition between the formation of true micelles and simple aggregation. The CMC, in turn, represents that thereafter this concentration, 1,2-hexanediol tends to self-aggregate in a manner very similar to the conventional surfactants, giving rise to micelle-like aggregates. Interestingly, the CMC value coincides with the concentration where $x_L \sim 1$, which demonstrates the ability of the Py scale to identify the transition point of aggregation.

Besides that, as a surfactant is incrementally added to water, I_1/I_3 rapidly falls until a CMC is reached, whereupon I_1/I_3 decreases much more gradually. This behavior originates from a partitioning of the highly hydrophobic pyrene probe into the micellar pseudo-phase, giving rise to a noticeably lower I_1/I_3 at the onset of micelle formation (PAL; SINGH; PANDEY, 2015). This describes the behavior of 1,2-hexanediol, in which the plateau for the Py scale quickly appears at low concentrations (see Figure 4.17).

Figure 4.21. Local composition of hydrotrope around the pyrene probe for aqueous solutions of 1,2-pentanediol $\diamond$ and 1,2-hexanediol $\blacklozenge$ with their corresponding CC (—) and CMC (—) values. Dashed lines are guides to the eyes.



As can be seen in Figure 4.21, it is notorious that both 1,2-pentanediol and 1,2-hexanediol quickly grow to x_L values close to the unit, with 1,2-hexanediol seeming in some cases to exceed this value (possibly due to experimental uncertainty for Py scale, which is ± 0.03). The 1,2-alkanediols, in this sense, apparently form larger and more compact aggregates than 1,n-alkanediols, and from a certain point, increasing the alkyl chain, 1,2-alkanediols may aggregate in a micellar structure.

The results presented above suggest that since the solubility enhancement of solutes has been sensitive to both the apolarity of the solute and hydrotrope, the polarity scales of a solvent (such as π^* and Py) are relevant tools to identify the dominant intermolecular interactions and the degree of aggregation of aqueous solvents since the microenvironmental features display a more sensitive variation with chain length and its respective apolarity. Since the method used to estimate the local composition of hydrotrope around pyrene is quite sensitive to the uncertainty of the experimental I_1/I_3 , it would be interesting to confirm experimentally by dynamic light scattering (DLS), small-angle neutron scattering (SANS), or other measurements, the nature of the 1,2-alkanediol aggregates.

Since many natural and industrial processes take place in solvents, this work disposes of a good collection of data on aqueous alkanediols systems, including solubility measurements, the solvatochromic parameters, and also the useful Py polarity scale.

4.2.5. Conclusions

In this work, we systematically investigated the aggregation behavior of 1,2 and 1,n-alkanediols as hydrotropes, by measuring the solubility curves and using polarity probes, namely the Kamlet–Taft solvatochromic parameters π^* and the P_y polarity scales.

From the solubility curves, the parameter m of the cooperative hydrotrope model was determined, and correlated with the increase in the solubility enhancement, demonstrating that for pyrene more molecules of hydrotropes were required to cover the solute volume for the solubilization process. For all 1,n-alkanediols, strong linear correlations ($R^2 > 0.96$) between π^* and P_y scales were found, which shows that the dipolarity/polarizability is the main contribution to the dissolution process. On the other hand, for some of the 1,2-alkanediols tested, the correlation was poor, particularly for the 1,2-hexanediol, which suggests that other forces influence the solvation. In this case, the formation of micelles may be responsible for this behavior, as the local composition suggests. From those measurements, it was also possible to determine the local composition of alkanediols around pyrene. The results clearly show a transition from hydrotrope aggregation to typical micelle-like aggregation for 1,2-hexanediol. In such case, the local composition is rapidly increasing, confirmed by CMC values from the literature.

To sum up, the polarity scales studied in this work are worthy tools to understand hydrotrope aggregation and to interpret the kind of intermolecular forces relevant to the solubilization enhancement by alkanediols as hydrotropes.

4.3 COSMO-RS PREDICTIONS ON ALKANEDIOLS SOLUBILITY

4.3.1 Preliminary results and perspectives

This subchapter aims to report the predictions by COSMO-RS, obtained for our solubility measurements using alkanediols as hydrotropes.

After considering several assumptions, the solubility of a solid compound in a liquid can be described by the following solid-liquid equilibrium equation (GMEHLING *et al.*, 2019; PRAUSNITZ; LICHTENTHALER; AZEVEDO, 1998):

$$\ln(x_s \gamma_s) = \frac{\Delta_m h}{R} \left(\frac{1}{T_m} - \frac{1}{T} \right) \quad (4.9)$$

where x_s is the saturation mole fraction (solubility) of the solute in the system, γ_s is its activity coefficient, T_m is its melting temperature, $\Delta_m h$ is its enthalpy of fusion (at temperature T_m), R is the ideal gas constant, and T is the absolute temperature of the system.

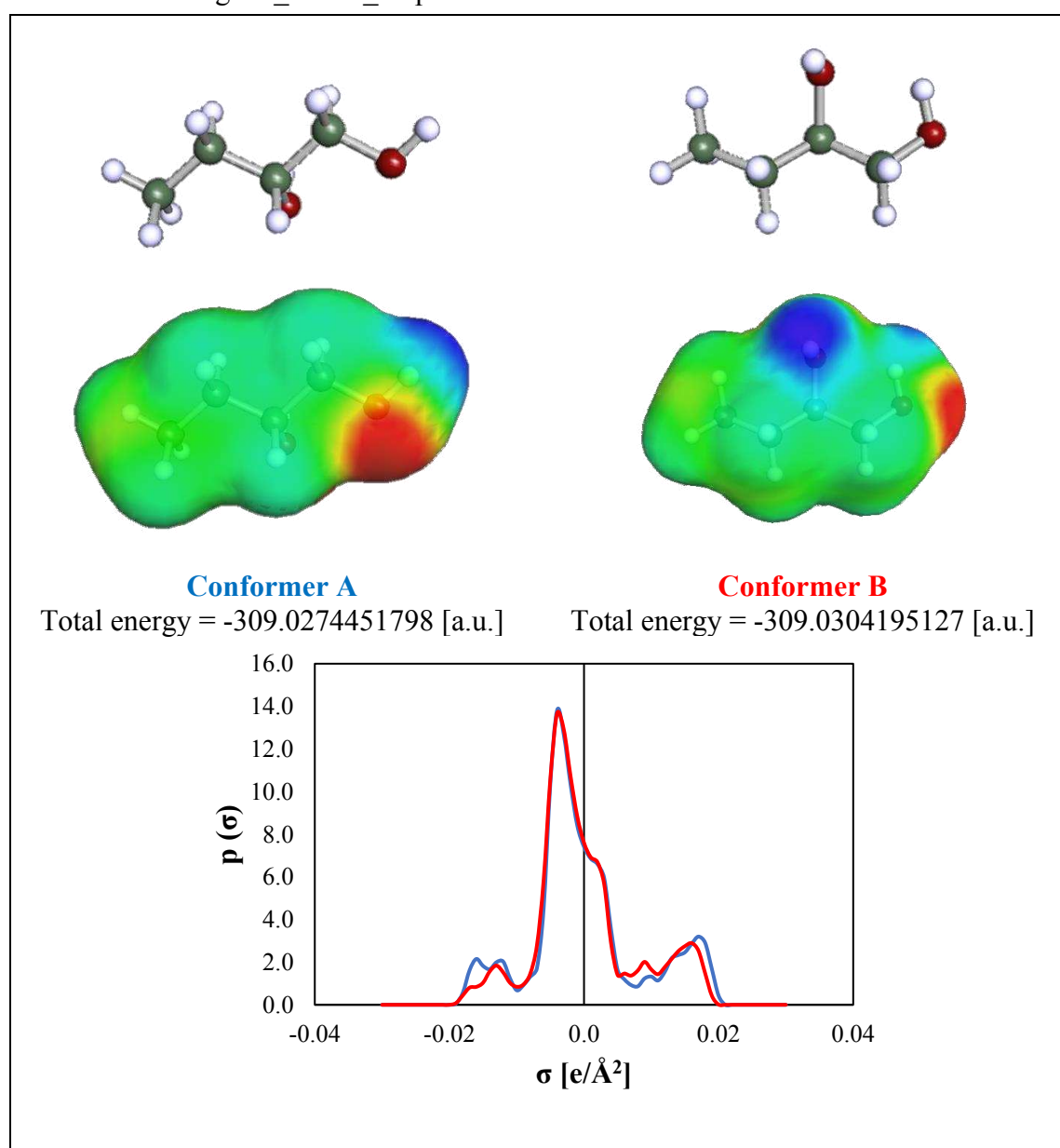
Equation 4.9, when coupled with a thermodynamics model, can be used to predict the solubility of a substance if its fusion properties are known (T_m and $\Delta_m h$). As such, in this work, COSMO-RS is used to predict the activity coefficient of syringic acid and pyrene in all hydrotropic solutions studied and is coupled with Equation 4.9 to predict its solubility. The fusion properties used throughout this work are $T_m = 480.3$ K and $\Delta_m h = 33.7$ kJ·mol⁻¹ (QUEIMADA *et al.*, 2009) and $T_m = 422.4$ K and $\Delta_m h = 16.7$ kJ·mol⁻¹ (ROJAS; OROZCO, 2003) for syringic acid and pyrene respectively.

The COSMO-RS solubility predictions were performed using the software package COSMOtherm with the BP_TZVP_21 parametrization (ECKERT; KLAMT, 2014). Each molecule studied in this work (water, syringic acid, pyrene and all the alkanediols listed in the previous Table 4.1 and their conformers) was optimized using TURBOMOLE, through its interface TmoleX, by adopting the COSMO-BP-TZVP template available in the software package. This template uses a def-TZVP basis set for all atoms, a DFT with the B-P86 functional and the COSMO solvation model (continuum with infinite permittivity).

A molecule prefers to occupy the states of low total energy and arranges its atoms accordingly. Rotating around single bonds (a hydrogen atom, for example), compounds with the same molecular formula can form geometrical isomers, arranging

their atoms in different, non-equivalent positions to each other (FREIRE *et al.*, 2008). For 1,n-alkanediol, one conformer only was tested. In this case, probable intramolecular interactions were not considered due to the position and distance between OH groups. However, for 1,2-alkanediols, two conformers were evaluated since intramolecular interactions can occur. The different conformers, namely different sigma surfaces, sigma profiles, and total energy are shown in Figure 4.22 for 1,2-butanediol. The structures of the other alkanediols are presented in Figures S1-S5 of **Appendix F**.

Figure 4.22. Sigma surface, sigma profile and total energy for different conformers of 1,2-butanediol (conformer A – blue lines and conformer B- red lines) obtained from CosmoThermX using BP_TZVP_21 parametrization.



Figures 4.23 and 4.24 depict the solubility curves of syringic acid in aqueous solutions of alkanediols obtained using COSMO-RS (lines) and the experimental datapoints.

Figure 4.23. Solubility enhancement (x/x_0) in logarithmic scale of syringic acid in aqueous solutions of 1,2-ethanediol $\blacklozenge$, 1,3-propanediol $\blacklozenge$, 1,4-butanediol $\blacklozenge$, 1,5-pentanediol $\blacklozenge$, 1,6-hexanediol $\blacklozenge$, and 1,7-heptanediol $\blacklozenge$ experimentally measured and with the predicted curves (dashed line) obtained from COSMO-RS. The left side corresponds to the entire hydrotrope concentration range and the right side to the more diluted hydrotrope region.

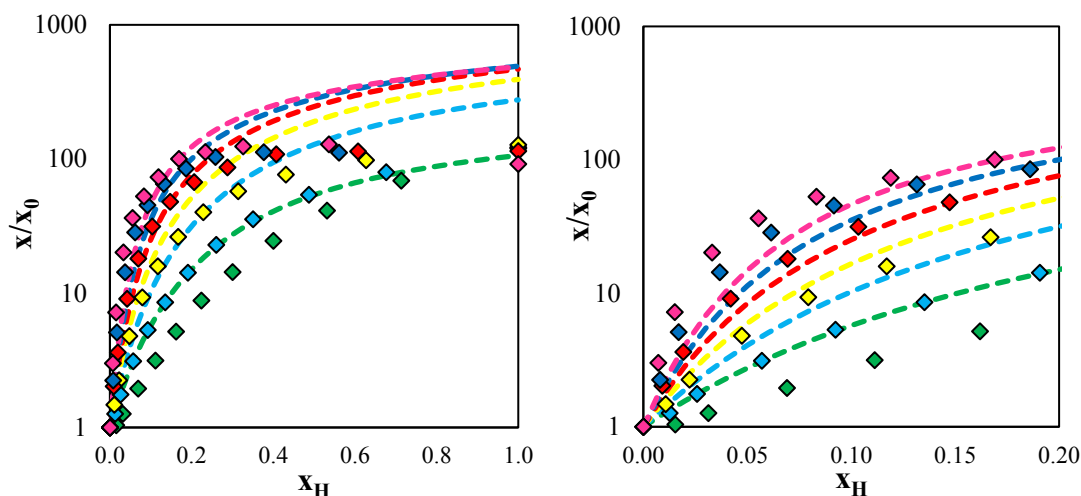
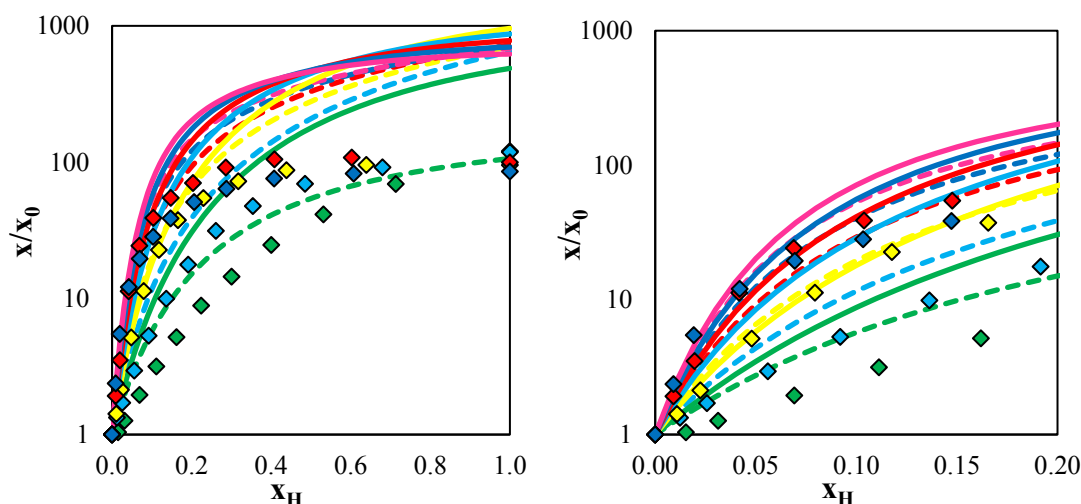


Figure 4.24. Solubility enhancement (x/x_0) in logarithmic scale of syringic acid in aqueous solutions of 1,2-ethanediol $\blacklozenge$, 1,2-propanediol $\blacklozenge$, 1,2-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, 1,2-hexanediol $\blacklozenge$, and 1,2-heptanediol $\blacklozenge$ experimentally measured and for different conformers: dashed (conformer A) and solid lines (conformer B), obtained from COSMO-RS. The left side corresponds to the entire hydrotrope concentration range and the right side to the more diluted hydrotrope region.



The results depicted in Figures 4.23 and 4.24 show that COSMO-RS provides better predictions in the diluted region, showing a significant failure as the concentration of the hydrotrope increases. For both 1,n-alkanediols and 1,2-alkanediols, in the non-dilute region (above 0.2 mole fraction), COSMO-RS could not describe the maxima observed in some solubility enhancement experimental curves, not providing an adequate description of the solubility in the pure hydrotrope whenever it was possible to measure this value. Comparing the predictions for 1,2 and 1,n-alkanediols, 1,n-alkanediols show better results, probably due to a better approximation of the optimized molecule to the real one.

For 1,2-alkanediols, the conformer A (dashed lines in Figure 4.24) provides the best predictions for the solubility enhancement of syringic acid in the diluted region. For conformer A, the H^+ from the hydroxyl group is more polarized if compared to the H^+ from conformer B, what is also demonstrated by the small pick perceptible in the sigma profile of conformer A ($\sigma = -0.017$ – Figure 4.22). In the first case, intermolecular interactions are preferred since the molecule is immersed in a polar medium (water + syringic acid). On the contrary, in conformer B there is an approximation between O-H which favors intramolecular interactions (i.e., internal hydrogen bonding) instead of intermolecular interactions. Interesting, despite the inappreciable difference in the total energy between the two conformers (around 0.003 a.u.) and the similarity of the sigma profiles, the conformer A performed better to predict the solubility data than conformer B, which demonstrates that small differences in the optimization of the molecules to find the best conformation (despite similar low energy), substantially changes the quality of the predict data.

Solubility predictions were made also for pyrene solubilization by alkanediols in aqueous solutions using COSMO-RS. The results are in line with those reported above for syringic acid, as depicted in Figures 4.23 and 4.24, for all the alkanediols studied. In the case of pyrene (see Figures 4.25 and 4.26), the data was shown not in terms of solubility enhancement (x/x_0) but in mole fractions (x). The reason for this change is that the water solubility prediction of pyrene ($x=4.5\cdot 10^{-7}$) is quite higher than the experimentally measured value ($x=1.48\cdot 10^{-8}$). As a consequence, the predictions using the solubility enhancement (x/x_0 scale) are not feasible for comparison purposes. In this

scenario, the description for pyrene in the concentrated hydrotrope region is reasonable, while the prediction in the diluted hydrotrope region worsens.

Figure 4.25. Mole fraction solubility (x) of pyrene in aqueous solutions of 1,2-ethanediol $\blacklozenge$, 1,3-propanediol $\blacklozenge$, 1,4-butanediol $\blacklozenge$, 1,5-pentanediol $\blacklozenge$, 1,6-hexanediol $\blacklozenge$, and 1,7-heptanediol $\blacklozenge$ experimentally measured and with the predicted curves (dashed line) obtained from COSMO-RS. The left side corresponds to the entire hydrotrope concentration range and the right side to the diluted hydrotrope region.

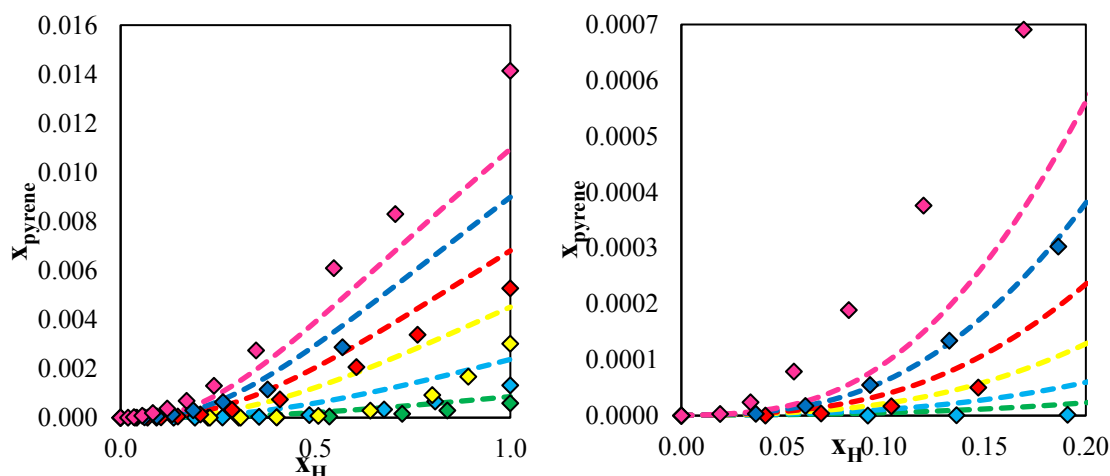
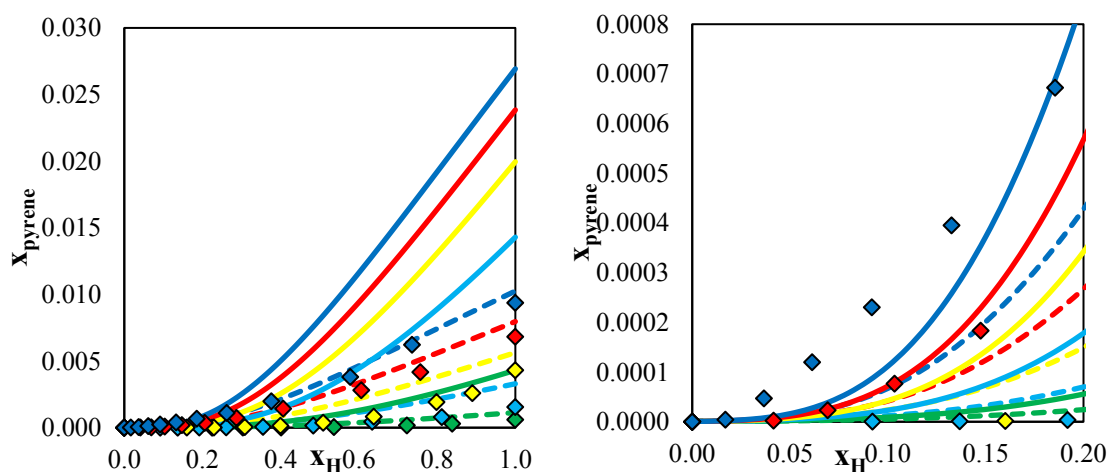


Figure 4.26. Mole fraction solubility (x) of pyrene in aqueous solutions of 1,2-ethanediol $\blacklozenge$, 1,2-propanediol $\blacklozenge$, 1,2-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, 1,2-hexanediol $\blacklozenge$ and 1,2-heptanediol $\blacklozenge$ experimentally measured and for different conformers: dashed (conformer A) and solid lines (conformer B), obtained from COSMO-RS. The left side corresponds to the entire hydrotrope concentration range and the right side to the diluted hydrotrope region.



To sum up, using the conformers tested in this work, COSMO-RS was not able to predict the maximum solubility increase and to cover the hydrotrope concentration

ranges (in diluted or non-diluted regions) for both syringic acid and pyrene. However, the results obtained so far to predict the water solubility of the pure solutes and the solubility enhancements using hydrotropes should be considered preliminary.

In this work, the quantum chemical level BP_TZVP_21 was used, but the BP_TZVPD_FINE_21 parametrization of COSMOtherm can also be applied. The TZVPD_FINE parametrization allows a finer mesh generation with a different hydrogen bond interaction term and a modified van der Waals dispersion term (ECKERT, 2013). Depending on the system studied TZVP or TZVPD_FINE can be selected according to the quality of the predictions. Some preliminary tests using TZVPD_FINE parametrizations are shown in Figures 4.27 and 4.28 for syringic acid and pyrene respectively.

Figure 4.27. Solubility enhancement (x/x_0) in logarithmic scale of syringic acid in aqueous solutions of butanediols $\blacklozenge$ experimentally measured and with the predicted COSMO-RS curves using TZVP (yellow lines) and TZVPD_FINE (orange lines) parametrizations. Different conformers of 1,2-butanediol were distinguish in the right side by dashed lines (conformer A) and solid lines (conformer B).

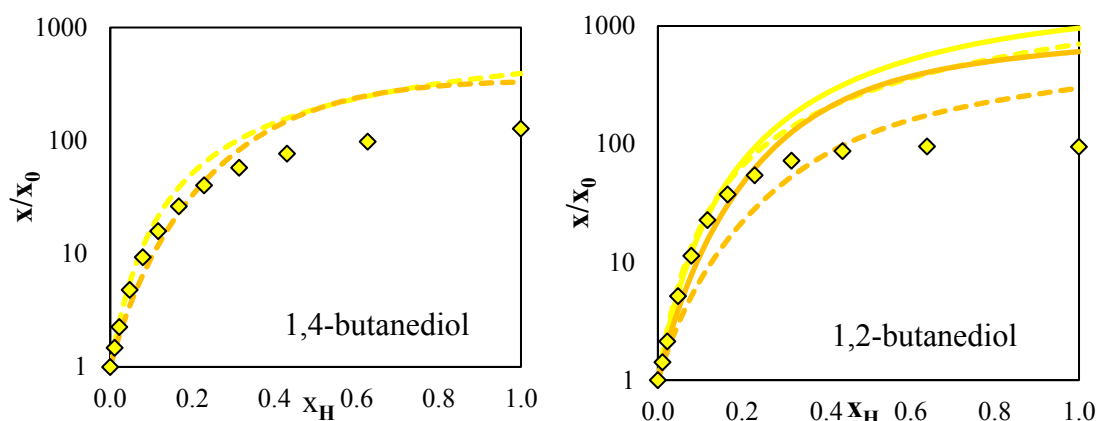
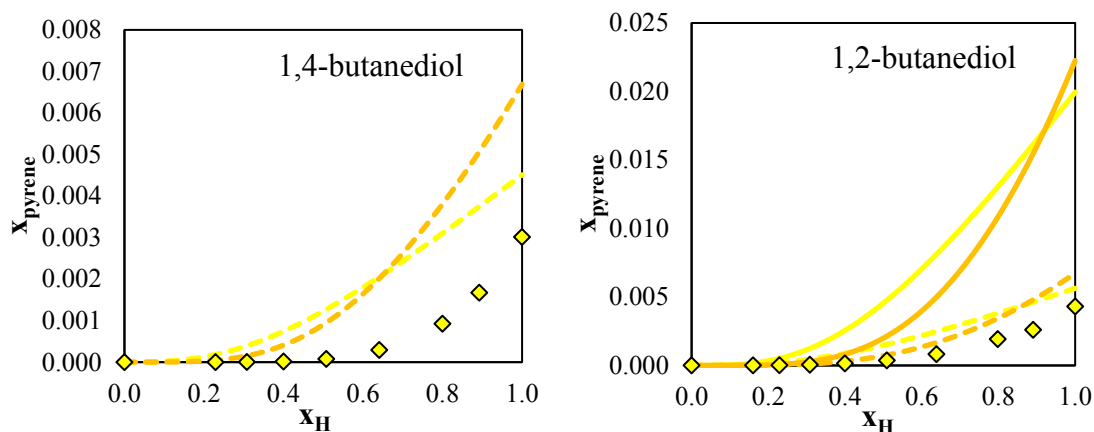


Figure 4.28. Molar solubility (x) of pyrene in aqueous solutions of butanediols $\blacklozenge$ experimentally measured and with the predicted COSMO-RS curves using TZVP (yellow lines) and TZVPD_FINE (orange lines) parametrizations. Different conformers of 1,2-butanediol were distinguish in the right side by dashed lines (conformer A) and solid lines (conformer B).



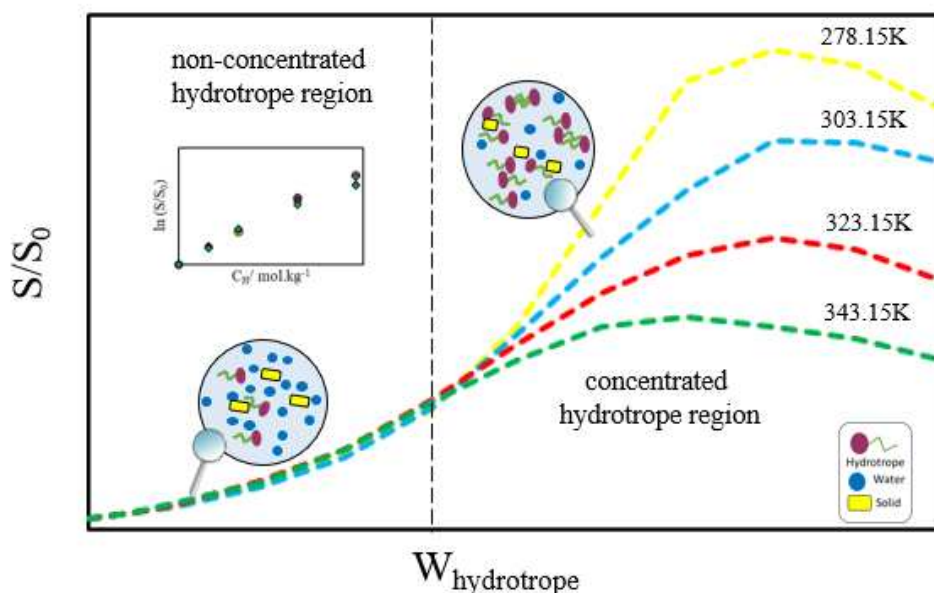
As can be seen, for syringic acid the TZVPD_FINE parametrization allows to predict the solubility data for 1,2-butanediol and 1,4-butanediol slightly better than the TZVP. Besides that, TZVPD_FINE seems to improve the description of the maximum solubility enhancement predictions. For pyrene, TZVPD_FINE does not present substantial differences.

For future work, a more detailed study of the effect of the conformers on the solubility in mixed solvents, including a more judicious choice of the conformers, using COSMOconf, may lead to better predictions. COSMOconf is a flexible toolbox able to generate multiple conformers that allow the selection of the most relevant conformers (with the minimum local energy and higher stability) in various media such as vacuum, polar, or non-polar liquid media. With these modifications, COSMO-RS may be able to describe the solubility curves better. In a near future, these predictions will be a tool to rationalize and interpret the solubility data and other thermodynamic data concerning the hydrotropic phenomena.

5. CHAPTER V - THE TEMPERATURE EFFECT

5.1. THE EFFECT OF TEMPERATURE ON THE SOLUBILITY OF AQUEOUS SOLUTIONS OF NON-IONIC HYDROTROPES

This chapter is unpublished.



Synopsis: two different mechanisms of aggregation observed with the increment in temperature for aqueous solutions of non-ionic hydrotropes

5.1.1. Abstract

The syringic acid solubility in aqueous solutions of six selected non-ionic hydrotropes, namely, 1,2-propanediol, 1,3-butanediol, 1,2-pentanediol, 3-methoxypropan-1,2-diol ([1.0.0]); 2-ethoxyethanol and cyrene was measured at $T = 278.2$ to 343.2 K, at different concentrations from pure water to pure hydrotrope, at atmospheric pressure. In general, the solubility (S , in g/L) was found to increase with the temperature, being glycol ether and 2-ethoxyethanol, the hydrotropes inducing the highest solubility enhancements, increasing the syringic acid solubility more than 195-fold than that in pure water at $T = 278.2$ K. Two different regions were identified: the first region (diluted hydrotrope) is controlled by water dissolution of the solute modified by the hydrotrope, where similar Setschenow constants and solubility enhancements (S/S_0) were found despite temperature variations. The second region (concentrated hydrotrope) is dominated by the hydrotrope dissolution of solute, where the self-aggregation phenomena are more evident, and the S/S_0 is higher for lower temperatures. Based on the van't Hoff equation,

the solution properties (ΔG_{dis}^0), (ΔH_{dis}^0), and (ΔS_{dis}^0) have been estimated, indicating the dissolution process of syringic acid is endothermic and entropy-driven ($\Delta H_{dis}^0 > 0$ and $\Delta S_{dis}^0 > 0$). This analysis paves the way for a rational design and selection of non-ionic hydrotropes for specific applications, in which both temperature and hydrotrope concentration are critical parameters to be considered.

keywords: hydrophobic solute solubilization; non-ionic hydrotropes; Setschenow constant; hydrotrope-hydrotrope interactions; dissolution thermodynamics.

5.1.2. Introduction

Hydrotropes are mild surface active amphiphilic compounds with a hydrophobic part comparatively smaller than that of a conventional surfactant (DHAPTE; MEHTA, 2015). They may be anionic, cationic, or neutral freely soluble organic molecules that considerably enhance the aqueous solubility of hydrophobic compounds (DHAPTE; MEHTA, 2015; SUBBARAO *et al.*, 2012).

There are some main chemical families or compounds that reveal hydrotropic potential. The aromatics, represented by anionic salts of petroleum origin are the most common hydrotropes; sodium benzene sulfonate (SBS), sodium toluene sulfonate (STS), and sodium xylene sulfonate (SXS) being probably the best examples (DHAPTE; MEHTA, 2015; HODGDON; KALER, 2007). Over the years, molecules without an aromatic ring and even without charge, as mentioned before, turned out to act as hydrotropes. The ethylene glycol ethers and the propylene glycol ethers are the most common non-ionic alkyl-hydrotropes (MOLINIER; AUBRY, 2014). In turn, glycerol ethers are also volatile non-ionic hydrotropes (ABRANCHES *et al.*, 2020a; LEBEUF *et al.*, 2016; MOITY *et al.*, 2013; SOARES *et al.*, 2020) that are becoming a promising alternative to commonly used solvo-surfactants such as ethylene or propylene glycol ethers, which are produced from the petrochemical industry. Glycerol ethers show similar properties to glycol ethers and present lower vapor pressures and higher boiling points than their glycol counterparts (QUESTE *et al.*, 2007).

The tunability of the solvent properties of glycerol ethers made them model molecules for studying the mechanisms of action of hydrotropes, as enunciated in the previous **Chapter 3.1** and **3.2**. The same can be said of alkanediols, which are amphiphilic di-alcohols, presenting a tunability character, and are excellent hydrotropic agents

(**Chapter 4.1**). Cyrene, recently proposed as a green solvent, has also been studied as a hydrotrope (DE BRUYN *et al.*, 2019). It is a polar compound that in a certain medium it can be represented in two different structural ways. Abranches and co-authors (ABRANCHES *et al.*, 2020b, 2020c) studied both forms of cyrene (ketone and diol) for the solubilization of hydrophobic solutes and correlated this data to the parameters obtained by the cooperative model of hydrotrope and COSMO-RS predictions to describe the ketone/water/diol equilibrium.

Classical aromatic anionic hydrotropes have been studied from the perspective of thermodynamic properties, particularly concerning the temperature effect (DESAI; PARIKH, 2009; JAYAKUMAR; GANDHI, 2010; WAGLE; KOTHARI; GAIKAR, 2007). However, similar approaches for aqueous solutions of non-charged or non-aromatic hydrotropes have not yet been explored to the same extent, although these compounds are routinely used in industrial applications.

As a continuation of the previous chapters, this chapter addresses the solubility measurements at different temperatures (from 278.2 to 343.2 K) of syringic acid in aqueous solutions of six selected hydrotropes, namely: the three alkanediols 1,2-propanediol, 1,3-butanediol, and 1,5-pentanediol; the glycerol ether 3-methoxypropan-1,2-diol [1.0.0]; the glycol ether 2-ethoxyethanol; and cyrene. Furthermore, to analyze the thermodynamic aspects of solvation, the apparent solution properties (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) of the syringic acid dissolution processes in the six investigated aqueous hydrotropes solutions were estimated by the van't Hoff equation.

5.1.3. Material and Methods

5.1.3.1. Chemicals

The chemical substances used in this work are listed in Table 5.1 along with their CAS number, source, and mass purity. The chemical structure of all hydrotropes studied is depicted in Figure 5.1. Water was double distilled, passed across a reverse osmosis system, and further treated with a Milli-Q plus 185 water purification apparatus. The glycerol ether used [1.0.0] was part of a batch synthesized in a previous work (see **Chapter 3.1**).

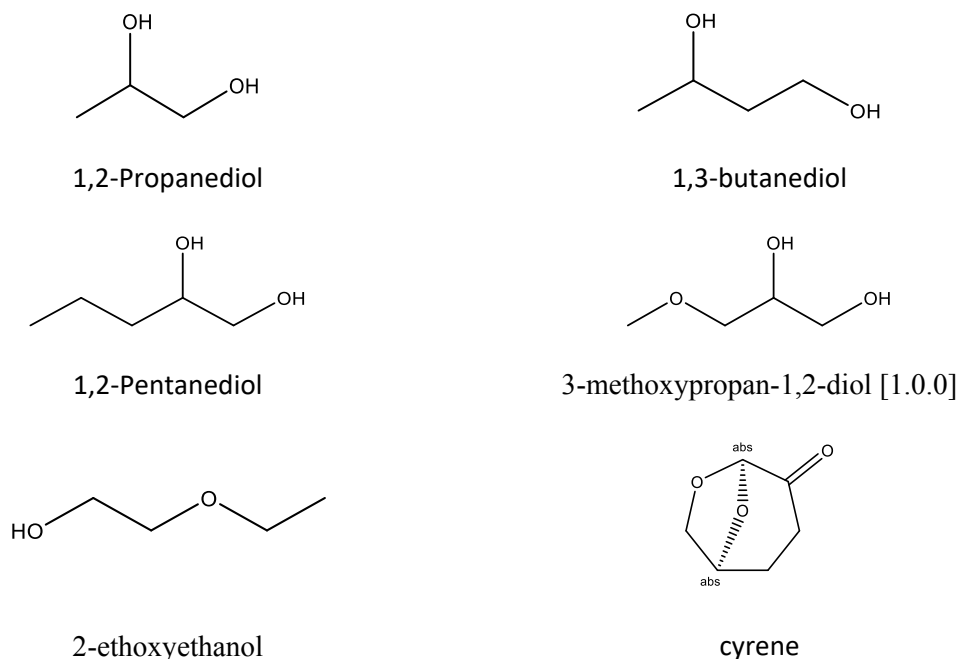
Table 5.1. List of substances experimentally used in this work, their CAS number, supplier, and purity

Substance	CAS number	Source	Purity (wt %)
1,2-propanediol	57-55-6	Sigma-Aldrich	99.5
1,3-butanediol	107-88-0	Sigma-Aldrich	99.5
1,2-pentanediol	5343-92-0	TCI	97.0
3-methoxypropan-1,2-diol [1.0.0]	623-39-2	Synthesized	>99.0
2-ethoxyethanol	110-80-5	Acros Organics	99.0
cyrene	53716-82-8	Sigma-Aldrich	99.0
syringic acid	530-57-4	Acros Organics	>98.0
vannilic acid	121-34-6	Acros Organics	>97.0

5.1.3.2. Solubility Measurements

At atmospheric pressure, syringic acid solubilities in aqueous solutions of hydrotropes (1,2-pentanediol, 1,3-butanediol, 1,2-pentanediol, 3-methoxypropan-1,2-diol, 2-ethoxyethanol and cyrene) at temperatures between 278.2 to 343.2 K were measured using the analytical isothermal shake-flask methodology, previously described in detail elsewhere (HEFTER; TOMKINS, 2003). Syringic acid was added in slight excess to each hydrotrope solution or pure compound. The samples were equilibrated in an air oven at (303.2 ± 0.5) K under constant stirring (1800 rpm) using an Eppendorf Thermomixer Comfort equipment. After at least 72 h (the thermodynamic equilibrium of the systems at higher temperatures required up to 120 h of agitation), the samples of the liquid phase were carefully collected, filtered using syringe filters, and diluted in ultra-pure water. The amount of syringic acid was quantified by UV-spectroscopy using a SHIMADZU UV-1700, Pharma-Spec spectrometer at 267 nm using a pre-determined calibration curve (Figure S1 in **Appendix G**). The systems of water + syringic acid and pure hydrotropes + syringic acid were measured at six different temperatures (278.2, 288.2, 303.2, 323.2, 333.2, and 343.2 K), while the aqueous hydrotrope + syringic acid systems were measured for four different temperatures (278.2, 303.2, 323.2 and 343.2 K).

Figure 5.1. Chemical structure of the hydrotropes used in this work.



5.1.3.3. Thermodynamic properties of dissolution

Standard thermodynamic parameters usually refer to parameters that describe the thermodynamic properties of the dissolution system, and the subtle change of the system state can be expressed by the total differential of the state function (ANIYA *et al.*, 2017; SHA *et al.*, 2021). The thermodynamic properties of the syringic acid dissolution process in aqueous solutions of hydrotropes can be evaluated using the van't Hoff equation as follows (LI *et al.*, 2022a):

$$\Delta H_{dis}^0 = -R \left[\frac{\partial \ln x_1}{\partial (1/T - 1/T_{hm})} \right] = -R \cdot slope \quad (5.1)$$

$$(5.2)$$

$$\Delta G_{dis}^0 = -RT_{hm} \cdot intercept \quad (5.3)$$

$$\Delta S_{dis}^0 = \frac{\Delta H_{dis}^0 - \Delta G_{dis}^0}{T_{hm}} \quad (5.4)$$

$$T_{hm} = \frac{n}{\sum_{i=1}^n \frac{1}{T_i}}$$

herein, T_{hm} is the harmonic mean temperature (K), n denotes the number of different temperature conditions for the solubility (x_l) measurements, and R is the universal gas constant ($R = 8.314 \text{ J}\cdot\text{mol}^{-1} \cdot \text{K}^{-1}$). The *slope* in Eq. (5.1) and *intercept* in Eq. (5.2) can be obtained by the linear regression of $\ln x_l$ versus $(1/T - 1/T_{hm})$.

Additionally, the relative contributions by enthalpy ζ_H and entropy ζ_{TS} toward the standard Gibbs energy change (ΔG_{dis}^0) in the dissolution process can be assessed as:

$$\zeta_H = \frac{|\Delta H_{dis}^0|}{|\Delta H_{dis}^0| + |T_{hm} \cdot \Delta S_{dis}^0|} \quad (5.5)$$

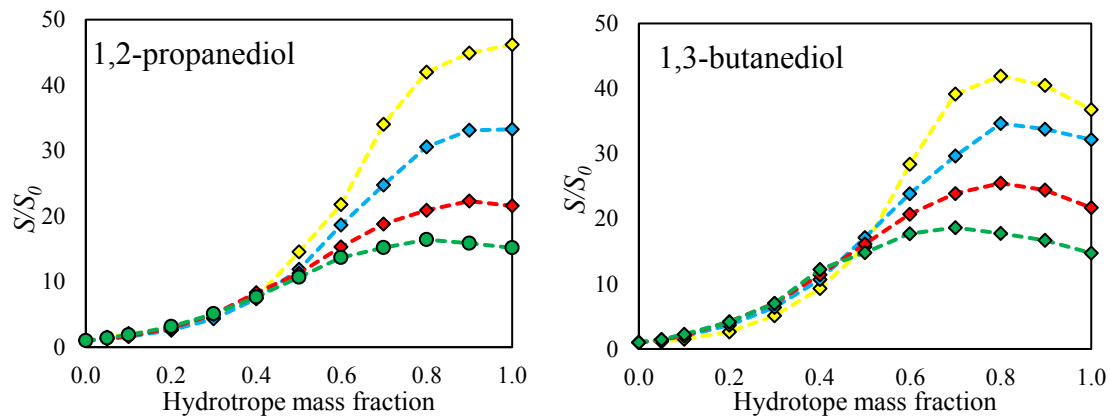
$$\zeta_{TS} = \frac{|T_{hm} \cdot \Delta S_{dis}^0|}{|\Delta H_{dis}^0| + |T_{hm} \cdot \Delta S_{dis}^0|} \quad (5.6)$$

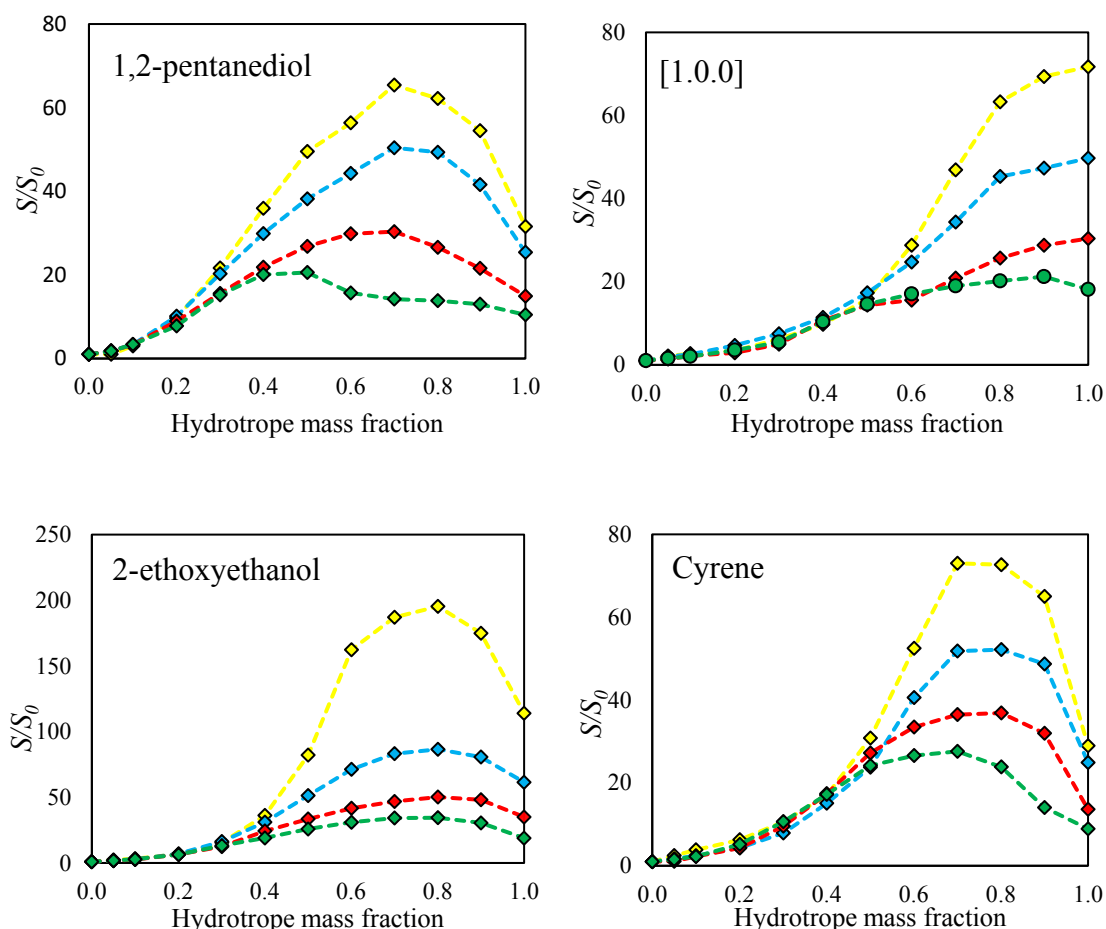
5.1.4. Results and Discussion

5.1.4.1. Solubility Enhancement

The solubility data of syringic acid in the six selected aqueous hydrotropes were experimentally determined in a series of hydrotrope weight fractions (0.0, 0.05, 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9 and 1.0) at $T = 278.2 \text{ K}$ to 343.2 K , as shown in Figure 5.2 and described in detail in Table S1 in **Appendix G**.

Figure 5.2. Solubility enhancement (S/S_0) of syringic acid in aqueous solutions of hydrotropes, namely 1,2-propanediol, 1,3-butanediol, 1,2-pentanediol, [1.0.0], 2-ethoxyethanol and cyrene with various weight fractions at different temperatures (278.2 K $\blacklozenge$, 303.2 K $\blacklozenge$, 323.2 K $\blacklozenge$ and 343.2 K $\blacklozenge$). Lines are guides for the eyes.





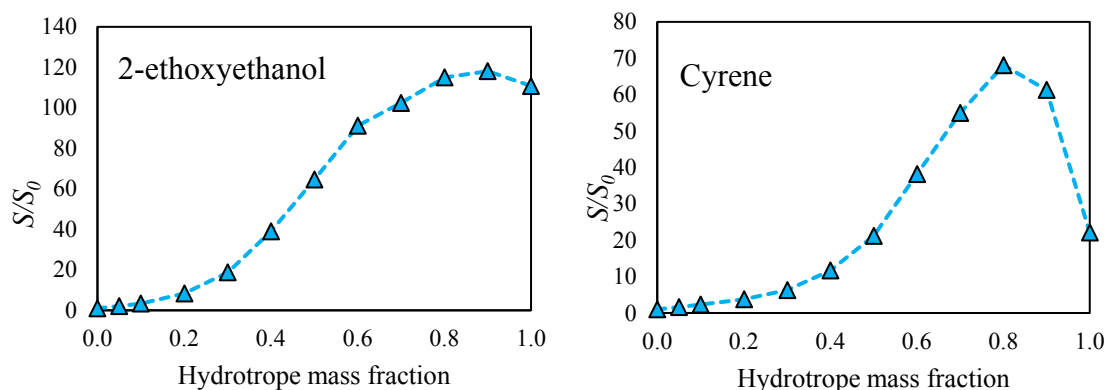
The solubility of syringic acid in the pure solvents follows 2-ethoxyethanol > [1.0.0] > 1,2-propanediol > 1,3-butanediol > 1,2-pentanediol > cyrene > water. In the studied temperature range solubility increases with the temperature, for both pure compounds and aqueous mixtures. A step of 20 K was chosen for the solubility measurements, except for the lowest temperature studied of 278.2 K, which was chosen in order to verify the effect of low temperatures (below room temperature) on the solubility change, and consequently on the effect of hydrotropes on the solute under these conditions. As can be seen in Figure 5.2, for all hydrotropes evaluated, the lower the temperature the higher the increases in solubility enhancement observed. Interestingly, the S/S_0 curves for each compound increase uniformly, except for the glycol ether 2-ethoxyethanol, for which the enhancement observed on the solubility curve at the lowest temperature (278.2 K) is visibly more pronounced than at the other temperatures. In fact, 2-ethoxyethanol achieved for all concentrations, and temperatures, the highest solubility enhancement when compared to the others compounds. From the alkanediols group, the one that showed the highest increases in solubility was 1,2-pentanediol which possesses

the highest apolar volume, as discussed before in **Chapter 4.1** for different families of alkanediols, since the hydrophobic interactions are one of the main driving forces of hydrotrophy.

Interestingly, the solubility curves of 1,3-butanediol, 1,2-pentanediol, 2-ethoxyethanol, and cyrene pass through a maximum at intermediate concentrations, while for 1,2-propanediol and [1.0.0] the maximum solubility was observed for the pure hydrotrope (sigmoidal shape). A different behavior was found for [1.0.0] in the solubilization of gallic acid, which also reached a maximum (SOARES *et al.*, 2020), while in aqueous cyrene solutions the solubility curves showed a maximum of solubility enhancement at different cyrene concentrations (0.5 for salicylic and ferulic acids; 0.6 for benzoic acid; 0.7 for ibuprofen and around 0.4 weight fraction for caffeine).

To compare the results obtained with solutes showing different hydrophilicity, the solubility curves for vanillic acid at 303.2 K for 2-ethoxyethanol and cyrene were also measured, and the results depicted in Figure 5.3 (in detail in the Table S2 of **Appendix G**). The same tendency for syringic acid was found for vanillic acid solubilization by cyrene, with the maximum S/S_0 at 0.9 weight fraction and a different maximum for 2-ethoxyethanol (0.9 for vanillic acid and 0.8 weight fraction for syringic acid) was observed. Despite that, the S/S_0 for vanillic acid was higher than those found for syringic acid for both cyrene and 2-ethoxyethanol. The differences observed were a result of the apolarity of the solutes, which is also one of the key factors in hydrotrophy, since the hydrotropic phenomena are maximized when the apolarities of the solute and the hydrotrope match (ABRANCHES *et al.*, 2020a; SOARES *et al.*, 2020).

Figure 5.3. Solubility enhancement (S/S_0) of vanillic acid in aqueous solutions of 2-ethoxyethanol and cyrene with various weight fractions at 303.2 K. Lines are guides to the eyes.



It is also important to mention that the concentration of the maximum solubility enhancement for the six hydrotropes studied was not much influenced by temperature, with the exception of 1,2-pentanediol at 343.2 K, where the maximum was obtained for 0.5 mass fraction, while for the other temperatures it was achieved close to 0.7 mass fraction. As previously reported in the **Chapter 4.1**, the driving force for self-aggregation is greater in aqueous solutions of 1,2-pentanediol than in their less apolar counterparts. The self-aggregation effect is only noticeable when hydrotrope-hydrotrope interactions become more likely (i.e., concentrated-hydrotrope region) and negatively impact the solvent performance as a hydrotrope. For 1,2-pentanediol the increase in temperature seems to increase the self-aggregation. The temperature effect on solubility may be explored by applying thermodynamic models such as the cooperative model of hydrotropy (SHIMIZU; MATUBAYASI, 2016) and chemical environment probes to confirm the different degrees of self-aggregation (ABRANCHES *et al.*, 2022).

In Figure 5.2 is possible to observe two well-defined regions. One is in the non-concentrated hydrotrope region, where the solubility increase is smaller, and the curves practically overlap. A different behavior is observed in the concentrated hydrotrope region, where the increase in solubility starts to differentiate and become more pronounced with decreasing temperature. These two regions will be the focus of the next section.

5.1.4.2. The non-concentrated and concentrated hydrotrope regions

The Setschenow constant (SETSCHENOW, 1989) (K_S) can be considered a measure of the effectiveness of a hydrotrope, at any given conditions of hydrotrope concentration and system temperature. In this work, it is defined as:

$$\ln(S/S_0) = K_S \cdot C_H \quad (5.7)$$

where S and S_0 are the molar solubilities (mol/L) of the solute in the aqueous hydrotrope solution and pure water, respectively, and C_H is the concentration (mol/kg_{solution}) of the hydrotrope on a solute-free basis. Analyzing the non-concentrated hydrotrope region for each compound (up to 40 wt%) by the Setschenow constant, each solubility curve previously reported was fitted using Eq. (5.7). The experimental results obtained in the

diluted region are depicted in Figure 5.4 and the Setschenow constants showed in Table 5.2.

Figure 5.4. Natural logarithm of the relative solubility enhancement of syringic acid, at different temperatures (278.2 K $\blacklozenge$, 303.2 K $\blacklozenge$, 323.2 K $\blacklozenge$ and 343.2 K $\blacklozenge$) as a function of the hydrotrope concentration, namely 1,2-propanediol, 1,3-butanediol, 1,2-pentenediol, [1.0.0], 2-ethoxyethanol and cyrene.

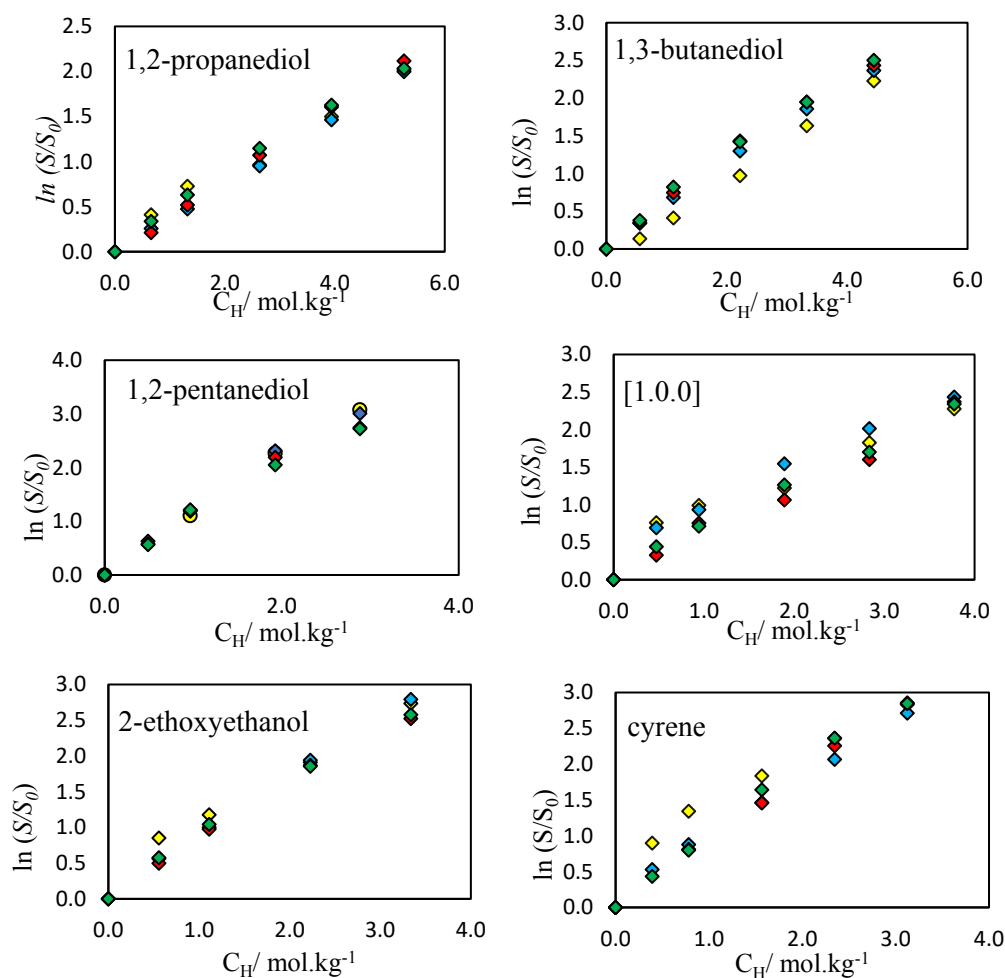


Figure 5.4 reveals that in the non-concentrated hydrotrope region, there are no significant differences in the hydrotropic effect, as the fitted models practically overlap for all compounds due to temperature changes. This can be checked by the K_S values presented in Table 5.2. The difference between the K_S for different temperatures is $< 7\%$ (corresponding to the ratio between standard deviation and average). These small differences between the K_S values seem to agree with the observation of the solubility curves presented in Figure 5.2, where despite the temperature increase there are no significant differences in the solubility enhancement.

It is important to highlight that the Eq. (5.7), is a linear correlation between $\ln(S/S_0)$ and C_H and is only valid at low values of C_H when these variables possess a linear relationship, as confirmed by the R-squared around 1.00 for all compounds and all temperatures showed in the Table 5.2. Since the Setschenow constant is similar at different temperatures it can be established that, for each compound, K_s follows the order: 1,2-pentanediol > cyrene > 2-ethoxyethanol > [1.0.0] > 1,3-butanediol > 1,2-propanediol. Interestingly, the values obtained in the dilute region are in contradiction with the initial qualitative analysis from Figure 5.2, in which 2-ethoxyethanol showed the highest solubility enhancement. These results are, however, in agreement with our previous work, regarding the use of glycerol ethers as hydrotropes discussed in **Chapter 3.2** that suggested the apolar volume of a hydrotrope to be directly connected with its capability to enhance the solubility of a solute in the non-concentrated hydrotrope region. In fact, the progressive increase, through the presence of methyl groups (in the linear chain hydrotropes) seems to correlate positively with the Setschenow constants obtained.

Table 5.2.. Setschenow constant (K_s) and determination coefficient (R^2) values for the fitted linear model, at different temperatures, for the hydrotropes studied in this work.

Compound	T (K)	K_s (kg.mol ⁻¹)	$\overline{K_s}$ (kg.mol ⁻¹)	$\pm s$ (kg.mol ⁻¹)	R^2
1,2-propanediol ($w_H = 0.4$)	278.2	0.359	0.383	0.021	0.983
	303.2	0.377			0.999
	323.2	0.409			0.999
	343.2	0.385			0.994
1,3-butanediol ($w_H = 0.4$)	278.2	0.518	0.540	0.018	0.994
	303.2	0.533			0.997
	323.2	0.552			0.992
	343.2	0.556			0.993
1,2-pentanediol ($w_H = 0.2$)	278.2	1.177	1.145	0.057	1.000
	303.2	1.196			0.999
	323.2	1.138			0.998
	343.2	1.067			0.990
[1.0.0] ($w_H = 0.4$)	278.2	0.537	0.582	0.031	0.945
	303.2	0.608			0.968
	323.2	0.591			0.985
	343.2	0.591			0.993
2-ethoxyethanol ($w_H = 0.3$)	278.2	0.768	0.780	0.034	0.976
	303.2	0.830			0.998
	323.2	0.760			0.994
	343.2	0.760			0.993
cyrene ($w_H = 0.4$)	278.2	0.825	0.876	0.055	0.939
	303.2	0.833			0.993
	323.2	0.916			0.998
	343.2	0.930			0.993

5.1.4.3. Thermodynamic properties of dissolution

For the calculations of thermodynamic properties of dissolution, the density of the corresponding hydrotropes was required to obtain de solubility (in x_1 units). For each compound the quadratic or linear correlation of density as a function of temperature was estimated from the literature data for 1,2-propanediol (GEORGE; SASTRY, 2003; LI; SU; WANG, 2007), 1,3-butanediol (GEORGE; SASTRY, 2003; NAIN, 2007), 1,2-pentanediol (PARSA; HAGHRO, 2008), [1.0.0] (LEAL-DUASO *et al.*, 2017a), 2-ethoxyethanol (SHI *et al.*, 2019) and cyrene (BAIRD *et al.*, 2019). The corresponding density correlations and determination coefficients are given in Table S3 in **Appendix G**.

To estimate the thermodynamic properties of dissolution for syringic acid in the six selected aqueous hydrotrope solutions and pure hydrotropes, all the plots of $\ln x_1$ vs. $(1/T-1/T_{hm})$ must show good linear correlations. In this sense, since (ΔH_{dis}^0) and (ΔS_{dis}^0) are determined by derivation, in the dataset of hydrotrope + water mixtures, a minimum change of 0.01 in R^2 can introduce an uncertainty up to 60% of the mean value for the enthalpy of dissolution, and even higher for the entropy of dissolution. Furthermore, solubility data were collected just at four temperatures (278.2, 303.2, 323.2, and 343.2 K), and since syringic acid is a hydrophobic solute, solubility measurements precision is a challenge. For these reasons, the accuracy of the values determined for (ΔH_{dis}^0) and (ΔS_{dis}^0) have high uncertainty. For this dataset (hydrotrope + water) the values obtained from the van't Hoff equation will be analyzed as a global trend. The linear correlation (R^2), and the calculated results of (ΔG_{dis}^0) , (ΔH_{dis}^0) , (ΔS_{dis}^0) and the relative contribution of enthalpy ζ_H and entropy ζ_T for each (hydrotrope + water) mixture and pure hydrotrope are shown in Tables S4 to S9, and the linear correlations in detail in Figures S2 to S7 in **Appendix G**.

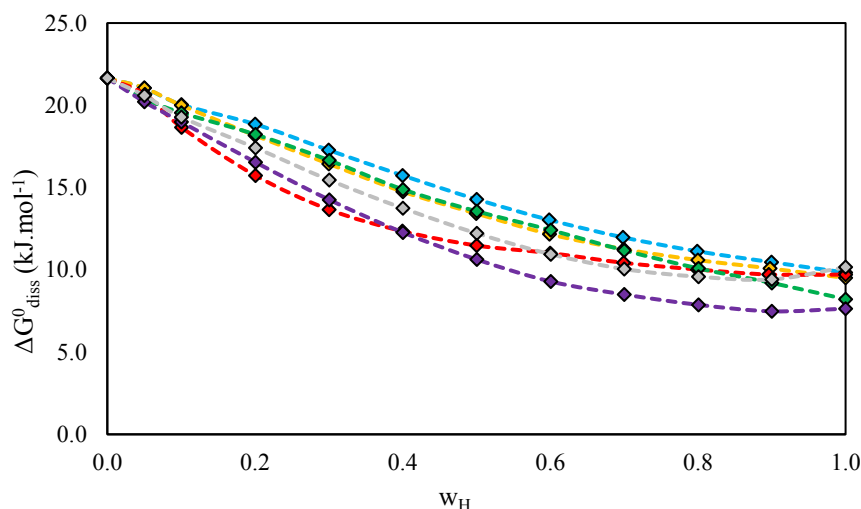
On the other hand, the data obtained for the pure hydrotropes were measured for six temperatures (278.2, 288.2, 303.2, 323.2, 333.2, and 343.2 K) and present a good correlation with acceptable standard deviations, as can be seen in Table 5.3. Still, the (ΔG_{dis}^0) data for both hydrotrope + water mixtures and pure hydrotropes present low standard deviations since the apparent dissolution Gibbs free energy change is directly calculated from solubility data.

Table 5.3. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in cyrene aqueous solutions ($p = 101.3$ kPa).

Solvent	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
water	0.9866	22.04 (0.38)	31.04 (5.03)	29.04 (14.99)	0.78	0.22
1,2-propanediol	0.9897	9.83 (0.19)	17.70 (2.51)	25.40 (7.49)	0.69	0.31
1,3-butanediol	0.9953	9.51 (0.14)	19.18 (1.83)	31.22 (5.45)	0.66	0.34
1,2-pentanediol	0.9848	9.71 (0.24)	18.13 (3.13)	27.19 (9.34)	0.68	0.32
[1.0.0]	0.9948	8.21 (0.11)	13.81 (1.39)	18.07 (4.14)	0.71	0.29
2-ethoxyethanol	0.9816	7.64 (0.14)	9.96 (1.89)	7.50 (5.64)	0.81	0.19
cyrene	0.9816	10.16 (0.24)	16.57 (3.2)	20.69 (9.39)	0.72	0.28

It turns out that both ΔG_{dis}^0 or and ΔH_{dis}^0 of syringic acid dissolution processes in the six selected aqueous hydrotropes solutions and pure solvents are positive, suggesting that the syringic acid dissolution process in all investigated solvent systems is endothermic and non-spontaneous, which is in agreement with the experimental observation that the syringic acid solubility increases with the temperature. Furthermore, positive Gibbs energy changes reflect the low solubilities of syringic acid, and is characteristic of hydrophobic solutes (SOLOMONOV; SEDOV, 2006). As the weight fraction of hydrotropes increases, the values of ΔG_{dis}^0 decrease, while the syringic acid solubility increases, indicating the solute dissolution in the six investigated hydrotropes solutions is inversely proportional to the values of ΔG_{dis}^0 as demonstrated clearly in Figure 5.5.

Figure 5.5. ΔG_{dis}^0 at various mass fractions (w_H) of 1,2-propanediol $\blacklozenge$, 1,3-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, [1.0.0] $\blacklozenge$, 2-ethoxyethanol $\blacklozenge$ and cyrene $\blacklozenge$. Lines are guides for the eyes.



From Figure 5.5 it is also evident that despite the ΔG_{dis}^0 for pure cyrene being similar to the values found for the short alkanediols, the aqueous cyrene solutions present lower ΔG_{dis}^0 values. The opposite happens with [1.0.0], which despite having ΔG_{dis}^0 of the pure compound close to the value of 2-ethoxyethanol, presents values in water close to those of short alkanediols, which agrees with the solubility enhancement achieved by 1,2-propanediol, 1,3-butanediol, and [1.0.0] water mixtures in the same order of magnitude. In fact, 2-ethoxyethanol showed the lowest values of ΔG_{dis}^0 in agreement with the highest syringic acid solubility enhancements achieved by that compound.

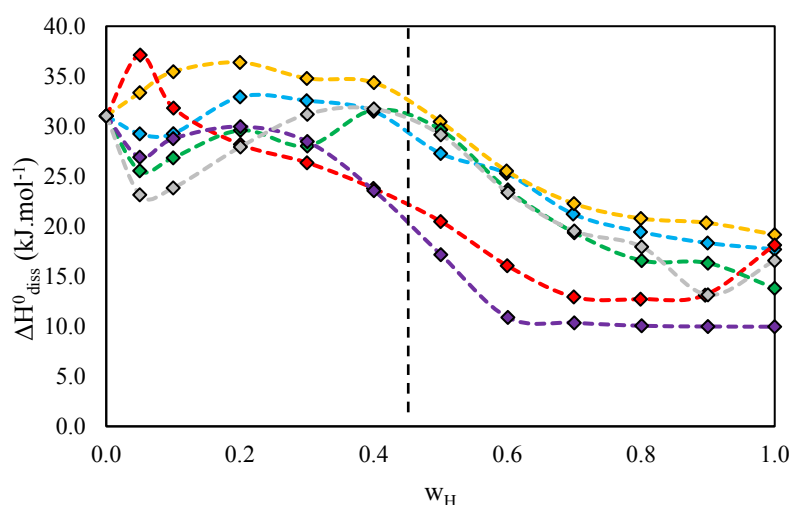
From the apparent thermodynamic properties of solutions data, it is also evident that the molar heat of solution is unfavorable for all solvents at all concentrations since they always present a positive value. The high values of the enthalpy suggest that more energy is required for overcoming the cohesive force of the solute and the hydrotrope in the solution process, which also indicates a strong temperature dependence on solubility (ANIYA *et al.*, 2017; LI *et al.*, 2022a). Naturally, their magnitude indicates about the temperature effect on the solubility, which is of the same level in the pure alkanediols (1,2-propanediol, 1,3-butanediol, and 1,2-pentanediol) and pure cyrene, higher than in pure 2-ethoxyethanol and pure [1.0.0]. Once again, pure 2-ethoxyethanol, which presented the lowest molar heat ($9.96 \text{ kJ}\cdot\text{mol}^{-1}$) achieved higher enhancements in syringic acid solubility. Consequently, water has the highest value of ΔH_{dis}^0 indicating the most unfavorable scenario for dissolution.

Figure 5.6 shows the ΔH_{dis}^0 as a function of mass fraction for all studied hydrotropes. It is possible to see that the values of ΔH_{dis}^0 initially have a minimum at 0.05 mass fraction for 1,2-propanediol, 1.0.0, 2-ethoxyethanol, and cyrene. This minimum is possibly due to the weak interaction between water and hydrotrope in the diluted region. After that, the ΔH_{dis}^0 increases with the increment of the mass fraction. As the hydrotrope concentration increases, the cavity opened to insert the hydrotrope-solute aggregates becomes increasingly larger and requires more energy. The energy associated with the solubilization involves the disruption of the intermolecular interactions of the syringic acid and the solvent molecules within themselves, followed by the establishment of interactions between the three components, which seems to be reflected by the increased ΔH_{dis}^0 values. In this region, the solvation regimen seems to be dominated by water. The exception of this global trend is 1,2-pentanediol, as previous discussed in the **Chapter**

4.2, the mechanism of aggregation is superior to common hydrotropes (with smaller alkyl chains), close to the micelle-like structure.

Then, a transition region is observed (around 0.4 mass fraction) followed by a decrease and stabilization of ΔH_{dis}^0 , at values similar to those of the pure hydrotrope, as shown in Figure 5.6. The stabilization region is a result of hydrotrope-dominated solvation. Here, instead of the water, the solvation is governed by the interaction between the nonpolar part of the hydrotrope and the solute.

Figure 5.6. ΔH_{dis}^0 at various mass fractions (w_H) of 1,2-propanediol $\blacklozenge$, 1,3-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, [1.0.0] $\blacklozenge$, 2-ethoxyethanol $\blacklozenge$ and cyrene $\blacklozenge$. Lines are guides for the eyes.

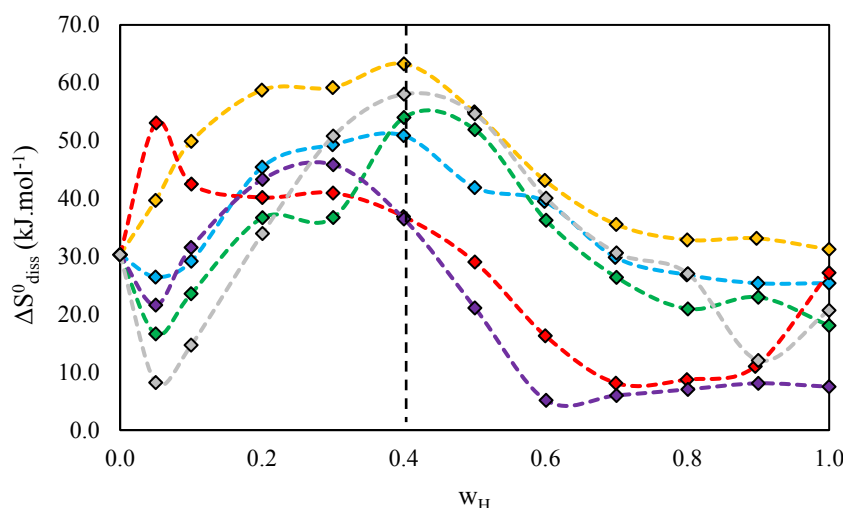


Furthermore, from Figure 5.6, all the ΔH_{dis}^0 values are positive, indicating that the hydrotrope-solute interaction force (by dispersion forces mainly) might be stronger than other interactions (solute-solute and hydrotrope-hydrotrope interaction) in the syringic acid dissolution process by aqueous hydrotropes (ANIYA *et al.*, 2017; SHA *et al.*, 2021; SHAKEEL *et al.*, 2016). The lower values achieved after the maximum corroborates with the two distinguished regions discussed before: in the first region the solute-solute and water-solute interactions drove the dissolution process, mediated by small concentrations of hydrotropes (higher values of ΔH_{dis}^0 were found since more energy was required). After achieving a maximum of ΔH_{dis}^0 the hydrotrope-solute interactions become more pronounced as the hydrotrope-hydrotrope interactions, showing significant differences of S/S_0 at different temperatures and lower values of ΔH_{dis}^0 . Additionally, the minimum value of ζ_H in all investigated hydrotrope aqueous

systems is 63% and the vast majority are higher than 70%, indicating that the energetic factor is the most important contributor to ΔG_{dis}^0 during the syringic acid dissolution process.

A similar trend observed for the ΔH_{dis}^0 is shown by ΔS_{dis}^0 : a minimum were observed at 0.05 mass fractions, and the entropy is enhanced in the non-concentrated hydrotrope region until the transition region, where ΔS_{dis}^0 starts to decrease, as shown in Figure 5.7.

Figure 5.7. ΔS_{dis}^0 at various weight fractions of 1,2-propanediol $\blacklozenge$, 1,3-butanediol $\blacklozenge$, 1,2-pentanediol $\blacklozenge$, [1.0.0] $\blacklozenge$, 2-ethoxyethanol $\blacklozenge$ and cyrene $\blacklozenge$. Lines are guides for the eyes.



For all hydrotropes the solution entropy is positive, favoring the dissolution process (PINHO; FERREIRA, 2012). Despite that, in the concentrated-hydrotrope region, the low values found for 2-ethoxyethanol and 1,2-pentanediol hinder the dissolution process possibly due to a more considerable ordering of the hydrotrope molecules to form hydrotrope-solute and hydrotrope-hydrotrope clusters. However, in this region the entropy contribution to the ΔG_{dis}^0 is very small, values lower than 20% of ζ_{TS} were found for 2-ethoxyethanol and up to 32% for 1,2-pentanediol. For the last the entropy seems to have more impact, somewhat disfavoring the syringic acid dissolution process despite ΔH_{dis}^0 favored it in a higher percentage. In other words, in the non-concentrated hydrotrope region, the highest values of ΔH_{dis}^0 disfavored the syringic acid dissolution that is in a less favored by ΔS_{dis}^0 . On the other hand, in the concentrated-hydrotrope region, while the enthalpy contributes to the dissolution process, entropy (in

a lower percentage) disfavors the solubilization. In general, the addition of hydrotropes after 0.4 in mass fraction will improve the syringic acid solubility to a certain extent.

5.1.5. Conclusions and perspectives

In this work, the effect of temperature on the solubility of syringic acid in non-ionic hydrotropes was studied and revealed two different mechanisms of action of hydrotropes: (i) in the low-concentration hydrotrope region, the water as continuous media drives the dissolution process and the solubility enhancements do not appear to be very significant with the temperature increment while (ii) in the concentrated-hydrotrope region, the hydrotrope-solute interactions govern the dissolution and the solubility enhancement appears to be more pronounced.

Additionally, the solution properties (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) were calculated using the van't Hoff equation and showed that the dissolution of syringic acid is synergistically affected by enthalpy and entropy, despite the energetic factor being the most important for the solubility changes. Although the data obtained for (ΔH_{dis}^0) and (ΔS_{dis}^0) show high variability (significant standard deviations) they reveal an interesting global trend. For (ΔH_{dis}^0) and (ΔS_{dis}^0) the two different regions of the solubility mechanism were also observed, while (i) is favored by the balance between enthalpy and entropy, (ii) is promoted by enthalpy and disadvantaged by entropy. Solubility measurements for at least two more temperatures within the studied range (278.2 to 343.2 K) would be necessary to reduce the errors associated with determinations derived from the van't Hoff equation.

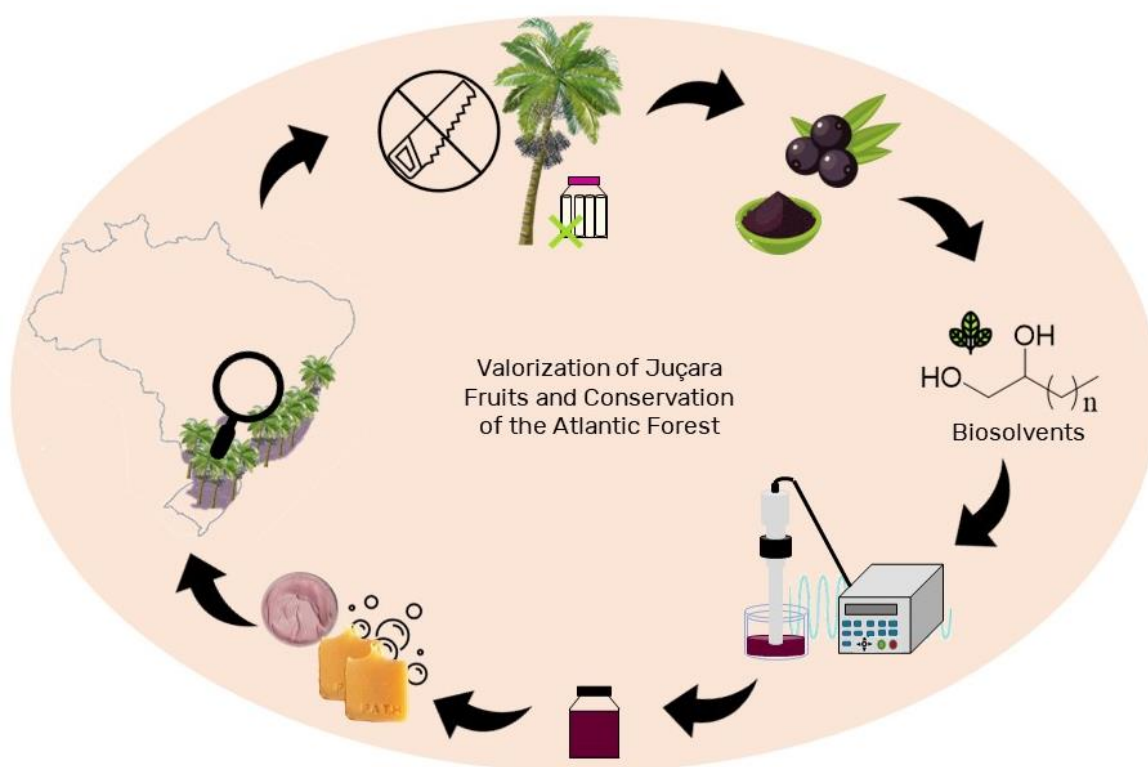
To delve further into the study of the mechanisms of self-aggregation of hydrotrope molecules in the concentrated region, the modeling data by thermodynamic models, COSMO-RS predictions, and other complementary experimental measurements (such as the study of the hydrotropes solutions using chemical environment probes) could provide further insights on the solvation mechanisms. Despite that, the data obtained in this work is a suitable guidance to improve the solubility of hydrophobic solutes, such as syringic acid, by non-ionic hydrotropes, taking into consideration both temperature and hydrotrope concentration.

6. CHAPTER VI - APPLICATION OF HYDROTROPES TO NATURAL MATRICES

6.1. JUÇARA FRUIT (EUTERPE EDULIS MARTIUS) VALORIZATION COMBINING EMERGENT EXTRACTION TECHNOLOGIES AND AQUEOUS SOLUTIONS OF ALKANEDIOLS

Chapter based on the published manuscript:

Bruna P. Soares, Ana M. Ferreira, Marina Justi, Luiz Gustavo Gonçalves Rodrigues, José Vladimir Oliveira, Simão P. Pinho and João A. P. Coutinho, Juçara Fruit (*Euterpe edulis* Martius) Valorization Combining Emergent Extraction Technologies and Aqueous Solutions of Alkanediols, *Molecules* v. 28 (4), p. 1607, 2023.



Synopsis: valorization of juçara fruits through alkanediols aqueous solutions and emergent extraction methodologies followed by cosmetic formulations.

6.1.1. Video abstract

The video abstract of this work can be accessed at:
<https://youtu.be/nB8NSkqyknA>

6.1.2. Abstract

Anthocyanins from juçara fruits were extracted by pressurized liquid extraction (PLE) or ultrasound-assisted extraction (UAE), using aqueous solutions of 1,2-alkanediols and glycerol ethers as biobased solvents. The PLE (100 bar, 13 min, 1 mL/min flow rate) in the optimal extraction conditions (55.2 wt% of 1,2-propanediol, 387.2 K, and pH 7.0), originated 23.1 mg_{anthocyanins.g_{dry biomass}⁻¹}. On other hand, the UAE was 10 min long, and the optimal conditions using 1,2-propanediol were 42.6 wt%, 160 W, and pH 7.0, leading to 50 mg_{anthocyanins.g_{dry biomass}⁻¹}. Extractions at the UAE optimized conditions, with aqueous solution of five different 1,2-alkanediols, and three of glycerol ethers were performed, and compared to water and ethanolic extracts. The biobased solvents solutions presented anthocyanin yields up to 33% higher than water, and showed to be as efficient as ethanol/water, but generating extracts with higher antioxidant capacity. The anthocyanin-rich extract of juçara, obtained under optimized conditions with 1,2-propanediol, was used in the production of a natural soap and incorporated into a cream, showing that the addition of the juçara extract resulted in an antioxidant capacity in both products.

keywords: pressurized liquid extraction; ultrasound-assisted extraction; 1,2-alkanediols; glycerol ethers; anthocyanins; product formulation.

6.1.3. Introduction

Juçara (*Euterpe edulis* Martius) is a tropical palm tree of the Atlantic Forest that produces a spherical purple fruit popularly known as juçara, a derived product of palm-heart extraction from the palm tree (SCHULZ *et al.*, 2016; TREVISAN *et al.*, 2015). The valorization of the juçara fruit contributes to environmental sustainability, and the recovery and conservation of the Atlantic Forest biodiversity, since for the harvest of the fruits, it is not necessary to cut the tree, as happens in the extraction of the palm heart, thus threatening the palm tree with extinction (CARVALHO *et al.*, 2017; GAIOTTO; GRATTAPAGLIA; VENCovsky, 2003; SCHULZ *et al.*, 2016). The valorization of juçara fruits generates an income source for local communities, employing collectors and providing a work base for handicrafts (leaves, seeds, rachillas, pigments, etc.), food products (juice, frozen pulp), cosmetics, and even medicinal products (GARCIA *et al.*, 2019; TREVISAN *et al.*, 2015).

Juçara fruit is considered a “superfruit” due to its high nutritional value, rich in phenolic compounds such as anthocyanins and phenolic acids (CARDOSO *et al.*, 2018). With a high concentration of the anthocyanins cyanidin-3-O-rutinoside and cyanidin-3-O-glucoside, these are the main constituents of the phenolic juçara fruits’ profile (around 73% of the total phenolic compounds content) (VIEIRA *et al.*, 2017). Anthocyanins are associated with juçara’s intense dark purple color, and one of their main advantages is their antioxidant properties, making them valuable compounds to the food and pharmaceutical industries (BICUDO; RIBANI; BETA, 2014; CARDOSO *et al.*, 2018; VIEIRA *et al.*, 2017). Regarding the phenolic acid profile, hydroxybenzoic (gallic, protocatechuic), p-hydroxybenzoic (vanillic, syringic), and hydroxycinnamic (chlorogenic, caffeic, p-coumaric, sinapinic, and ferulic) are some of the main phenolic acid constituents (BICUDO; RIBANI; BETA, 2014; CARDOSO *et al.*, 2018). However, phenolic compounds, such as anthocyanins, are easily degradable, and bioconverted in phenolic acids by temperature, light, solvents, and pH changes, affecting their bioavailability (BRAGA *et al.*, 2018a, 2018b; PERON; FRAGA; ANTELO, 2017). Aiming to overcome these limitations, the use of innovative and environmentally-friendly extraction technologies (reducing time, energy, and solvent consumption), such as pressurized liquid extraction (PLE) and ultrasound-assisted extraction (UAE), have been applied. These technologies have been explored in a wide range of valuable compounds found in low-cost natural products (TURNER, 2006; VINATORU, 2001; VINATORU; MASON; CALINESCU, 2017; WEN *et al.*, 2020).

PLE employs solvents at high pressure and temperatures. The potential of PLE is correlated to (i) facilitate the disruption of analyte-matrix by exerting pressure, (ii) increases the solubility, and (iii) improve the mass transfer by the use of liquid solvents at temperatures above their atmospheric boiling point (MUSTAFA; TURNER, 2011; TURNER, 2006). On the other hand, UAE is a low-pressure green extraction technology that involves the use of ultrasounds, which mechanically increase the contact between solvents and natural matrices, allowing the disruption of the plant cell wall in the case of plants matrices, decreasing the mass transfer resistance (SHIRSATH; SONAWANE; GOGATE, 2012; WEN *et al.*, 2020). For UAE, the cavitation generated by liquid bubbles collapse is an important driving force in the extraction process, since the cavitation results in extremely high-speed microjets of solvent that reach the vegetal matrix, facilitating the

penetration of the solvent for swelling and solvation process (VINATORU, 2001; VINATORU; MASON; CALINESCU, 2017).

The choice of solvent and temperature is crucial for both extraction methods. Differences in temperature and solvent composition change the solvents dielectric constant (ϵ_0), and thus their polarity (CHEMAT *et al.*, 2019). High temperatures destroy the water hydrogen-bond structures, reducing its polarity and allowing a better dissolution of polar and non-polar substances (CHEMAT *et al.*, 2019; MUSTAFA; TURNER, 2011). Organic solvents such as ethanol and methanol, commonly used extraction solvents, and present lower dielectric constants than water (MOHSEN-NIA; AMIRI; JAZI, 2010). In this perspective, it is possible to combine the polarity of organic solvents and water to generate environmentally aqueous solutions with the desired polarity by changing the concentration of the solvent and the applied temperature. Amphiphilic compounds, such as alkanediols and glycerol ethers, can offer this desired polarity tunability and can be a worth opportunity for enhancing the performance and extending the applicability of these emerging extraction methods (HENRY; YONKER, 2006; HERRERO *et al.*, 2015; PENA-PEREIRA; NAMIEŚNIK, 2014).

Various compounds have been proposed as solvent additives for subcritical water extraction (MACHMUDAH *et al.*, 2018; TOMMASI *et al.*, 2017), including aqueous solutions of DES for the extraction of anthocyanins from jaboticaba peels using pressurized liquid extraction (BENVENUTTI; ZIELINSKI; FERREIRA, 2022). Some works propose the use of alkanediols as DES precursors for the extraction and recovery of bioactive compounds from natural sources (RUESGAS-RAMÓN; FIGUEROA-ESPINOZA; DURAND, 2017; WAN MAHMOOD *et al.*, 2019; WANG *et al.*, 2019; WOJEICCHOWSKI *et al.*, 2021). Recently, the use of biobased solvents in PLE applications has been expanded. There are reports of studies on the use of limonene (GOLMAKANI *et al.*, 2014), glycerol (HUAMÁN-CASTILLA *et al.*, 2020), and other renewable biosolvents (BERMEJO *et al.*, 2013; HU *et al.*, 2011). However, the applications of biobased solvents in the extraction of bioactive compounds combined with alternative extractions have been seldom addressed.

Biobased solvents are produced from renewable resources (BEHR *et al.*, 2008; GARCÍA; GARCÍA-MARÍN; PIRES, 2014). One of the most studied biobased solvents is the 1,2-propanediol, also known as propyleneglycol, which can be produced from de-oxy sugars using microorganisms for its fermentation and through hydrogenolysis of

glycerol, a by-product of biodiesel production (BEHR *et al.*, 2008; GARCÍA; GARCÍA-MARÍN; PIRES, 2014; SAXENA *et al.*, 2010). The 1,2-propanediol is considered a GRAS solvent (Generally Recognized as Safe) for food, cosmetics, and drug applications (FDA, 2018; GÜNTHER VERHEUGEN, 2006). Its use includes the production of unsaturated polyester resins, as an additive in nutrition products, coloring, and flavoring agent, and also as a component of lubricants and anti-freezing agent in cosmetics (BEHR *et al.*, 2008). Although alkanediols, such as 1,2-propanediol, are industrial biosolvents and have been adopted for many applications, they have been poorly explored as extractive solvents so far. Recently, Vieira *et al.* (VIEIRA *et al.*, 2020), studied glycerol and a series of alkanediols (1,2-ethanediol, 1,2-propanediol, 1,3-propanediol, 1,3-butanediol, 1,2-pentanediol, 1,5-pentanediol, and 1,2-hexanediol) for the extraction of bioactive compounds from *Juglans regia* L. leaves, by the heat-assisted method. In that work, the alkanediols 1,2 and 1,3-propanediol and 1,3-butanediol showed the best results (high phenolic concentration and cytotoxicity potential against cervical carcinoma cells). To the best of our knowledge, the extraction techniques reported in the literature using alkenodiols and glycerol were all carried out at low-pressure and mostly applying high concentrations of the alkanediols, from 25 to 100% (v/v). There are very few works in which alkanediols are applied in UAE (BARANAUSKAITE *et al.*, 2016; SUNGPUD *et al.*, 2020).

This work aims to extract anthocyanins from juçara fruit using aqueous solutions of the 1,2-alkanediols and to apply alternative extraction technologies, such as PLE and UAE. First, a kinetic study using 1,2-propanediol aqueous solutions was carried for PLE and UAE. Then, experimental conditions for each extraction technology, namely, temperature/amplitude, solvent concentration and pH, were optimized by response surface methodology (RSM). Since the preliminary results showed the UAE technique to give the highest extraction yields of anthocyanins (in a shorter time and with a lower solid-to-liquid ratio compared to PLE), the screening of several aqueous solutions of 1,2-alkanediols (1,2-ethanediol, 1,2-propanediol, 1,2-butanediol, 1,2-pentanediol, and 1,2-hexanediol) and glycerol ethers [1.0.1], [2.0.2] and [2.0.0], at the optimized conditions by UAE, was conducted concerning extraction yield of anthocyanins and extracts antioxidant activity. Finally, to demonstrate the potential of the anthocyanin-rich extract in cosmetic products, the extract obtained by UAE using 1,2-propanediol under the

optimized conditions was directly used as additive to produce natural soap and incorporated in a cream formulation.

6.1.4. Material and methods

6.1.3.1. Raw Material

The juçara fruit (*Euterpe edulis* Martius) pulps were produced and kindly supplied by Duas Rodas company (Jaraguá do Sul (SC) – Brazil), harvested in June 2020, in Garuva, Jaraguá do Sul and Joinville (SC) – Brazil. The pulps were pasteurized, providing a shelf life of 36 months under refrigerated conditions 255.2 K). The juçara pulp samples were frozen in 5.0 kg packages at the Laboratory of Thermodynamics and Supercritical Technology (LATESC, UFSC, Brazil). Frozen portions of the sample were freeze-dried (Liotop, model LD101, São Paulo, Brazil) for 48 h in order to reduce the moisture content from approximately 96% to 4-6%, determined according to 012 IV method (INSTITUTO ADOLFO LUTZ, 2008). Dried samples were ground with a knife mill (average particle diameter = 0.345 mm) and stored in polyethylene bags at 255.2 K until further use.

6.1.3.2. Chemicals

The chemicals used in this work are detailed in Table 6.1, along with their CAS, sources and mass purities. Water was double distilled, passed across a reverse osmosis system, and further treated with a Milli-Q plus 185 water purification apparatus. The glycerol ethers used were synthesized as described in detail in a previous work (SOARES *et al.*, 2020). All oils, butter, essential oils and sodium lactate for soap production were purchased from Jabonarium. The Versatile™ base cream was purchased from Fagron Iberica S.A.U. (Barcelona, Spain). Bio-propylene glycol from Orlen was kindly supplied by Brenntag Portugal.

Table 6.1. List of compounds used in this work along with their CAS number, source, and purity.

Compound	CAS number	Source	Purity (wt%)
1,2-ethanediol	107-21-1	Fisher Scientific	>99.0
1,2-propanediol	57-55-6	Sigma-Aldrich	99.5
1,2-butanediol	584-03-2	Sigma-Aldrich	>98.0
1,2-pentanediol	5343-92-0	TCI	97.0
1,2-hexanediol	6920-22-5	Alfa Aesar	97.0
1,3-dimethoxypropan-2-ol [1.0.1]	623-69-8	Synthesized	>99.0
1,3-diethoxypropan-2-ol [2.0.2]	4043-59-8	Synthesized	>99.0
3-Ethoxypropane-1,2-diol [2.0.0]	1874-62-0	Synthesized	>99.0
2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS)	30931-67-0	Sigma-Aldrich	98.0
Acetone	67-64-1	Fisher Scientific	>99.0
Acetonitrile	75-05-8	Fisher Scientific	99.9
Cyanidin 3-O-glucoside chloride	7084-24-4	Sigma-Aldrich	>95.0
Cyanidin-3-O-rutinoside chloride	18719-76-1	Sigma-Aldrich	>98.0
Ethanol	64-17-5	Fisher Scientific	99.8
Folin-Ciocalteu reagent 2M	-	Panreac	-
Gallic acid	149-91-7	Sigma-Aldrich	99.5
Hydrochloric acid (HCl)	7647-01-0	Fisher Scientific	37.0
Methanol	67-56-1	Fisher Scientific	99.8
Potassium persulfate (K ₂ S ₂ O ₈)	7727-21-1	Scharlau	99.0
Sodium carbonate (Na ₂ CO ₃)	497-19-8	Prolabo	99.0
Sodium hydroxide (NaOH)	1310-73-2	Fisher Scientific	98.0
Sulfuric acid (H ₂ SO ₄)	7664-93-9	Sigma-Aldrich	95.0
Trolox	53188-07-1	Acros Organics	97.0

6.1.3.3. Extraction Methodologies

The obtention of the anthocyanin-rich extracts from juçara dried pulps was performed by combining the use of aqueous solutions of alkanediols with emergent extraction techniques, namely pressurized liquid extraction (PLE) and Ultrasound-Assisted Extraction (UAE).

6.1.3.4. Pressurized Liquid Extraction

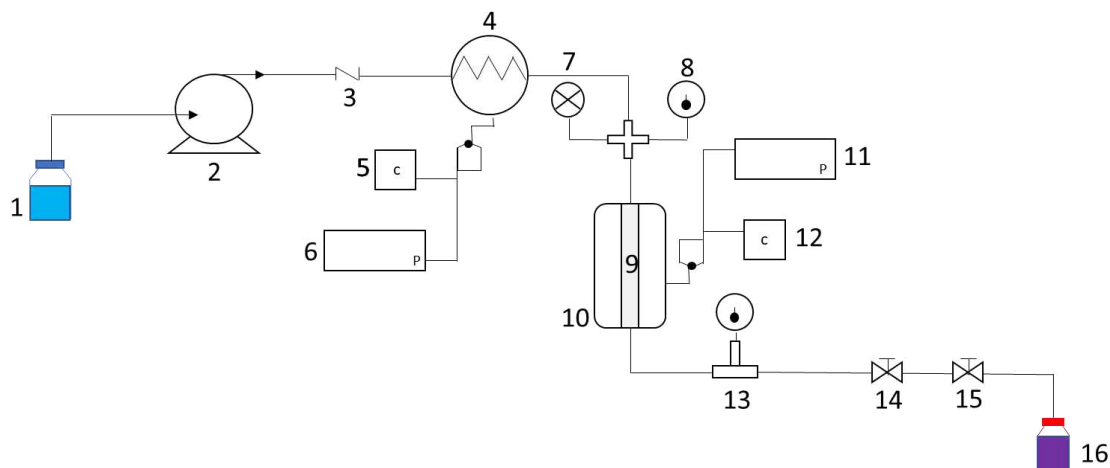
A self-assembled PLE unit was used to recover anthocyanins from juçara dried pulps. The customized apparatus was built to perform subcritical water extractions (PLAZA; TURNER, 2015). The apparatus contains an extraction vessel of 17.3 mL

(internal diameter of 9.40 mm and a height of 250 mm) made of 316/316L stainless steel. The vessel was heated using an electrical heating jacket system, using 250 W resistances (designed to withstand up to 573.2 K). A pre-heater system (with two resistances of 250 W with steel coil), brings the solvent up to the operating temperature before entering the extraction vessel.

The temperature of the extraction vessel and the pre-heater was controlled by a digital temperature controller (PXF4 Fuji electric, Tokyo, Japan). The thermocouple (model ADAM 4019+-AE, Advantech, California, USA) allowed for the acquisition of temperature data using a type-k temperature sensor (model BT.MiK1.2.1,5/3.15/35/50, Bresimar Automação, Aveiro, Portugal). The extraction pressure was monitored by a digital manometer (model PTI-S-AG250-15AV, Swagelok, London, United Kingdom). The solvent was pumped by a compact HPLC pump (PU-4580 Jasco, Maryland, USA) into the extraction vessel containing the biomass, where the pressure was controlled by a needle valve (model SS-41GS2, Swagelok, USA).

A schematic diagram of the PLE customized apparatus is illustrated in Figure 6.1, containing the main constituents of a supplying pump, a pre-heating and a temperature-controlled system, an extraction vessel and a pressure-controlled valve.

Figure 6.1. Schematic design of the customized PLE unit: 1) Solvent reservoir; 2) Solvent pump; 3) Check Valve; 4) Pre-heater exchanger; 5 and 12) Automatic temperature controller; 6 and 11) Data acquisition by computer; 7) Manometer; 8) Thermocouple; 9 and 10) Extraction vessel with electrical heating jacket; 13) Regulator needle valve; 14) Regulator valve; 15) Block valve and 16) Amber glass flask.



The runs were made at continuous mode, with pressure fixed at 10 MPa, a flow rate of 1 mL/min, and a time of 13 min, which was defined through an extraction kinetic (performed with 1 g of sample, at 353.2 K, 30 wt% of 1,2-propanediol solution as the solvent and pH of 4.5) based in the yield of extraction of anthocyanins ($\text{mg}_{\text{anthocyanins}} \cdot \text{mL}^{-1}$). A pressure of 10 MPa was chosen to perform the high-pressure extractions because it has been reported in the literature that a pressure higher than 10 MPa has a negligible effect on the solubilization and recovery of the target compounds (MUSTAFA; TURNER, 2011; PLAZA; TURNER, 2015). One of the explanations for this is that the change in density with pressure is generally insignificant due to the non-compressibility of the liquid (LI *et al.*, 2022b). In addition, the use of higher-pressure conditions are directly associated with higher energy requirements.

After the extractions, the samples were immediately collected in amber flasks, diluted, and quantified by HPLC-DAD on the same day.

6.1.3.5. Ultrasound-Assisted Extraction (UAE)

UAE was carried out according to the method described by Cheok *et al.* (CHEOK *et al.*, 2013). The assays were performed using an UAE equipment (Branson 450 Digital Sonifier), where a titanium probe was immersed right below the sample, containing 0.2 g of juçara dried pulps, added to 10 mL of solvent (1,2-propanediol/water mixture) in a sonication time of 10 min, defined from preliminary tests (performed for 30

wt % of 1,2-propanediol at pH 7.0 and 30% of amplitude). After extraction, the samples were centrifuged for 10 min at room temperature at 10,000 rpm in an Eppendorf centrifuge 5425, collected in amber flasks, diluted, and quantified by HPLC-DAD on the same day. It is important to mention that amplitude affects the amount of power applied during the experiment, where 100% of amplitude in this equipment corresponds to 400W.

6.1.3.6. Response Surface Methodology

The central composite rotatable design (CCRD) is a response surface methodology, in which a multivariate statistical tool is here applied to optimize the extraction conditions of anthocyanins from juçara pulps. The relationship between the response and the independent variables was described according to the following polynomial equation:

$$y = \beta_0 + \sum_{i=1}^k \beta_i X_i + \sum_{i=1}^k \beta_{ii} X_i^2 + \sum_{i < j}^k \beta_{ij} X_i X_j \quad (6.1)$$

where β_0 represents the intercept or regression coefficient, β_i , β_{ii} , and β_{ij} are the linear, quadratic, and interaction coefficients, respectively, X_i and X_j are the independent variables, and k is the number of variables studied that can influence the response y . In this work, the independent variables were subjected to factorial planning of 2^3 (3 variables, and 2 levels) to optimize the yield of extraction of anthocyanins ($\text{mg}_{\text{anthocyanins}}.\text{mL}^{-1}$) as the measured response function. Nineteen experiments were performed, including five replicates at the central point. A 90% confidence level was chosen to analyze the results. To verify the significance of the model parameters analysis of variance (ANOVA) was implemented. The coefficient of determination (R^2) and adequate precision (F calculated value and p-value) were used to evaluate the adequacy of the polynomial equation for the response and optimum values. The experimental design, statistical analysis, and regression model were accomplished using Statistica version 13.5.0.17.

In this work, 2 factorial planning were executed, for both techniques of extraction, PLE and UAE. The first factorial planning was applied for PLE, in which the chosen levels of the independent variables were temperature (333.2, 353.2, and 373.2 K), 1,2-propanediol concentration in water (15, 30, and 45 wt%), and pH (3.0, 4.5, and 6.0). The second factorial planning was carried out for UAE, in witch amplitude (18, 30, and

42 %), 1,2-propanediol concentration in water (15, 30, and 45 wt%), and pH (4.0, 7.0, and 10.0) were varied. All coded levels of independent variables used in the factorial planning for the optimization of operating conditions for both PLE and UAE will be given in detail in Sections 6.1.4.1 and 6.1.4.2, respectively. For both extractions methodologies the pH of the solutions was adjusted from 0.1 M solutions of HCl or NaOH.

6.1.3.7. Anthocyanin-Rich Extracts Characterization

The characterization of both anthocyanins, cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside, was carried out by HPLC-DAD (Shimadzu, model Prominence, Kyoto, Japan). Chromatographic analyses were performed with an analytical C18 reversed-phase column (250×4.60 mm), Kinetex 5 μ m C18 100 Å, from Phenomenex. The separation was conducted in a gradient system of pure methanol (phase A) and 0.1% of sulfuric-acid-ultra-pure water (phase B). The separation was carried out using the following gradient mode: 0 min 5% of phase A; 1 min 13% of phase A; 2 min 15% of phase A; 3 min 18% of phase A; 3.5 min 20% of phase A; 4 min 25% of phase A; 4.5 min 27% of phase A; 6.5 min 50% of phase A; 8.5 min 5% of phase A; 20 min 5% of phase A. The flow rate was 1 mL/min, with a volume injection of 20 μ L, and DAD was set at 520 nm. The column oven was operated at a controlled temperature of 328.2 K. Each sample was analyzed at least in duplicate. Calibration curves were prepared using the pure anthocyanins with samples dissolved in 0.1% sulfuric-acid-ultra-pure water. Cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside displayed a retention time of 8.0 and 8.7 min, respectively. The results were expressed in mg of anthocyanin per g of dry biomass ($\text{mg} \cdot \text{g}_{\text{dry biomass}}^{-1}$). Henceforth, the sum of both anthocyanins concentrations expresses the yield of extraction in mg of anthocyanins per g of dry biomass ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$).

6.1.3.8. Comparing Alkanediol Aqueous Solutions with Other Solvents

To compare different solvents, the UAE technique was chosen given (i) the higher extraction yield of anthocyanin achieved in the optimization compared to PLE; (ii) the shortest extraction time (10 minutes for UAE and 13 minutes for PLE) and maintenance between two consecutive experiments (around 5 minutes for UAE and 1 hour for PLE) and (iii) the lower solid-liquid ratio required (1:50 g/mL for UAE versus 1:13 g/mL for PLE). Here, the effect of the type of solvent on UAE yield and antioxidant

activity was evaluated using the optimized conditions. Aqueous solutions of the 1,2-alkanediol series (1,2-ethanediol, 1,2-propanediol, 1,2-butanediol, 1,2-pentanediol and 1,2-hexanediol) and the glycerol ethers [1.0.1], [2.0.2] and [2.0.0] were selected in the following of previous studies using these compounds (ABRANCHES *et al.*, 2022; SOARES *et al.*, 2020) and compared with water and ethanol used as control solvents. The chemical structure of the solvents mentioned above is represented in Figure S1 in the **Appendix H**. Besides the anthocyanins yield, defined by HPLC-DAD quantification, the following analyses were also conducted to evaluate the total phenolic content and antioxidant activity of the extracts recovered using different solvents:

6.1.3.9. Total Phenolic Content (TPC)

The TPC of juçara pulps extracts obtained for different solvents (1,2-alkanediol series, glycerol ethers, water, and ethanol) was determined by the Folin-Ciocalteu method, according to Koşar *et al.* (KOŞAR; DORMAN; HILTUNEN, 2005), with a few modifications. The extracts obtained were initially diluted 10 times in the correspondent extraction solvent. Then, 10 µL of the sample solution, 600 µL of distilled water, and 50 µL of Folin–Ciocalteu reagent were mixed. After 1 min, 150 µL of Na₂CO₃ 20% (w/v) were added, and the volume was completed to 1 mL with distilled water. Aliquots of 300 µL of each sample are transferred to wells in a microplate. After 2 h, at 298.2 K in the absence of light, the absorbance was measured at 760 nm in a UV-spectroscopy using a BioTeck Synergy 4 HT microplate reader. The analyses are realized in quintuplicate and the TPC was calculated according to a standard curve prepared with gallic acid. The results are expressed in mg of Gallic Acid Equivalent per g of dry biomass (mg_{GAE}/g_{dry biomass}).

6.1.3.10. ABTS Radical Scavenging Assay

The mechanism involved in ABTS⁺ radical (2,2'-Azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt) cation quenching follows an electron transfer mechanism. The ABTS assay of the extracts obtained for different solvents (1,2-alkanediol series, glycerol ethers, water, and ethanol) was realized according to Re *et al.* (RE *et al.*, 1999), with some modifications. First, 7 mM ABTS solution and 2.45 mM potassium persulfate solution was allowed to react (1:1) for 16 h at room temperature to generate the radical ABTS⁺. Then, ABTS⁺ solution was diluted

with distilled water until an absorbance of 0.700 (± 0.05) at 734 nm. The extracts were diluted 10 to 100 times in the corresponding extraction solvent. After that, 20 μL of each sample solution and 280 μL of ABTS^+ were added to a microplate. Finally, after 30 min of incubation in darkness, the absorbance was read at 734 nm. The analysis was performed in quintuplicate accompanied by a control (20 μL of diluted solvent, 280 μL of ABTS^+) and the blank for each sample (20 μL of sample, 280 μL of distilled water). Trolox was used as a reference standard, and the concentrations were calculated from a standard curve. The results are expressed in terms of μmol of Trolox equivalent per g of dry biomass ($\mu\text{mol}_{\text{TE}}/\text{g}_{\text{dry biomass}}$) for quintuplicate measurements.

6.1.3.11. Statistical Analysis

The analysis of variance (ANOVA), followed by the Tukey test, were applied using ORIGIN 2018 to compare the significance of the extraction yields, TPC, and antioxidant activity obtained for the two anthocyanins extracted with the different solvents studied (1,2-alkanediol series, glycerol ethers, water, and ethanol), using a significance level of 90% ($p < 0.1$).

6.1.3.12. Cosmetic Formulations using Juçara Anthocyanins-rich Extract as Additive

6.1.3.12.1. Natural Soap Production by Hot Process Saponification

The natural soaps were manufactured using 5% of juçara extract and 30% of distilled water (% over the total mass of oils). The soaps were prepared by hot saponification or hot processing method. Sodium hydroxide (NaOH), or lye, was used as the base. A lye calculator was used to determine the rate of saponification for each oil used to formulate the soap, and it indicated that 15% of NaOH (over the total mass of oils) was required. A mixture of oils, including coconut oil (30%), sunflower oil (25%), castor oil (10%), and karite butter (35%), was heated at 373.2 K, then, without heating sodium lactate (2%) was added and mixed. Immediately, the NaOH aqueous solution was prepared and promptly added to the oil mixture and properly blended to mix the ingredients homogeneously. After the texture changed, the additives were incorporated into the soap: a mixture of essential oils (5%) and juçara extract (5%). Parchment paper-lined molds were filled with the soap mixture and saponified for 24 h at room temperature.

After saponification, each soap loaf was removed from the mold, weighed (approximately 0.75 kg), and dimensions were measured ($0.045 \times 0.05 \times 0.225$ m). The loaves were cut into nine identical bars using a commercial wire cutter. Each bar had a similar shape and weight, measuring ($0.045 \times 0.05 \times 0.025$ m), and weighing approximately 0.08 kg. The soap bars were placed in small wooden containers and stored at room temperature in a dark room with controlled humidity for one week. After curing, the soap shavings were wrapped with aluminium foil paper and kept at 193.2 K for further analysis. The abbreviations used to delineate the natural soaps evaluated in this study are: BB = base bar and JB = juçara bar. For the base, the same recipe was used but without the juçara extract, which was replaced by rosemary oil, a conventional antioxidant for natural herbal soaps.

6.1.3.12.2. Incorporation of the Extract in a Cosmetic Cream

Juçara extract, rich in anthocyanins, was incorporated into the cosmetic cream Versatile™ enriched oil in water (O/W). The cosmetic cream used as a cream base contains natural lipids with moisturizing and protective properties compatible with the incorporation of a wide range of active ingredients. According to the manufacturer, this base cream contains purified water, natural oil, O/W emulsifiers, emollients, lubricants, pro-liposome, vitamin E, chelating agent, silicone, and preservatives, and is free of fragrances, dyes, parabens, mineral oil, SLS, propylene glycol and 1,4-dioxane. In the preparation of the formulations, 50 mg of the extract was mixed into 1 g of base cream. Three different cream formulations were evaluated: CC = control cream (without the addition of extract); JC = juçara cream (with direct addition of the final UAE extract); and CJC = concentrated juçara cream (juçara extract was concentrated twice by reducing the water content).

6.1.3.12.3. Antioxidant Activity Analysis and pH of Cosmetic Formulations

To perform the analyses of antioxidant activity of the studied cosmetic products, 100 mg of each soap or 200 mg of each cream were extracted with 1 ml of acetone:ethanol (1:1 v/v) for one hour in a Tryster device (Multi Bio RS -24, Biosan) with an orbital rotation of 50 rpm. The sample was centrifuged, at 10,000 rpm for 15 min, and the supernatant was carefully decanted without disturbing the pellet. The supernatant was filtered using syringe filters, and the filtrate was used directly to determine the antioxidant

activity of the formulations. The antioxidant activity for the formulations was determined by the ABTS method (section 6.1.3.5.2), and the results expressed in milligrams of trolox equivalent per gram of formulation ($\text{mg}_{\text{TE}}/\text{g}_{\text{formulation}}$). In the case of the soap/cream quantifications, the ABTS^+ solution was diluted with ethanol instead of distilled water, due to the high level of fat ingredients present in these formulations.

For measured the pH 1 g of the final formulation (soap or cream) was diluted in 10 mL of distilled water and the resulting solution was measured using a pH meter calibrated before each measurement (SevenExcellence, Mettler Toledo).

6.1.5. Results and discussion

6.1.4.1. Optimization of PLE Extraction Conditions

The extraction kinetics was firstly established at 10 MPa, 353.2 K, 1 mL/min, 30 wt% of 1,2-propanediol concentration and pH 4.5 to enable the definition of the extraction time, and then, this time was applied for all assays and solvents studied. The overall extraction curve (OEC) representing the change of the yield of extraction ($\text{mg}_{\text{anthocyanins}}.\text{mL}^{-1}$) with the extraction time is presented in Figure S2 (**Appendix H**).

The analysis of the kinetics behavior for PLE from juçara extracts showed the following mass-transfer mechanisms: constant extraction rate period (CER), falling extraction rate period (FER), and diffusion-controlled period (DC). In addition to that, the extraction time was fixed at 13 min for all assays, because it represents the beginning of the DC period where most of the soluble anthocyanins were already extracted (after 13 min, the yield increment is less than 3%).

All PLE assays were performed according to CCRD plan and considering the fixed conditions of pressure, 10 MPa, flow rate 1mL/min, and time, 13 min, for the recovery of extract samples rich in anthocyanins. The extracts were analyzed according to the yield of extraction ($\text{mg}_{\text{anthocyanins}}.\text{g}_{\text{dry biomass}}^{-1}$), and the results are presented in Table 6.2.

Table 6.2. Central Composite Rotatable Design (CCRD) for the independent variables considered for the PLE extraction with aqueous solutions of 1,2-propanediol from juçara fruit samples at the pressure of 10 MPa and flow rate of 1mL/min and the results obtained for the extraction assays in terms of yield of extraction of anthocyanins.

Assay	Temperature (K)	C _{alkanediol} (wt%)	pH	C _{cyanidin-3-O-glucoside} (mg·g _{dry biomass} ⁻¹)	C _{cyanidin-3-O-rutinoside} (mg·g _{dry biomass} ⁻¹)	Yield of Extraction (mg _{anthocyanins} ·g _{dry biomass} ⁻¹)
1	333.2 (-1)	15 (-1)	3.0 (-1)	2.73	9.62	12.35
2	373.2 (+1)	15 (-1)	3.0 (-1)	3.52	12.08	15.60
3	333.2 (-1)	45 (+1)	3.0 (-1)	3.40	11.51	14.91
4	373.2 (+1)	45 (+1)	3.0 (-1)	4.06	13.82	17.88
5	333.2 (-1)	15 (-1)	6.0 (+1)	3.62	12.94	16.56
6	373.2 (+1)	15 (-1)	6.0 (+1)	4.72	15.81	20.53
7	333.2 (-1)	45 (+1)	6.0 (+1)	3.86	12.72	16.58
8	373.2 (+1)	45 (+1)	6.0 (+1)	4.81	15.91	20.72
9	320.2 (-1.67)	30 (0)	4.5 (0)	2.70	9.52	12.22
10	387.2 (+1.67)	30 (0)	4.5 (0)	2.91	10.35	13.26
11	353.2 (0)	4.8 (-1.67)	4.5 (0)	2.79	10.12	12.91
12	353.2 (0)	55.2 (+1.67)	4.5 (0)	3.89	12.88	16.77
13	353.2 (0)	30 (0)	2.0 (-1.67)	2.70	9.52	12.22
14	353.2 (0)	30 (0)	7.0 (+1.67)	4.62	15.87	20.49
15	353.2 (0)	30 (0)	4.5 (0)	2.97	10.51	13.48
16	353.2 (0)	30 (0)	4.5 (0)	3.32	11.17	14.48
17	353.2 (0)	30 (0)	4.5 (0)	3.27	11.23	14.50
18	353.2 (0)	30 (0)	4.5 (0)	2.92	10.19	13.11
19	353.2 (0)	30 (0)	4.5 (0)	3.13	10.89	14.01

The model was tested at 10% significance level. From the Pareto chart, obtained before the successive adjustments (Figure S3 in the **Appendix H**) the three variables were found to be significant for the extraction efficiency (*Y*) of anthocyanins, namely: x_1 (temperature), x_2 (C_{alkanediol}), x_3 (pH); the quadratic effect of both x_2 (C_{alkanediol}) and pH and the interaction between temperature and pH. After excluding the parameters that were not statistically significant from the model, the linear correlation of x_2 was also not included and the final model obtained is described in Equation 6.2:

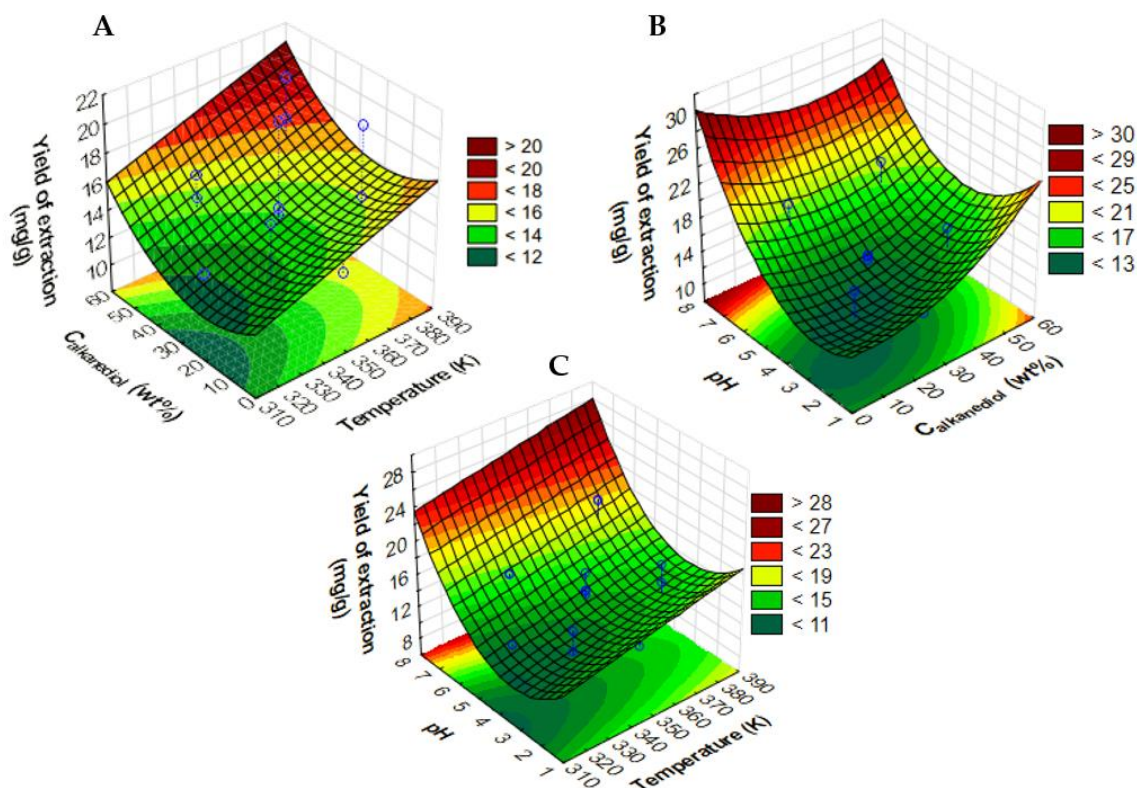
$$Y = -3.68 + 5.88 * 10^{-2}x_1 + 3.20 * 10^{-3}x_2^2 - 3.14x_3 + 5.99 * 10^{-1}x_3^2 - 2.98 * 10^{-2}x_2x_3 \quad (6.2)$$

The regression coefficients and statistical parameters for the model can be seen in Table S1 of **Appendix H**. This model presents a $R^2 = 0.81$, and the F test confirms the global significance of the model (p-value, $1.89 * 10^{-6}$). According to the proposed model and three-dimensional (3D) response (see Figure 6.2), the temperature (x_1) has a positive effect on the anthocyanins extraction, although pH (x_3) is the most significant variable.

From Table 6.2, it is also evident that keeping the same temperature and concentration conditions, higher increases in extraction yield were found for pH 6.0 compared to pH 3.0. Although in the literature it is accepted that lower pH favors the stability of anthocyanins, since generally under acidic conditions anthocyanins exist as flavylium cation form, which shows lower degradation (HUBBERMANN *et al.*, 2006), in this work pH around the neutrality, favors the extraction of anthocyanins by alkanediols.

The results depicted in Figure 6.2 evidence that for PLE, the extreme upper values in alkanediol concentration, temperature, and pH present the highest yield of anthocyanins. In fact, the predicted values from the model show that aqueous solutions of 1,2-propanediol at 55.2 wt%, 387.2 K, and pH around 7.0, provide the highest yield of extraction of anthocyanins. The accuracy of the model was checked by an experimental validation test at the optimum operational conditions with a low deviation (6.2%) compared to the predicted value (experimental: $23.11 \pm 0.06 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$; predicted: $24.63 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). The optimal conditions coincide with the most extreme conditions used experimentally, which were chosen considering restrictions that must be considered: (i) higher concentrations of 1,2-propanediol increase the solvent viscosity to a level not supported by the PLE pump and (ii) high temperatures accelerate the thermal degradation of anthocyanins by oxidation, hydrolysis, and polymerization reactions (ENARU *et al.*, 2021).

Figure 6.2. Response surface plots obtained for the CCRD (2^3) applying PLE regarding the alkanediol concentration ($C_{\text{alkanediol}}$ in wt%), temperature (K), and pH in terms of yield of extraction of anthocyanins ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). (A) $C_{\text{alkanediol}}$ and temperature, (B) pH and $C_{\text{alkanediol}}$, and (C) Temperature and pH.



In this work, the highest temperature tested (387.2 K) was the optimal condition for extraction of anthocyanins from juçara pulps. Different results were obtained by Garcia-Mendoza *et al.* (GARCIA-MENDOZA *et al.*, 2017), i.e., the highest anthocyanins yield ($1.76 \pm 0.09 \text{ mg/g dr}$) in PLE was obtained from juçara residues at 313.2 K using acidified water, since at higher temperatures (333.2 and 353.2 K) the authors report a reduction in the anthocyanins content. On the contrary, in the work of Benvenuti (BENVENUTTI; ZIELINSKI; FERREIRA, 2022), the highest content of anthocyanins from jaboticaba peels by PLE, and using DES aqueous solutions, was found at temperatures around 363.2 K.

The differences observed in these works may be related to the solution chemistry and kinetics of dissolution. For instance, in the case of cyanidin 3-O-glucoside the predominant flavylum cation, a highly water-soluble form (due to the structure moieties of sugars) is found until pH values around 4.0 (OLIVAS-AGUIRRE *et al.*, 2016), but as the pH rises the flavylum cation gives rise to the prevalent quinoidal form. Torskangerpoll and Andersen (TORSKANGERPOLL; ANDERSEN, 2005), studied the

changes in colour, solubility and stability regarding the pH range for three anthocyanins, including cyanidin 3-O-glucoside, and they showed different times of dissolution for this specific anthocyanin at different pH buffer solutions: at pH around 5.0 two days were necessary for complete dissolution, at pH from 6.0 to 6.9 one day was required and at the pH 7.2 it took just 1 hour.

In this work, for PLE extraction, the neutral pH was the most appropriate for anthocyanins solubilization and extraction, considering the short extraction time of 13 minutes. At the end of the extraction applying the optimum conditions (387.2 K, pH 7.0, and 55.2 wt% of alkanediol) the pH of the extracts was measured and hovered to 4.5, which shows that the biomass, containing a high concentration of anthocyanins, acidified the extract. In fact, the pH in juçara fruit and pulp is around 4.5 to 5.6 (SCHULZ *et al.*, 2016), acidifying the final extract. For comparison, three replicas of the optimal conditions, but changing the pH from 7.0 to 2.0, were performed, and the extraction response was lower ($15 \pm 2 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$), as expected. The final extracts pH for these last assays was 4.0, which means that the biomass increased the pH of the alkanediol solution during the extraction. It is important to note that in the work of Torskangerpoll and Andersen (TORSKANGERPOLL; ANDERSEN, 2005), an increase of only 0.3 in the pH was sufficient to decrease the time of dissolution of cyanidin 3-O-glucoside from one day to one hour, which is probably a result of different forms of anthocyanins in equilibrium.

Since the concentration of alkanediol was not one of the most significant variables for the anthocyanin extraction by PLE, contrary to the pH, which was the most impacting factor, the range of pH evaluated for UAE was extended (from 1.9 to 12.0), keeping the same alkanediol concentrations studied for the PLE and evaluating the effect of temperature through changes in the percentage of the system power (amplitude).

6.1.4.2. Optimization of UAE Extraction Conditions

For the UAE, the optimization of the extraction conditions was performed considering amplitude (x_1), corresponding to a percentage of the power of the ultrasonic probe (400 W), evaluated from 10-50%, the concentration of the 1,2-propanediol (x_2 , $C_{\text{alkanediol}}$) in the same range used with PLE (from 4.8 to 55.2 %wt) and pH (x_3), evaluated from 1.9 to 12.0 based on a CCDR (2^3). The time was set up to 10 min due to preliminary kinetic tests (see Figure S4 of **Appendix H**). For method comparison, the response used

in the predictive model was the same as PLE optimization, the yield of extraction of anthocyanins (expressed in $\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). The yields of extraction of both anthocyanins are shown in Table 6.3, along with the corresponding extracting conditions.

Table 6.3. Central Composite Rotatable Design (CCRD) for the independent variables considered for the UAE extraction with aqueous solutions of 1,2-propanediol from juçara fruit samples at 10 min and the results obtained for the extraction assays in terms of yield of extraction of anthocyanins.

Assay	Amplitude (%)	C _{alkanediol} (wt%)	pH	C _{cyanidin-3-O-glucoside} ($\text{mg} \cdot \text{g}_{\text{dry biomass}}^{-1}$)	C _{cyanidin-3-O-rutinoside} ($\text{mg} \cdot \text{g}_{\text{dry biomass}}^{-1}$)	Yield of Extraction ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$)
1	18 (−1)	15 (−1)	4.0 (−1)	7.64	36.64	44.29
2	42 (+1)	15 (−1)	4.0 (−1)	8.72	40.67	49.39
3	18 (−1)	45 (+1)	4.0 (−1)	8.72	38.94	47.66
4	42 (+1)	45 (+1)	4.0 (−1)	9.45	41.50	50.95
5	18 (−1)	15 (−1)	10.0 (+1)	7.64	35.66	43.30
6	42 (+1)	15 (−1)	10.0 (+1)	8.54	39.55	48.10
7	18 (−1)	45 (+1)	10.0 (+1)	7.63	35.89	43.52
8	42 (+1)	45 (+1)	10.0 (+1)	9.12	40.51	49.63
9	10 (−1.67)	30 (0)	7.0 (0)	7.21	34.47	41.68
10	50 (+1.67)	30 (0)	7.0 (0)	8.98	40.20	49.18
11	30 (0)	4.8 (−1.67)	7.0 (0)	8.13	39.69	47.82
12	30 (0)	55.2 (+1.67)	7.0 (0)	8.83	40.18	49.00
13	30 (0)	30 (0)	1.9 (−1.67)	8.79	39.14	47.93
14	30 (0)	30 (0)	12 (+1.67)	7.69	36.62	44.30
15	30 (0)	30 (0)	7.0 (0)	8.98	40.19	49.16
16	30 (0)	30 (0)	7.0 (0)	9.29	40.87	50.16
17	30 (0)	30 (0)	7.0 (0)	9.35	40.72	50.08
18	30 (0)	30 (0)	7.0 (0)	9.48	41.32	50.80
19	30 (0)	30 (0)	7.0 (0)	9.30	40.84	50.14

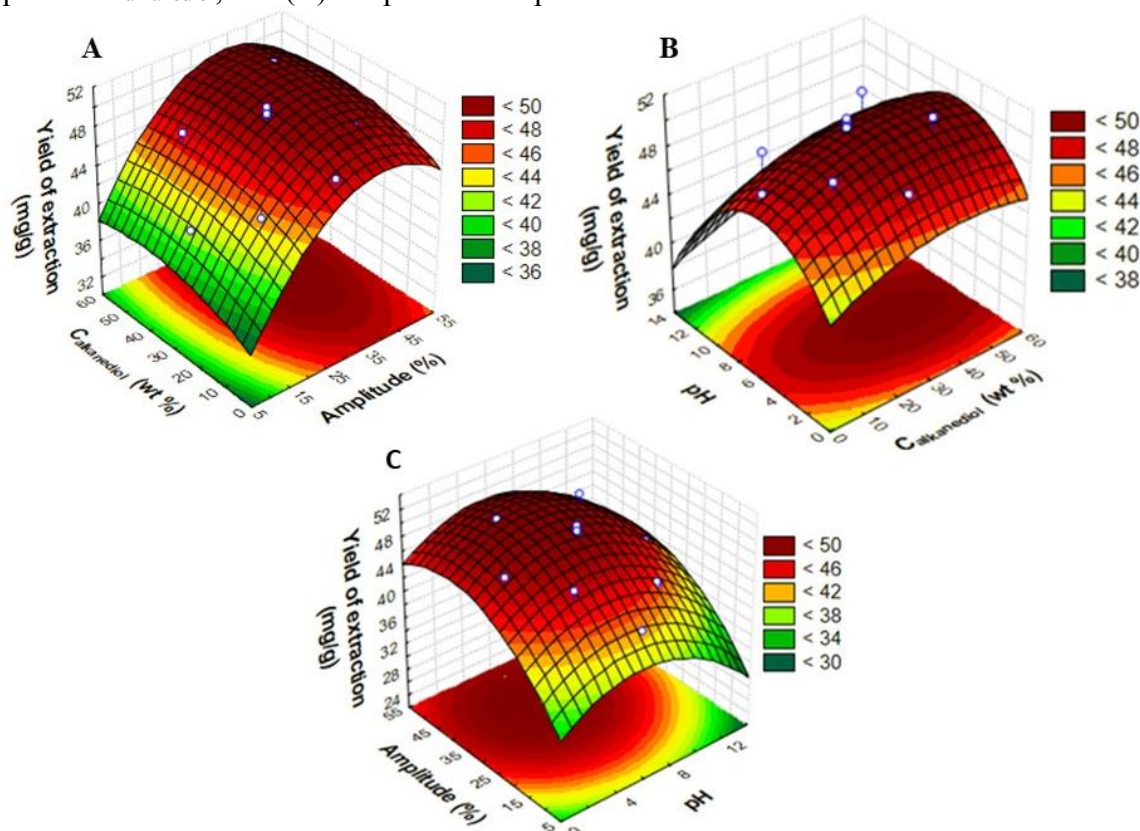
As obtained from the Pareto chart (Figure S5 of **Appendix H**), linear and quadratic forms of x_1 (amplitude), x_2 (C_{alkanediol}), and x_3 (pH) are significant to represent the extraction yield (Y). Following the usual procedure of successive fittings to ignore the non-significant statistical variables, the model is described by Equation 6.3:

$$Y = 2.73 \cdot 10^1 + 8.27 \cdot 10^{-1}x_1 - 1.05 \cdot 10^{-2}x_1^2 + 1.59 \cdot 10^{-1}x_2 - 1.94 \cdot 10^{-3}x_2^2 + 1.60x_3 - 1.39 \cdot 10^{-1}x_3^2 \quad (6.3)$$

Table S2 (**Appendix H**) compiles statistical information on the regressed model. This model presents an $R^2 = 0.95$, and the F test confirms the global significance of the model (p-value, $9.36 \cdot 10^{-7}$). According to the Equation 6.3 and three-dimensional (3D) response surfaces, depicted in Figure 6.3, the amplitude (x_1) increases the extraction yield

up to 40% of x_1 ; the alkanediol concentration (x_2) around 40 wt%, and the pH (x_3) until 6.0. This trend of the model makes intermediate values of pH, concentration, and amplitude to be considered as the optimal extraction values, as depicted by the curvature profile of the responses surfaces in Figure 6.3. In the case of UAE, aqueous solutions of 1,2-propanediol at 42.6 wt %, 40% of amplitude, and pH around 7.0 provide the highest yield of extraction of anthocyanins (the experimental value is, $50 \pm 1 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). The predicted value is ($51.18 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$) showing the very good accuracy of the model with a deviation to the experimental value close to 1.4%. Although there are differences between the variables using the two extraction methodologies (UAE or PLE), it is notorious that the yield of extraction obtained for the UAE is twice superior to the yields obtained by PLE, although the solid-liquid ratio used for UAE was lower (1:50 g/mL) than for PLE (1:13 g/mL).

Figure 6.3. Response surface plots obtained for the CCRD (2^3) applying UAE regarding the alkanediol concentration ($C_{\text{alkanediol}}$ in wt%), amplitude (%), and pH in terms of yield of extraction of anthocyanins ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). (A) $C_{\text{alkanediol}}$ and amplitude, (B) pH and $C_{\text{alkanediol}}$, and (C) Amplitude and pH.



As mentioned before, the pH of the solvent is an important factor for anthocyanins stability, and solubility, and consequently for its extraction from juçara. For

UAE, the solvent pH around 7.0 is the optimal condition to extract anthocyanins from juçara fruit, and the final pH value of these extracts was around 4.5, in line with what was observed for PLE. Surprisingly, when the optimal point was repeated, but changing the pH from 7.0 to 2.0, the final pH value of the extracts was around 2.5, which was more acidic than that found for PLE (final pH = 4.0). Moreover, the yield of extraction for this last assay at pH 2.0 is ($48 \pm 1 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$) much closer to the optimum UAE (pH 7.0 optimized value $50 \pm 1 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$) than for PLE (pH 7.0 optimized value $23.11 \pm 0.06 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$; pH 2.0 optimized value $15 \pm 2 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$) which reinforces that for UAE the variable with the most significant impact is amplitude and not pH.

For UAE the amplitude is correlated with two main factors: cavitation and temperature. Ultrasounds incite the formation of small bubbles in a cavitation phenomenon, which leads to the break of the juçara cell wall, rapidly increasing the local temperature in the system. Thus, higher ultrasonic amplitude generates a higher number of cavities, increasing the mass transfer and allowing better penetration of the medium, enhancing the extraction yield of anthocyanins (SHIRSATH; SONAWANE; GOGATE, 2012; VINATORU; MASON; CALINESCU, 2017). For each assay, the initial and final temperatures were measured (Table S3 of the **Appendix H**). While the initial temperature was always around room temperature, the final temperature (after 10 minutes) for almost all the amplitudes was between 365.4 and 372.3 K, except for 10% of the amplitude. From these values, it is possible to notice that temperature and amplitude do not have a linear correlation despite being closely related. The temperature rise gradient for larger amplitudes is higher, although the final temperatures are similar, increasing the extraction yield, as it is possible to note in Table 6.3, for the same pH and alkanediol concentration, higher yields of extraction were found for the amplitude of 42% compared to 18%, despite the final temperature difference were just about 5 K (Table S3 of the **Appendix H**). High-rise gradients of temperature decrease the solvents' viscosity and surface tension, facilitating their penetration into the biomass matrix.

Regarding the solvent concentration until 42.6 wt%, the increased alkanediol proportion reduces the solution's polarity, favoring the interaction energy between the hydrophobic and hydrophilic target compounds and the solvent molecules (CHEMAT *et al.*, 2019; MUSTAFA; TURNER, 2011). In other works, concerning the extraction of anthocyanins from juçara pulps/residuals, concentrations around 50-70% (v/v) of ethanol

exhibit higher anthocyanins yields, suggesting the intermediate concentrations of organic solvents to be the best condition for anthocyanins recovery (GARCIA-MENDOZA *et al.*, 2017; MADALÃO *et al.*, 2021; VIEIRA *et al.*, 2013).

In summary, in addition to the effects of pH (on anthocyanin stability) and solvent type and concentration (on extraction yield), the specific conditions of each extraction methodologies should also be considered when extracting anthocyanins. While temperature and pressure (up to 10 MPa (MUSTAFA; TURNER, 2011)) are the most important parameters for PLE, power, intensity and frequency are the most important parameters for UAE, all related to amplitude. In addition, temperature also has a strong influence on UAE extraction, as it can lead to altered solvent properties and affects sonochemical effects related to cavity bubble collapse (CHEMAT *et al.*, 2017). Note that in the case of UAE, temperature is also an intrinsic parameter related to amplitude. The results obtained in this work support all these facts, since temperature and amplitude are significant variables for PLE and UAE, respectively.

6.1.4.3. Solvent Effect on UAE Recovery of Anthocyanins-Rich Extract

Subsequently, anthocyanin-rich extracts were obtained by UAE at the optimum extraction conditions established in the previous section using different solvents, namely the 1,2-alkanediol series (1,2-ethanediol, 1,2-propanediol, 1,2-butanediol, 1,2-pentanediol and 1,2-hexanediol), the glycerol ethers [1.0.1], [2.0.2] and [2.0.0], and water and ethanol, as control. The judicious choice of the alkanediols in question was based on three particularities of these solvents: (i) their cost, (ii) applicability and (iii) previous experience (ABRANCHES *et al.*, 2022; SOARES *et al.*, 2020). The 1,2-alkanediols, and also the glycerol ethers, showed to be good solvents for the solubility enhancement of phenolic compounds. The results regarding the yield of extraction, TPC and antioxidant activity by ABTS are summarized in Figure 6.4 and Table 6.4, respectively.

Figure 6.4. Comparison of different solvents in the extraction of anthocyanins (in terms of yield of extraction in $\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$) and their corresponding standard deviations. Purple bars represent the 1,2-alkanediols, and light pink bars represent the glycerol ethers studied. Different letters represent statistically different values ($p \leq 0.10$) while the same letters represent statistically equal values ($p > 0.10$) by Tukey test from ANOVA.

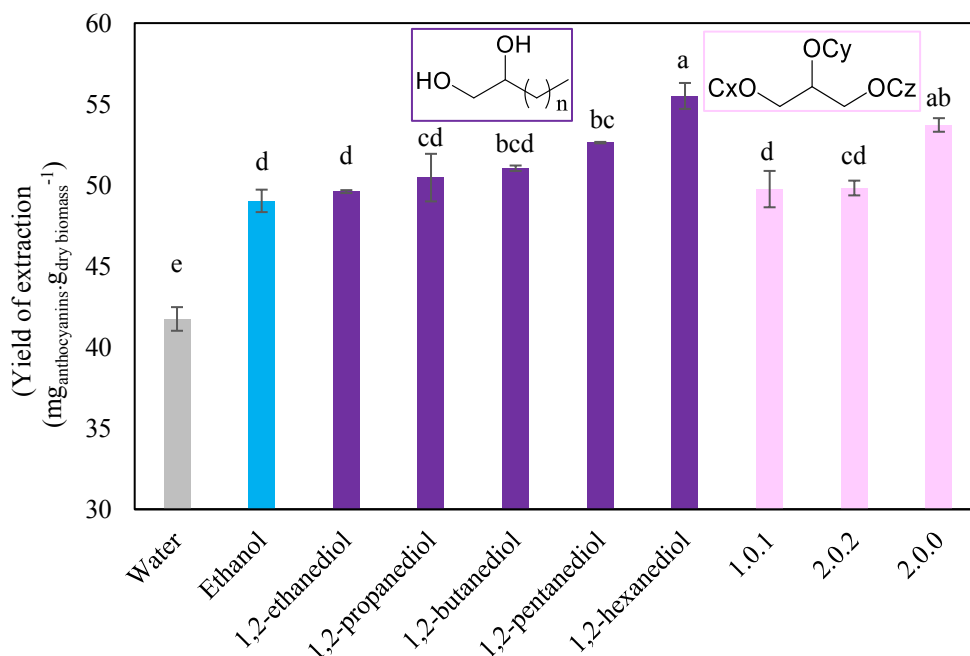


Table 6.4. Characterization of the juçara's extracts obtained by UAE, using different solvents in terms of yield of extraction, TPC, and ABTS and the corresponding standard deviation (s).

Solvent	Yield of Extraction $\pm s$ ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$)	TPC $\pm s$ ($\text{mg}_{\text{GAE}} \cdot \text{g}_{\text{dry biomass}}^{-1}$)	ABTS $\pm s$ ($\mu\text{mol}_{\text{TE}} \cdot \text{g}_{\text{dry biomass}}^{-1}$)
Water	41.8 $\pm$ 0.7 ^e	104.1 $\pm$ 0.8 ^d	1163 $\pm$ 21 ^d
1,2-ethanediol	49.60 $\pm$ 0.09 ^d	107 $\pm$ 8 ^d	1241 $\pm$ 8 ^{bc}
1,2-propanediol	50 $\pm$ 1 ^{cd}	117 $\pm$ 5 ^c	1302 $\pm$ 32 ^a
1,2-butanediol	51.0 $\pm$ 0.2 ^{bcd}	130 $\pm$ 6 ^a	1192 $\pm$ 16 ^{cd}
1,2-pentanediol	52.63 $\pm$ 0.05 ^{bc}	127 $\pm$ 4 ^{ab}	1267 $\pm$ 35 ^{ab}
1,2-hexanediol	55.5 $\pm$ 0.8 ^a	126 $\pm$ 5 ^{abc}	1302 $\pm$ 48 ^a
Ethanol	49.0 $\pm$ 0.7 ^d	120 $\pm$ 5 ^{abc}	1229 $\pm$ 26 ^{bc}
[1.0.1]	50 $\pm$ 1 ^d	125 $\pm$ 11 ^{abc}	1222 $\pm$ 13 ^{bc}
[2.0.2]	49.8 $\pm$ 0.5 ^{cd}	126 $\pm$ 1 ^{abc}	1215 $\pm$ 25 ^{bcd}
[2.0.0]	53.7 $\pm$ 0.4 ^{ab}	119 $\pm$ 2 ^{bc}	1255 $\pm$ 35 ^{ab}

Different letters represent statistically different values ($p \leq 0.10$) while the same letters represent statistically equal values ($p > 0.10$) by Tukey test from ANOVA.

The yield of extraction ranged from 41.8 to 55.5 $\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$, with the highest value obtained for 1,2-hexanediol (33% higher than water), followed by the glycerol ether [2.0.0] (28.7% higher than water). The results reported in Table 6.4 and Figure 6.4 suggest that increasing the alkyl chain size of alkanediols increases the yield

of extraction of anthocyanins, due to the change in the polarity of the solvent system that favors the solubilization. Although anthocyanins solubility studies are scarce, mainly due to their high commercial value in high purities, the study of other model molecules (also presented in juçara fruit pulps), such as phenolic acids, indicates that increasing the alkyl chain size of alkanediols favors the solubilization of these compounds (ABRANCHES *et al.*, 2022). As mentioned before, anthocyanins are generally soluble in water. However, the polyphenolic structure adds a measure of hydrophobic character, which gives relevance to organic solvents. The combination of this polar and non-polar anthocyanins nature makes alkanediol/water mixtures good solvents for their solubilization since alkanediols also exhibit an amphiphilic structure.

Regarding TPC the highest values were found for 1,2-butanediol, 1,2-pentanediol, and 1,2-hexanediol alkanediols and the symmetric glycerol ethers [1.0.1] and [2.0.2] (being 20-25% higher than in water). This is probably a consequence of the extraction of more apolar phenolic compounds such as phenolic acids and flavonoids. To evaluate the functional properties of the juçara pulps extracts, the antioxidant activity was assessed by ABTS, and the results showed that the extracts which exhibit the best antioxidant capacity were those from 1,2 propanediol and 1,2-hexanediol. In this case, the glycerol ethers extracts presented similar ABTS values as the ethanolic ones.

These results suggest that both solvents families tested, alkanediols and glycerol ethers, have a higher ability to extract anthocyanins than water and ethanol, solvents commonly used to extract these target compounds. The 1,2-hexanediol presented a higher yield of extraction of anthocyanins, and selectivity, since even other alkanediols can extract with a similar yield of phenolic compounds (regarding TPC values), 1,2-hexanediol generates extracts with higher anthocyanins concentration and with good antioxidant capacity.

Although it is well established in the literature that more acidic pH is more suitable for anthocyanin extractions, the results presented in this work suggest that, when using alkanediols, a pH close to 7.0 can generate extracts with higher anthocyanin extraction yields than those with a more acidic pH. In terms of the process, this is an advantage of the alkanediols, since the aqueous solutions of these compounds, in the optimized concentration (46.2 wt%), present a pH close to neutrality, which eliminates the pH adjustment step (see Table S4 in the **Appendix H**).

In addition, depending on the application, alkanediols are ingredients that offer advantages to the final product (at a regulated concentration by safety assessments), as in the case of cosmetics, in which short-chain alkanediols, such as those studied in this work, confer properties such as viscosity reducers, skin conditioning, humectants and they are part of the chemical solvent constitution of cosmetics. This is an advantage over Volatile Organic Compounds (VOCs) such as ethanol, methanol, or acetone as these alkanediols may be retained in the extract (at specific concentrations) depending on the final application.

At this point, the extracts obtained using 1,2-propanediol (propylene glycol) reached the highest antioxidant activity by ABTS, which is a great advantage of this compound over the other alkanediols studied since statistically, the values obtained for the extraction yield were similar between 1,2 propanediol, 1,2 butanediol, and 1,2 pentanediol (see Figure 6.4). The biggest advantage of 1,2-propanediol compared to others is that this solvent is considered GRAS. Besides that, it is widely used in cosmetic formulations, which makes the anthocyanins-rich extracts obtained, potential ingredients for cosmetic application, with a high content of antioxidant compounds.

6.1.4.4. Juçara's Formulations Characterization

To demonstrate the potential of using the anthocyanin-rich extract from juçara in cosmetic products, this extract was used as an additive in the manufacture of natural soap and natural cream. The extract used in these products was the final juçara extract obtained by UAE under optimized conditions (42.6% 1,2-propanediol, pH 7.0, at 160 W for 10 min). Juçara's soap (JB) was prepared using the final juçara extract as an additive, without additional polishing or purification steps. On the other hand, juçara cream was formulated with the final juçara extract (JC), but also with the final juçara extract concentrated (JCC).

The production scheme of juçara natural soap is shown in Figure 6.5. After one week of curing, the pH of the soaps (JB and BB) was measured and the final pH for both samples was about 9.5. The acceptable pH range for natural soaps is between 8.0 and 10, since a pH above 10 expresses a high percentage of unknown and unsaponifiable substances due to incomplete alkaline hydrolysis. In addition, the high alkalinity of soaps destroys the protective acid mantle of human skin, which acts as a protective barrier

against the penetration of microbes. On the other hand, the use of a weakly alkaline soap is less harmful to the skin and favors detergency properties (ATOLANI *et al.*, 2016).

Figure 6.5. Juçara's natural soap production. (A)- oils and butter heating; (B) – melted butter and oils mixture; (C)- soap mass before additives; (D)- juçara extract (42.6% of 1,2-propanediol in 1:50 g/mL of juçara dried pulp); (E)- parchment paper-lined molds filled with soap mass; (F)- loaf of soap after 24h; (G)- soap bars; and (H)-soap bars after cure.



Due to the antimicrobial and antifungal effects of essential oils, cosmetic preparations such as natural herbal soaps do not necessarily require an additional chemical preservative if they contain an essential oil or a single compound as an active agent (SARKIC; STAPPEN, 2018), such as rosemary oil or, in the case of this work, juçara extract. The antioxidants present in essential oils and juçara fruits are free radical scavengers that protect soaps and human skin from oxidative stress, and also help prevent the decomposition of fats or oils by oxidation (RAMBABU *et al.*, 2020).

Regarding the three creams studied (Figure 6.6), the pH of each cream was about 4.0, which corresponds to an acceptable pH for topical application (between 4.0 and 6.0) (LUKIĆ; PANTELIĆ; SAVIĆ, 2021). In addition, the pH values obtained also show that the juçara extracts do not alter the final pH of the creams.

Figure 6.6. Formulation creams evaluated in this work. (CC) – Control cream, (JC) – Juçara cream and (CJC) Concentrated juçara cream.



Table 6.5 shows the antioxidant activity by ABTS for the soaps and creams produced. Comparing the two soaps, the antioxidant activity by ABTS method is 2.4 times higher for JB than for BB. It is important to mention that, in the formulation of both soaps (BB and JB) essential oils additives were incorporated with antioxidant activity, as confirmed by the ABTS value found for BB = $6.6 \mu\text{mol}_{\text{TE.gformulation}}^{-1}$. For the creams, the antioxidant activity for the control was around zero, since the cream base formulation did not contain antioxidants as additives. On the other hand, the addition of juçara extracts in the cream formulation resulted in an increase in antioxidant activity, with the concentrated juçara cream (JCC) showing 2.2-fold antioxidant activity compared to juçara cream (JC), what means that, for creams, concentrating the juçara extract in 50% also increases the antioxidant activity of the final creams in around 50%. In conclusion, the use of juçara extracts as an additive in cosmetic products (soap and cream) leads to an increase in antioxidant activity. In this particular case, a similar increase in antioxidant activity was observed in both products studied.

Table 6.5. Antioxidant activity by the ABTS method for soap bars and creams formulations.

Formulation	ABTS ($\mu\text{mol}_{\text{TE.gformulation}}^{-1}$)
Base bar (BB)	6.6 ± 0.6
Juçara bar (JB)	16.0 ± 0.5
Control cream (CC)	0.10 ± 0.01
Juçara cream (JC)	1.5 ± 0.1
Concentrated juçara cream (CJC)	3.3 ± 0.2

Cream formulations using bio propylene glycol instead of commercial propylene glycol in UAE extractions were also performed, in order to evaluate the direct

applicability of a solvent from renewable sources. Without major surprises, similar levels of antioxidant activity were found by ABTS, suggesting that bio-propylene glycol-based extracts can also be used as a formulation ingredient for the production of 100% natural cosmetic products. It is important to note that propylene-glycol is widely used as an ingredient for cosmetic formulations, including the production of soaps and creams, acting as a humectant, skin-conditioning agent, viscosity reducer and also as a fragrance ingredient (BEHR *et al.*, 2008; GÜNTER VERHEUGEN, 2006). In this sense, the use of juçara rich-anthocyanins extracts promotes both antioxidant activity and improvement of cosmetic formulations through the inherent physical-chemical properties of the solvent.

6.1.6. Conclusions

The development of new approaches to valorize the juçara fruits is essential for maintaining the Atlantic Forest biodiversity and encouraging innovative ideas for its sustainable use. This work proposes the use of juçara fruit pulp as a rich source of bioactive antioxidant compounds. Therefore, this work aims to study the extraction of phenolic compounds and anthocyanins by two emergent methods (PLE and UAE) using aqueous solutions of biobased solvents, namely alkanediols and glycerol ethers.

For the valorization of juçara fruits, the optimum PLE and UAE conditions, using aqueous solutions of 1,2-propanediol as reference solvent, were established. For PLE the best processing conditions were 55.2 (wt%) of 1,2-propanediol, 387.2 K, and pH 7.0 (achieving the extraction yield of $23.11 \pm 0.06 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). For UAE the best conditions were 42.6 (wt%) of 1,2-propanediol, 40% amplitude, and pH 7.0 (yield of $50 \pm 1 \text{ mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$). The results found for UAE exceeded twice the yield of extraction by PLE, and that method was selected for a solvent screening with alkanediols and glycerol ethers. For the optimized conditions, 1,2-hexanediol was the best solvent for anthocyanins extraction, presenting extracts with high antioxidant capacity and overcoming the extraction yield with conventional solvents like water or ethanol.

In the literature it is well established that acidic pH is more suitable for obtaining anthocyanin-rich extracts. This work showed that using alkanediols, a pH close to 7.0 confers higher extraction yields, which is an advantage of these compounds since their aqueous solutions have a pH close to neutrality. In some applications, such as in the cosmetics industry, some alkanediols work as formulations ingredients, which allows juçara extracts to be used without polishing steps, differing from what is expected for

volatile organic solvents such as ethanol, as the case of both formulations presented in this work, juçara natural soap and incorporated juçara creams. Therefore, alkanediols are green alternatives to replace organic solvents and for juçara fruits valorization.

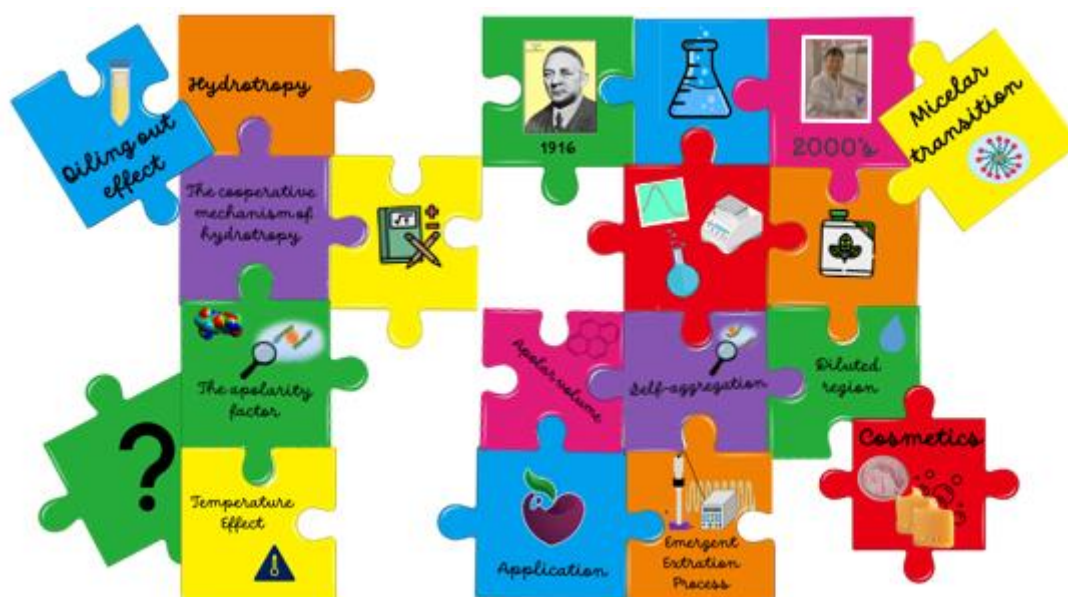
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7. CONCLUSIONS AND PERSPECTIVES

The present work reports a comprehensive study to disclose some aspects (pieces) of the big puzzle that are the mechanism of hydrotrope (see Figure 7.1). It is also an important contribution to the design of hydrotropes for specific demands and the development of novel applications for these compounds, such as the solubilization of highly hydrophobic compounds; the extraction of bioactive molecule from natural matrices; the direct use of the rich-phenolic extracts for industrial applications; and the recovery, purification, and crystallization of solutes by aqueous hydrotrope solutions.

Figure 7.1. The action puzzle of the hydrotropy mechanism: from the hydrotropes proposal in 1916 until the findings proposed by this work.



Source: the authors. This scheme was presented as a part of an oral communication on the conference Research Summit 2022, in Aveiro, Portugal.

First, inspired by the lack of information regarding the mechanism of action of hydrotropes and in the recent statistical thermodynamics-based cooperative model of hydrotropy, two classes of hydrotropes were selected as model hydrotropes namely, the glycerol ethers and the alkanediols. These families of non-ionic hydrotropes were used as a powerful way to understand the overall mechanisms and rationally design better green hydrotropes. Solubilities curves, proton nuclear magnetic resonance spectroscopy (^1H -NMR), and the chemical environment probes, such as Kamlet-Taft solvatochromic parameter π^* and P_y (pyrene) polarity scales were successfully measured and determined.

These data provided the information that was rationalized using the cooperative-hydrotrope and Setschenow models to clarify the molecular mechanisms.

The solubility measurements using monoglycerol ethers and alkanediols as hydrotropes have demonstrated the exceptional ability of these solvents to increase the solubility of hydrophobic compounds. Increasing the size of the alkyl side chain of monoglycerol ethers provided key insight into the mechanism of hydrotropy revealing that the magnitude of the solubility enhancement of a given solute correlates well with the apolarity of the hydrotrope and reaches a maximum when the apolar surface area of the hydrotrope and solute match. This maximum was discussed in terms of competing solute-hydrotrope and hydrotrope-hydrotrope interactions.

The study of two different families of alkanediols (1,2-alkanediols and 1,n-alkanediols) to solubilize syringic acid revealed contradictory findings in the dilute and non-dilute hydrotrope regions, a consequence of both (i) the hydrotrope self-aggregation that negatively impacts the solubility enhancement and (ii) the apolar volumes of 1,2 and 1,n-alkanediols, that drive the different levels of aggregation between solute, hydrotrope, and water. Besides that, in addition to the hydrophobicity of the hydrotrope, the apolar volume of the solute for the formation of aggregations must also be considered for hydrotropic solubilizations, since large molecules (such as Reichardt's dye and pyrene) are not so easily aggregated by the hydrotrope as simple as phenolic acids.

Applying polarity scales (π^* and P_y) it was also possible to assess the contributions of different interactions on the solubilization using alkanediols as hydrotropes. A transition point from hydrotropic to micellar aggregation in the longer-alkyl chain alkanediols was identified. In future studies, it would be valuable to experimentally confirm the nature of the aggregates formed by alkanediols and other hydrotropes using techniques such as dynamic light scattering (DLS), small-angle neutron scattering (SANS), or other relevant measurements.

The temperature effect on hydrotrope was also evaluated for six selected non-ionic hydrotropes and showed, once more, two different mechanisms in the non-concentrated-hydrotrope and concentrated-hydrotrope regions. The first region is controlled by water dissolution of the solute, where similar solubility enhancements (S/S_0) were found despite temperature variations. The second region is driven by hydrotrope dissolution of solute, where the self-aggregation phenomena are more evident, and the S/S_0 is higher for lower temperatures. From the van't Hoff equation, the solution

thermodynamic properties (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) have been estimated, indicating the dissolution process of syringic acid in aqueous hydrotropes solutions as endothermic and entropy-driven. To gain a deeper understanding of the mechanism by which temperature affects hydrotropy, it would be beneficial to evaluate the self-aggregation mechanisms of hydrotrope molecules through the use of thermodynamic experimental and computational tools, including COSMO-RS predictions and chemical environment probes.

While several factors play a role in governing hydrotropy, the solubilization of vanillic acid by glycol ethers revealed the oiling-out effect, which involves the formation of liquid-liquid phase separation. This phenomenon occurs only at specific hydrotrope concentrations and results from a combination of factors, including the interaction between hydrotrope and solute and the hydrophobicity of the solute. To better understand this effect, it would be crucial, in future research, to investigate the impact of temperature, as partial phase immiscibility depends on this factor. Additionally, accurately quantifying the liquid-liquid separation boundaries is crucial for designing and controlling crystallization and purification processes. Future studies can focus on precisely quantifying this phenomenon through experimental methodologies that determine liquid-liquid equilibrium and the corresponding phase diagrams.

As a final step, we examined the potential use of the hydrotropes studied in this work for extracting phenolic compounds, particularly anthocyanins, from juçara fruit, obtained from the juçara palm tree. By combining aqueous solutions of glycerol ethers or alkanediols with emergent extraction techniques, such as pressurized liquid extraction and ultrasound-assisted extraction, we were able to produce extracts with a high concentration of phenolic compounds and strong antioxidant activity. Future research can investigate the combination of multiple emerging extraction techniques to improve the recovery of target compounds, such as using SFE (sc-CO₂) followed by UAE. Lastly, we applied the anthocyanin-rich extracts directly in cosmetic formulations (soaps and creams), taking advantage of their powerful antioxidant capacity and the inherent properties of the solvents, such as their emollient and humectant properties, which are well-suited for cosmetic applications.

New approaches to valorize the juçara fruits are essential for maintaining the Atlantic Forest biodiversity and encouraging innovative ideas for its sustainable use. In addition to the scientific impact of this last work, an entrepreneurial component based on

the anthocyanins-rich extracts was implemented. Undoubtedly, the rich-phenolic extracts obtained from biomass sources are a good starting point for producing new cosmetics. The results achieved in this thesis provide a solid foundation for exploring other potential applications of phenolic-rich extracts, such as in skin care products (lotions and serums), shampoos, and conditioners, as well as in other industries, including food and beverages as flavor enhancers and colorants; pharmaceuticals as natural medicines; agriculture as natural pesticides, herbicides, and plant growth enhancers; and the textile industry as natural dyes and for fabric pre-treatment.

In addition to the comprehensive study of the hydrotropic phenomena proposed by this project, other important questions have been considered in this work, namely the use of bio-based solvents, the study of more efficient and less environmentally hazardous synthesis methodologies, the analysis of solvent recovery and reuse, the extraction of value-added compounds from biomass and also the application of the rich-phenolics extracts to cosmetic applications for juçara's fruit valorization, all converging with the principles proposed within the Green Chemistry approach. The milestones are to present real alternatives for solubilization and extraction of bioactive compounds and select or design hydrotropes for specific applications.

REFERENCES

ABBOTT, A. P. *et al.* Deep eutectic solvents formed between choline chloride and carboxylic acids: versatile alternatives to ionic liquids. **Journal of the American Chemical Society**, v. 126, n. 9, p. 9142–9147, 2004.

ABBOTT, S.; BOOTH, J. J.; SHIMIZU, S. Practical molecular thermodynamics for greener solution chemistry. **Green Chemistry**, v. 19, n. 1, p. 68–75, 2016.

ABRANCHES, D. O. *et al.* Unveiling the mechanism of hydrotropy: Evidence for water-mediated aggregation of hydrotropes around the solute. **Chemical Communications**, v. 56, n. 52, p. 7143–7146, 2020a.

ABRANCHES, D. O. *et al.* Solubility Enhancement of Hydrophobic Substances in Water / Cyrene Mixtures: A Computational Study. **Industrial & Engineering Chemistry Research**, v. 59, n. 40, p. 18247–18253, 2020b.

ABRANCHES, D. O. *et al.* The Perspective of Cooperative Hydrotropy on the Solubility in Aqueous Solutions of Cyrene. **Industrial & Engineering Chemistry Research**, v. 59, n. 41, p. 18649–18658, out. 2020c.

ABRANCHES, D. O. *et al.* The impact of the counterion in the performance of ionic hydrotropes. **Chemical Communications**, v. 57, p. 2951–2954, 2021.

ABRANCHES, D. O. *et al.* The impact of size and shape in the performance of hydrotropes: a case-study of alkanediols. **Physical Chemistry Chemical Physics**, v. 24, n. 13, p. 7624–7634, 2022.

ACREE, W. E. *et al.* Spectrochemical Investigations of Preferential Solvation. 2. Compatibility of Thermodynamic Models versus Spectrofluorometric Probe Methods for Tautomeric Solutes Dissolved in Binary Mixtures. **The Journal of Physical Chemistry**, v. 98, n. 10, p. 2537–2544, mar. 1994.

ACREE, W. E.; TUCKER, S. A.; WILKINS, D. C. Spectrochemical investigations of preferential solvation: fluorescence emission behavior of select polycyclic aromatic hydrocarbon solute probes dissolved in mixed solvents. **The Journal of Physical Chemistry**, v. 97, n. 43, p. 11199–11203, out. 1993.

AGUIAR, A. C. DE *et al.* Sequential high-pressure extraction to obtain capsinoids and phenolic compounds from biquinho pepper (*Capsicum chinense*). **The Journal of Supercritical Fluids**, v. 150, p. 112–121, 2019.

AL-LAL, A. M. *et al.* A new route to synthesize tert-butyl ethers of bioglycerol. **Fuel**, v. 93, p. 632–637, 2012.

ALMEIDA, J. R. M.; FÁVARO, L. C. L.; QUIRINO, B. F. Biodiesel biorefinery:

Opportunities and challenges for microbial production of fuels and chemicals from glycerol waste. **Biotechnology for Biofuels**, v. 5, p. 1–16, 2012.

ALMGREN, M. *et al.* Aggregation of Alkyltrimethylammonium Surfactants in Aqueous Poly(styrenesulfonate) Solutions. **Langmuir**, v. 8, n. 10, p. 2405–2412, 1992.

ALZAGAMEEM, A. *et al.* Lignocellulosic Biomass as Source for Lignin-Based Environmentally Benign Antioxidants. **Molecules**, v. 23, n. 10, p. 2664, out. 2018.

ANASTAS, P.; EGHBALI, N. Green Chemistry: Principles and Practice. **Chem. Soc. Rev.**, v. 39, n. 1, p. 301–312, 2010.

ANASTAS, P. T.; WARNER, J. C. **Green chemistry: theory and practice**. New York: Oxford University Press, 1998.

ANASTAS, P. T.; ZIMMERMAN, J. B. The periodic table of the elements of green and sustainable chemistry. **Green Chemistry**, v. 21, n. 24, p. 6545–6566, 2019.

ANIYA, V. *et al.* Measurement and Modeling of Solubility of para-tert-Butylbenzoic Acid in Pure and Mixed Organic Solvents at Different Temperatures. **Journal of Chemical and Engineering Data**, v. 62, n. 4, p. 1411–1421, 2017.

ANSARI, K. B.; GAIKAR, V. G. Green hydrotropic extraction technology for delignification of sugarcane bagasse by using alkybenzene sulfonates as hydrotropes. **Chemical Engineering Science**, v. 115, p. 157–166, 2014.

AROSO, I. M. *et al.* Natural deep eutectic solvents from choline chloride and betaine – physicochemical properties. **Journal of Molecular Liquids**, v. 241, p. 654–661, 2017.

ATOLANI, O. *et al.* Green synthesis and characterisation of natural antiseptic soaps from the oils of underutilised tropical seed. **Sustainable Chemistry and Pharmacy**, v. 4, p. 32–39, 2016.

AZEVEDO, N. *et al.* Phenolic compounds and carotenoids from four fruits native from the Brazilian Atlantic Forest. **Journal of Agricultural and Food Chemistry**, v. 62, p. 5072–5084, 2014.

BADHANI, B.; SHARMA, N.; KAKKAR, R. Gallic acid: a versatile antioxidant with promising therapeutic and industrial applications. **RSC Advances**, v. 5, n. 35, p. 27540–27557, 2015.

BAIRD, Z. S. *et al.* **Vapor Pressures, Densities, and PC-SAFT Parameters for 11 Bio-compounds**. [s.l.] Springer US, 2019. v. 40

BALLESTEROS-VIVAS, D.; MENDIOLA, J.; IBÁÑEZ, E. Extraction : supercritical fluid extraction. **Encyclopedia of Analytical Science**, v. 3, p. 127–133, 2019.

- BARANAUSKAITE, J. *et al.* Optimization of carvacrol, rosmarinic, oleanolic and ursolic acid extraction from oregano herbs (*Origanum onites* L., *Origanum vulgare* spp. *hirtum* and *Origanum vulgare* L.). **Natural Product Research**, v. 30, n. 6, p. 672–674, 2016.
- BAUDUIN, P. *et al.* Unified concept of solubilization in water by hydrotropes and cosolvents. **Langmuir**, v. 21, n. 9, p. 6769–6775, 2005.
- BEHR, A. *et al.* Improved utilization of renewable resources: New important derivatives of glycerol. **Green Chemistry**, v. 10, n. 1, p. 13–30, 2008.
- BELLETTINI, I. C. *et al.* Properties of aqueous solutions of hydrophobically modified polyethylene imines in the absence and presence of sodium dodecylsulfate. **Journal of Colloid and Interface Science**, v. 370, n. 1, p. 94–101, 2012.
- BELSITO, D. V. *et al.* **Safety Assessment of Alkane Diols as Used in Cosmetics**. Washington: [s.n.]. Disponível em: <<https://www.cir-safety.org/supplementaldoc/cir-report-format-outline%0Ahttps://www.cir-safety.org/sites/default/files/pentae032014final.pdf>>.
- BENVENUTTI, L.; ZIELINSKI, A. A. F.; FERREIRA, S. R. S. Pressurized aqueous solutions of deep eutectic solvent (DES): A green emergent extraction of anthocyanins from a Brazilian berry processing by-product. **Food Chemistry: X**, v. 13, p. 100236, 2022.
- BERMEJO, D. V. *et al.* Extraction of caffeine from natural matter using a bio-renewable agrochemical solvent. **Food and Bioproducts Processing**, v. 91, n. 4, p. 303–309, 2013.
- BICUDO, M. O. P.; RIBANI, R. H.; BETA, T. Anthocyanins, phenolic acids and antioxidant properties of Juçara Fruits (*Euterpe edulis* M.) along the On-tree ripening process. **Plant Foods for Human Nutrition**, v. 69, p. 142–147, 2014.
- BOOTH, J. J. *et al.* Hydrotrope accumulation around the drug: the driving force for solubilization and minimum hydrotrope concentration for nicotinamide and urea. **Physical Chemistry Chemical Physics**, v. 17, n. 12, p. 8028–8037, 2015.
- BOOTH, J. J.; ABBOTT, S.; SHIMIZU, S. Mechanism of hydrophobic drug solubilization by small molecule hydrotropes. **Journal of Physical Chemistry B**, v. 116, n. 51, p. 14915–14921, 2012.
- BORGES, G. D. S. C. *et al.* Chemical characterization, bioactive compounds, and antioxidant capacity of jussara (*Euterpe edulis*) fruit from the Atlantic Forest in southern Brazil. **Food Research International**, v. 44, n. 7, p. 2128–2133, 2011.

BRAGA, A. R. C. *et al.* Lactobacillus fermentation of jussara pulp leads to the enzymatic conversion of anthocyanins increasing antioxidant activity. **Journal of Food Composition and Analysis**, v. 69, p. 162–170, 2018a.

BRAGA, A. R. C. *et al.* Bioavailability of anthocyanins: Gaps in knowledge, challenges and future research. **Journal of Food Composition and Analysis**, v. 68, p. 31–40, maio 2018b.

BUCHECKER, T. *et al.* The impact of the structuring of hydrotropes in water on the mesoscale solubilisation of a third hydrophobic component. **Physical Chemistry Chemical Physics**, v. 19, n. 3, p. 1806–1816, 2017.

BYRNE, F. P. *et al.* Tools and techniques for solvent selection : green solvent selection guides. **Sustainable Chemical Processes**, v. 4, p. 1–24, 2016.

CAI, C. *et al.* Maleic acid as a dicarboxylic acid hydrotrope for sustainable fractionation of wood at atmospheric pressure and ≤ 100 °C: mode and utility of lignin esterification. **Green Chemistry**, 2020.

CARDOSO, A. L. *et al.* An Update on the Biological Activities of Euterpe edulis (Juçara). **Planta Medica**, v. 84, p. 487–499, 2018.

CARVALHO, C. DA S. *et al.* Climatic stability and contemporary human impacts affect the genetic diversity and conservation status of a tropical palm in the Atlantic Forest of Brazil. **Conservation Genetics**, v. 18, n. 2, p. 467–478, 2017.

CASTRO, M. D. L. DE; PRIEGO-CAPOTE, F. Soxhlet extraction: past and present panacea. **Journal of Chromatography A**, v. 1217, p. 2383–2389, 2010.

CHAN, C. *et al.* Microwave-assisted extractions of active ingredients from plants. **Journal of Chromatography A**, v. 1218, n. 37, p. 6213–6225, 2011.

CHANG, J. *et al.* Methylation of glycerol with dimethyl sulfate to produce a new oxygenate additive for diesels. **Industrial & Engineering Chemistry Research**, v. 51, p. 655–661, 2012.

CHEMAT *et al.* Review of Alternative Solvents for Green Extraction of Food and Natural Products: Panorama, Principles, Applications and Prospects. **Molecules**, v. 24, n. 16, p. 3007, ago. 2019.

CHEMAT, F. *et al.* Ultrasound assisted extraction of food and natural products. Mechanisms, techniques, combinations, protocols and applications. A review. **Ultrasonics Sonochemistry**, v. 34, p. 540–560, 2017.

ChemSpider database. <http://www.chemspider.com>.

- CHEN, L. *et al.* Inexpensive ionic liquids: [HSO₄]⁻-based solvent production at bulk scale. **Green Chem.**, v. 16, n. 6, p. 3098–3106, 2014.
- CHEN, L. *et al.* Rapid and near-complete dissolution of wood lignin at ≤80°C by a recyclable acid hydrotrope. **Science Advances**, v. 3, n. 9, p. e1701735, 15 set. 2017.
- CHEOK, C. Y. *et al.* Optimization of total monomeric anthocyanin (TMA) and total phenolic content (TPC) extractions from mangosteen (*Garcinia mangostana* Linn.) hull using ultrasonic treatments. **Industrial Crops & Products**, v. 50, p. 1–7, 2013.
- CHERUBINI, F. The biorefinery concept: using biomass instead of oil for producing energy and chemicals. **Energy Conversion and Management**, v. 51, p. 1412–1421, 2010.
- CHOUBEY, S. *et al.* Medicinal importance of gallic acid and its ester derivatives: a patent review. **Pharmaceutical Patent Analyst**, v. 4, n. 4, p. 305–315, jul. 2015.
- CLARK, J. H. *et al.* Circular economy design considerations for research and process development in the chemical sciences. **Green Chemistry**, v. 18, p. 3914–3934, 2016.
- CLARKE, C. J. *et al.* Green and sustainable solvents in chemical processes. **ACS Sustainable Chemistry & Engineering**, v. 118, p. 747–800, 2018.
- CLÁUDIO, A. F. M. *et al.* The magic of aqueous solutions of ionic liquids: ionic liquids as a powerful class of catanionic hydrotropes. **Green Chemistry**, v. 17, n. 7, p. 3948–3963, 2015.
- CLÁUDIO, A. F. M. *et al.* Separation and purification technology valorization of olive tree leaves: extraction of oleanolic acid using aqueous solutions of surface-active ionic liquids. **Separation and Purification Technology**, v. 204, p. 30–37, 2018.
- COFFMAN, R. E.; KILDSIG, D. O. Effect of Nicotinamide and Urea on the Solubility of Riboflavin in Various Solvents. **Journal of Pharmaceutical Sciences**, v. 85, n. 9, p. 951–954, set. 1996.
- CRAVEIRO, R. *et al.* Properties and thermal behavior of natural deep eutectic solvents. **Journal of Molecular Liquids**, v. 215, p. 534–540, 2016.
- CUCCINIELLO, R. *et al.* Synthesis of monoalkyl glyceryl ethers by ring opening of glycidol with alcohols in the presence of Lewis acids. **ChemSusChem**, v. 9, p. 1–5, 2016.
- CUI, Y. Parallel stacking of caffeine with riboflavin in aqueous solutions: The potential mechanism for hydrotropic solubilization of riboflavin. **International Journal of Pharmaceutics**, v. 397, n. 1–2, p. 36–43, set. 2010.
- CVJETKO BUBALO, M. *et al.* Green solvents for green technologies. **Journal of**

- Chemical Technology & Biotechnology**, v. 90, n. 9, p. 1631–1639, set. 2015.
- DAI, Y. *et al.* Natural deep eutectic solvents as new potential media for green technology. **Analytica Chimica Acta**, v. 766, p. 61–68, 2013.
- DAI, Y. *et al.* Tailoring properties of natural deep eutectic solvents with water to facilitate their applications. **Food Chemistry**, v. 187, p. 14–19, 2015.
- DAI, Y. *et al.* Application of natural deep eutectic solvents to the extraction of anthocyanins from *Catharanthus roseus* with high extractability and stability replacing conventional organic solvents. **Journal of Chromatography A**, v. 1434, p. 50–56, 2016.
- DALI, I. *et al.* Correlation and semi-empirical modeling of solubility of gallic acid in different pure solvents and in binary solvent mixtures of propan-1-ol + water, propan-2-ol + water and acetonitrile + water from (293.2 to 318.2) K. **Journal of Molecular Liquids**, v. 222, p. 503–519, 2016.
- DE ALBUQUERQUE, I.; MAZZOTTI, M. Crystallization process design using thermodynamics to avoid oiling out in a mixture of vanillin and water. **Crystal Growth and Design**, v. 14, n. 11, p. 5617–5625, 2014.
- DE BRUYN, M. *et al.* Geminal Diol of Dihydrolevoglucosenone as a Switchable Hydrotrope: A Continuum of Green Nanostructured Solvents. **ACS Sustainable Chemistry and Engineering**, v. 7, n. 8, p. 7878–7883, 2019.
- DESAI, M.; PARIKH, J. Thermodynamic study for aggregation behavior of hydrotropic solution. **World Academy of Science, Engineering and Technology**, v. 57, p. 227–229, 2009.
- DEVIerno KREUDER, A. *et al.* A Method for Assessing Greener Alternatives between Chemical Products Following the 12 Principles of Green Chemistry. **ACS Sustainable Chemistry & Engineering**, v. 5, n. 4, p. 2927–2935, abr. 2017.
- DHAPTE, V.; MEHTA, P. Advances in hydrotropic solutions: an updated review. **St. Petersburg Polytechnical University Journal: Physics and Mathematics**, v. 1, n. 4, p. 424–435, 2015.
- DONG, D. C.; WINNIK, M. A. The Py scale of solvent polarities. Solvent effects on the vibronic fine structure of pyrene fluorescence and empirical correlations with ET and Y values. **Photochemistry and Photobiology**, v. 35, n. 1, p. 17–21, jan. 1982.
- DONG, D. C.; WINNIK, M. A. The Py scale of solvent polarities. **Canadian Journal of Chemistry**, v. 62, n. 11, p. 2560–2565, 1984.
- DU, Y. *et al.* The liquid-liquid phase separation and crystallization of vanillin in 1-

propanol/water solution. **Fluid Phase Equilibria**, v. 409, p. 84–91, 2016.

DURAND, M. *et al.* Solubilizing and hydrotropic properties of isosorbide monoalkyl- and dimethyl-ethers. **Journal of Surfactants and Detergents**, v. 12, n. 4, p. 371–378, 2009.

EARLE, M. J. *et al.* The distillation and volatility of ionic liquids. **Nature**, v. 439, p. 831–834, 2006.

EASTOE, J.; HATZOPOULOS, M. H.; DOWDING, P. J. Action of hydrotropes and alkyl-hydrotropes. **Soft Matter**, v. 7, n. 13, p. 5917–5925, 2011.

ECKERT, F. **COSMOtherm User's Manual Version C3.0 Release 14.01, COSMOlogic GmbH & Co. Kg, Leverkusen, Germany**, 2013.

ECKERT, F.; KLAMT, A. **COSMOtherm. Version C3.0 Release 14.01. COSMOlogic. GmbH & Co. Kg, Leverkusen**, 2014.

ENARU, B. *et al.* Anthocyanins: Factors affecting their stability and degradation. **Antioxidants**, v. 10, n. 12, p. 1–24, 2021.

ERYTHROPEL, H. C. *et al.* The Green ChemisTREE: 20 years after taking root with the 12 principles. **Green Chemistry**, v. 20, n. 9, p. 1919–2160, 2018.

EYCKENS, D. J. *et al.* Determination of Kamlet-Taft parameters for selected solvate ionic liquids. **Physical Chemistry Chemical Physics**, v. 18, n. 19, p. 13153–13157, 2016.

FACHE, M.; BOUTEVIN, B.; CAILLOL, S. Vanillin production from lignin and its use as a renewable chemical. **ACS Sustainable Chemistry & Engineering**, v. 1, n. 4, p. 35–4, 2015.

FDA. **GRAS Substances (SCOGS) Database**. Disponível em: <<https://www.fda.gov/food/generally-recognized-safe-gras/gras-substances-scogs-database>>. Acesso em: 21 out. 2020.

FELZENSZWALB, I. *et al.* Toxicological evaluation of Euterpe edulis: a potential superfruit to be considered. **Food and Chemical Toxicology**, v. 58, p. 536–544, 2013.

FLETCHER, K. A. *et al.* Probing solute and solvent interactions within binary ionic liquid mixtures. **New Journal of Chemistry**, v. 27, n. 12, p. 1706–1712, 2003.

FRANK, H. S.; FRANKS, F. Structural Approach to the Solvent Power of Water for Hydrocarbons; Urea as a Structure Breaker. **The Journal of Chemical Physics**, v. 48, n. 10, p. 4746–4757, maio 1968.

FREIRE, M. G. *et al.* Aqueous biphasic systems: a boost brought about by using ionic liquids. **Chemical Society Reviews**, v. 41, n. 14, p. 4966, 2012.

FRIBERG, S. E. *et al.* The system water-ethanol-didodecyldimethylammonium bromide. Phase equilibria and vapor pressures. **Colloids and Surfaces**, v. 156, p. 145–156, 1999.

FRIBERG, S. E.; BLUTE, I. Hydrotropy. In: **Liquid Detergents (Second Edition)**. [s.l.] Taylor & Francis Group, LLC, 2006. p. 19–35.

FRIBERG, S. E.; RYDHAG, L. Solubility and association conditions of hydrotrope substances. **Tenside Surfactants Detergents**, v. 7, p. 80–83, 1970.

GABOV, K.; FARDIM, P.; GOMES DA SILVA JÚNIOR, F. Hydrotropic Fractionation of Birch Wood into Cellulose and Lignin: A New Step Towards Green Biorefinery. **BioResources**, v. 8, n. 3, p. 3518–3531, maio 2013.

GAIKAR, V. G.; SHARMA, M. M. Separations with hydrotropes. **Separations Technology**, v. 3, n. 1, p. 2–11, 1993.

GAIOTTO, F. A.; GRATAPAGLIA, D.; VENCOVSKY, R. Genetic Structure, Mating System, and Long-Distance Gene Flow in Heart of Palm (*Euterpe edulis* Mart.). **Journal of Heredity**, v. 94, n. 5, p. 399–406, 2003.

GARCIA-MENDOZA, M. DEL P. *et al.* Extraction of phenolic compounds and anthocyanins from juçara (*Euterpe edulis* Mart.) residues using pressurized liquids and supercritical fluids. **Journal of Supercritical Fluids**, v. 119, p. 9–16, 2017.

GARCÍA, A. *et al.* Extraction of phenolic compounds from virgin olive oil by deep eutectic solvents (DESs). **Food Chemistry**, v. 197, p. 554–561, 2016.

GARCIA, J. A. A. *et al.* Chemical composition and biological activities of Juçara (*Euterpe edulis* Martius) fruit by-products, a promising underexploited source of high-added value compounds. **Journal of Functional Foods**, v. 55, n. December 2018, p. 325–332, 2019.

GARCÍA, J. I. *et al.* Green solvents from glycerol. Synthesis and physico-chemical properties of alkyl glycerol ethers. **Green Chemistry**, v. 12, n. 3, p. 426–434, 2010.

GARCÍA, J. I. *et al.* Ecotoxicity studies of glycerol ethers in *Vibrio fischeri*: checking the environmental impact of glycerol-derived solvents. **Green Chemistry**, v. 17, n. 8, p. 4326–4333, 2015.

GARCÍA, J. I.; GARCÍA-MARÍN, H.; PIRES, E. Glycerol based solvents: synthesis, properties and applications. **Green Chem.**, v. 16, n. 3, p. 1007–1033, 2014.

GAUDIN, P. *et al.* Acid-Catalyzed Etherification of Glycerol with Long-Alkyl-Chain Alcohols. **ChemSusChem**, v. 4, p. 719–722, 2011.

GEORGE, J.; SASTRY, N. V. Densities, Dynamic Viscosities, Speeds of Sound, and

Relative Permittivities for Water + Alkanediols (Propane-1,2- and -1,3-diol and Butane-1,2-, -1,3-, -1,4-, and -2,3-Diol) at Different Temperatures. **Journal of Chemical and Engineering Data**, v. 48, n. 6, p. 1529–1539, 2003.

GLUSHKO, V.; THALER, M. S. R.; KARP, C. D. Pyrene Fluorescence Fine Structure as a Polarity Probe of Hydrophobic Regions: Behavior in Model Solvents. **Archives of Biochemistry and Biophysics**, v. 210, n. 1, p. 33–42, 1981.

GMEHLING, J. *et al.* **Chemical Thermodynamics for Process Simulation**. 1st. ed. [s.l.] Wiley, 2019.

GOLMAKANI, M. T. *et al.* Pressurized limonene as an alternative bio-solvent for the extraction of lipids from marine microorganisms. **Journal of Supercritical Fluids**, v. 92, p. 1–7, 2014.

GU, Y. *et al.* Heterogeneously catalyzed etherification of glycerol : new pathways for transformation of glycerol to more valuable chemicals. **Green Chemistry**, v. 10, p. 164–167, 2008.

GÜNTHER VERHEUGEN. **Cosmetic ingredients other than perfume and aromatic raw materials** **Official Journal of the European Union**, 2006.

HAJJI, M. S. Heat capacity studies of formation of micelle-like structure in aqueous solutions of some alkane polyols. **Journal of Colloid and Interface Science**, v. 257, n. 2, p. 364–370, 2003.

HEFTER, G. T.; TOMKINS, R. P. T. **The Experimental Determination of Solubilities**. [s.l.] Wiley, 2003.

HENRY, M. C.; YONKER, C. R. Supercritical fluid chromatography, pressurized liquid extraction, and supercritical fluid extraction. **Analytical Chemistry**, v. 78, n. 12, p. 3909–3915, 2006.

HERBINSKI, A. *et al.* An eco-compatible pathway to new hydrotropes from tetraols. **Green Chemistry**, v. 20, p. 1250–1261, 2018.

HERRERO, M. *et al.* Plants, seaweeds, microalgae and food by-products as natural sources of functional ingredients obtained using pressurized liquid extraction and supercritical fluid extraction. **Trends in Analytical Chemistry**, v. 71, p. 26–38, 2015.

HODGDON, T. K.; KALER, E. W. Hydrotropic solutions. **Current Opinion in Colloid and Interface Science**, v. 12, n. 3, p. 121–128, 2007.

HOPKINS HATZOPOULOS, M. *et al.* Are hydrotropes distinct from surfactants? **Langmuir**, v. 27, n. 20, p. 12346–12353, 2011.

HU, J. *et al.* Pressurized liquid extraction of ginger (*Zingiber officinale* Roscoe) with bioethanol: An efficient and sustainable approach. **Journal of Chromatography A**, v. 1218, n. 34, p. 5765–5773, 2011.

HUAMÁN-CASTILLA, N. L. *et al.* Glycerol as alternative co-solvent for water extraction of polyphenols from Carménère pomace: hot pressurized liquid extraction and computational chemistry calculations. **Biomolecules**, v. 10, n. 3, p. 1–14, 2020.

HUBBERMANN, E. M. *et al.* Influence of acids, salt, sugars and hydrocolloids on the colour stability of anthocyanin rich black currant and elderberry concentrates. **European Food Research and Technology**, v. 223, n. 1, p. 83–90, 2006.

INSTITUTO ADOLFO LUTZ. **Métodos físicos-químicos para análise de Alimentos**. IV ed. São Paulo: [s.n.].

JAYAKUMAR, C.; GANDHI, N. N. Thermodynamic study on hydrotropic aggregation behavior of benzamide. **Journal of Chemical and Engineering Data**, v. 55, n. 10, p. 4362–4368, 2010.

JESSOP, P. G. Searching for green solvents. **Green Chemistry**, v. 13, n. 6, p. 1391, 2011.

JESSOP, P. G. *et al.* Solvatochromic parameters for solvents of interest in green chemistry. **Green Chemistry**, v. 14, n. 5, p. 1245–1259, 2012.

JOHNSON, W. *et al.* Safety Assessment of 1,2-Glycols as Used in Cosmetics. **International Journal of Toxicology**, v. 31, p. 147S-168S, 2012.

KAMLET, M. J. The β -Scale of Solvent Hydrogen-Bond Acceptor (HBA) Basicities. **J. Am. Chem. Soc.**, v. 98, n. 2, p. 377–383, 1975.

KAMLET, M. J.; ABBOD, J. L. J. L.; TAFT, R. W. The Solvatochromic Comparison Method. 6. The π^* Scale of Solvent Polarities. **Journal of the American Chemical Society**, v. 99, n. 18, p. 6027–6038, ago. 1977.

KAMLET, M. J.; TAFT, R. W. The Solvatochromic Comparison Method. 2. The α -Scale of Solvent Hydrogen-Bond Donor (HBD) Acidities. **Journal of the American Chemical Society**, v. 98, n. 2, p. 377–383, 1976.

KAUR, H. *et al.* Critical analysis of research trends and issues in microwave assisted extraction of phenolics: have we really done enough. **Trends in Analytical Chemistry**, v. 85, p. 140–152, 2016.

KIM, J. I. Y. *et al.* Hydrotropic solubilization of poorly water-soluble drugs. **Journal of Pharmaceutical Sciences**, v. 99, p. 3953–3965, 2010.

KIRKWOOD, J. G.; BUFF, F. P. The statistical mechanical theory of solutions. I. **The**

- Journal of Chemical Physics**, v. 19, n. 6, p. 774–777, 1951.
- KLAMT, A. **COSMO-RS: From Quantum Chemistry to Fluid Phase Thermodynamics and Drug Design**. 1st. ed. [s.l.] Elsevier Science, 2005.
- KOŞAR, M.; DORMAN, H. J. .; HILTUNEN, R. Effect of an acid treatment on the phytochemical and antioxidant characteristics of extracts from selected Lamiaceae species. **Food Chemistry**, v. 91, p. 525–533, 2005.
- KUNZ, W.; HOLMBERG, K.; ZEMB, T. Hydrotropes. **Current Opinion in Colloid & Interface Science**, v. 22, p. 99–107, 2016.
- KUNZ, W.; NOSTRO, P. LO; NINHAM, B. W. The present state of affairs with Hofmeister effects. **Current Opinion in Colloid and Interface Science**, v. 9, p. 1–18, 2004.
- KUSUMOTO, Y. *et al.* **Preferential solvation studied by the fluorescence spectrum of pyrene in water-alcohol binary mixtures** **Chemistry Letters**, 1997.
- LAI, K.-Y. (ED.). **Liquid Detergents**. [s.l.] CRC Press, 2005.
- LAWRENCE, A. S. . The mechanism of detergence. **Nature**, v. 103, p. 1491–1494, 1959.
- LEAL-DUASO, A. *et al.* Glycerol as a source of designer solvents: physicochemical properties of low melting mixtures containing glycerol ethers and ammonium salts. **Phys. Chem. Chem. Phys.**, v. 19, n. 41, p. 28302–28312, 2017a.
- LEAL-DUASO, A. *et al.* Synthesis of 3-alkoxypropan-1,2-diols from glycidol: Experimental and theoretical studies for the optimization of the synthesis of glycerol derived solvents. **Green Chemistry**, v. 19, n. 17, p. 4176–4185, 2017b.
- LEAL-DUASO, A. *et al.* Glycerol-Derived Solvents: Synthesis and Properties of Symmetric Glyceryl Diethers. **ACS Sustainable Chemistry & Engineering**, v. 7, n. 15, p. 13004–13014, ago. 2019.
- LEBEUF, R. *et al.* Solvo-surfactant properties of dialkyl glycerol ethers: application as eco-friendly extractants of plant material through a novel Hydrotropic Cloud Point Extraction (HCPE) process. **ACS Sustainable Chemistry and Engineering**, v. 4, n. 9, p. 4815–4823, 2016.
- LEE, E. *et al.* The influence of alkane chain length on the skin irritation potential of 1,2-alkanediols. **International Journal of Cosmetic Science**, v. 33, n. 5, p. 421–425, 2011.
- LEE, J. *et al.* Hydrotropic solubilization of Paclitaxel: analysis of chemical structures for hydrotropic property. **Pharmaceutical Research**, v. 20, n. 7, p. 1022–1030, 2003.
- LEONETI, A. B. *et al.* Glycerol as a by-product of biodiesel production in Brazil :

- alternatives for the use of unrefined glycerol. **Renewable Energy**, v. 45, p. 138–145, 2012.
- LI, C. *et al.* Enhancement of clozapine solubility in three aqueous choline chloride-based deep eutectic solvents: Experimental and COSMO-RS prediction. **Journal of Molecular Liquids**, v. 359, p. 119298, 2022a.
- LI, J. *et al.* High-Pressure Technologies for the Recovery of Bioactive Molecules from Agro-Industrial Waste. **Applied Sciences (Switzerland)**, v. 12, n. 7, 2022b.
- LI, Q. S.; SU, M. G.; WANG, S. Densities and excess molar volumes for binary glycerol + 1-propanol, + 2-propanol, + 1,2-Propanediol, and + 1,3-propanediol mixtures at different temperatures. **Journal of Chemical and Engineering Data**, v. 52, n. 3, p. 1141–1145, 2007.
- LI, W. *et al.* Solubility modelling and thermodynamic properties of allopurinol in aqueous solutions of four deep eutectic solvents. **The Journal of Chemical Thermodynamics**, v. 132, p. 363–372, 2019.
- LIDIA, D.-A. *et al.* Microwave-Assisted Extraction of phytochemicals and other bioactive compounds. **Pure and Applied Biology**, v. 5, p. 48–57, 2017.
- LIU, F. *et al.* Catalytic etherification of glycerol with short chain alkyl alcohols in the presence of Lewis acids. **Green Chemistry**, v. 15, p. 901–909, 2014.
- LUKIĆ, M.; PANTELIĆ, I.; SAVIĆ, S. D. Towards optimal pH of the skin and topical formulations: From the current state of the art to tailored products. **Cosmetics**, v. 8, n. 3, 2021.
- MACHMUDAH, S. *et al.* Subcritical water extraction enhancement by adding deep eutectic solvent for extracting xanthone from mangosteen pericarps. **Journal of Supercritical Fluids**, v. 133, p. 615–624, 2018.
- MADALÃO, M. C. M. *et al.* Extraction of bioactive compounds from juçara pulp (*Euterpe edulis* M.) is affected by ultrasonic power and temperature. **Ciencia e Agrotecnologia**, v. 45, p. 1–11, 2021.
- MAUGERI, Z.; MARIA, P. D. Novel choline-chloride-based deep-eutectic-solvents with renewable hydrogen bond donors: levulinic acid and sugar-based polyols. **RSC Advances**, v. 2, p. 421–425, 2012.
- MAY, W. E. *et al.* Solution Thermodynamics of Some Slightly Soluble Hydrocarbons in Water. **Journal of Chemical and Engineering Data**, v. 28, n. 2, p. 197–200, 1983.
- MAZZUTTI, S. *et al.* Integrated green-based processes using supercritical CO₂ and

pressurized ethanol applied to recover antioxidant compounds from cocoa (*Theobroma cacao*) bean hulls. **The Journal of Supercritical Fluids**, v. 135, p. 52–59, 2018.

MCKEE, R. H. R. H. Use of hydrotropic solutions in industry. **Journal of Industrial and Engineering Chemistry**, v. 38, n. 4, p. 382–384, abr. 1946.

MOHSEN-NIA, M.; AMIRI, H.; JAZI, B. Dielectric constants of water, methanol, ethanol, butanol and acetone: Measurement and computational study. **Journal of Solution Chemistry**, v. 39, n. 5, p. 701–708, 2010.

MOITY, L. *et al.* Hydrotropic properties of alkyl and aryl glycerol monoethers. **Journal of Physical Chemistry B**, v. 117, n. 31, p. 9262–9272, 2013.

MOKHTARPOUR, M. *et al.* Study of naproxen in some aqueous solutions of choline-based deep eutectic solvents : Solubility measurements , volumetric and compressibility properties. **International Journal of Pharmaceutics**, v. 564, p. 197–206, 2019.

MOLINIER, V.; AUBRY, J.-M. Sugar-based hydrotropes: preparation, properties and applications. **Carbohydr. Chem.**, v. 40, p. 51–72, 2014.

MONTEALEGRE, R. R. *et al.* Phenolic compounds in skins and seeds of ten grape *Vitis vinifera* varieties grown in a warm climate. **Journal of Food Composition and Analysis**, v. 19, p. 687–693, 2006.

MOTA, F. L. *et al.* Aqueous Solubility of Some Natural Phenolic Compounds. **Industrial & Engineering Chemistry Research**, v. 47, n. 15, p. 5182–5189, ago. 2008.

MOTA, M. I. F. *et al.* Recovery of vanillin and syringaldehyde from lignin oxidation : a review of separation and purification processes. **Separation & Purification Reviews**, v. 45, n. 1–4, p. 227–259, 2016.

MUSTAFA, A.; TURNER, C. Pressurized liquid extraction as a green approach in food and herbal plants extraction: A review. **Analytica Chimica Acta**, v. 703, n. 1, p. 8–18, 2011.

NAIN, A. K. Densities and volumetric properties of binary mixtures of formamide with 1-butanol, 2-butanol, 1,3-butanediol and 1,4-butanediol at temperatures between 293.15 and 318.15 K. **Journal of Solution Chemistry**, v. 36, n. 4, p. 497–516, 2007.

NAKAJIMA, A. Solvent Effect on the Vibrational Structures of the Fluorescence and Absorption Spectra of Pyrene. **Bulletin of the Chemical Society of Japan**, v. 44, n. 12, p. 3272–3277, dez. 1971.

NANDI, L. G. *et al.* Properties of aqueous solutions of lentinan in the absence and presence of zwitterionic surfactants. **Carbohydrate Polymers**, v. 98, n. 1, p. 1–7, 2013.

NEUBERG, C. Hydrotropic phenomena. **Biochem. Z.**, v. 76, n. 1, p. 107–176, 1916.

NEUMANN, M. G. *et al.* The photophysical determination of the minimum hydrotrope concentration of aromatic hydrotropes. **Journal of Colloid and Interface Science**, v. 315, n. 2, p. 810–813, 2007.

NI, N.; YALKOWSKY, S. H. Prediction of Setschenow constants. **International Journal of Pharmaceutics**, v. 254, n. 2, p. 167–172, mar. 2003.

NIETO, A. *et al.* Pressurized liquid extraction: a useful technique to extract pharmaceuticals and personal-care products from sewage sludge. **Trends in Analytical Chemistry**, v. 29, p. 752–764, 2010.

NN, A. A Review on the extraction methods use in medicinal plants , principle, strength and limitation. **Medicinal & Aromatic Plants**, v. 4, p. 3–8, 2015.

NOUBIGH, A. *et al.* Solubility of some phenolic compounds in aqueous alkali metal nitrate solutions from (293.15 to 318.15) K. **Journal of Chemical Thermodynamics**, v. 40, n. 11, p. 1612–1616, 2008.

NOUBIGH, A. *et al.* Solubility of gallic acid in liquid mixtures of (ethanol + water) from (293.15 to 318.15) K. **J. Chem. Thermodynamics**, v. 55, p. 75–78, 2012.

NOUBIGH, A. *et al.* Measurement and correlation of the solubility of gallic acid in methanol plus water systems from (293.15 to 318.15) K. **Journal of Molecular Liquids**, v. 187, p. 226–229, 2013.

NOUBIGH, A.; AKERMI, A. Solubility and thermodynamic behavior of syringic acid in eight pure and water + methanol mixed solvents. **Journal of Chemical and Engineering Data**, v. 62, n. 10, p. 3274–3283, 2017.

OLIVAS-AGUIRRE, F. J. *et al.* Cyanidin-3-O-glucoside: Physical-chemistry, foodomics and health effects. **Molecules**, v. 21, n. 9, p. 1–30, 2016.

OROIAN, M.; ESCRICHE, I. Antioxidants: characterization, natural sources, extraction and analysis. **Food Research International**, v. 74, p. 10–36, 2015.

OZTURK, B.; GONZALEZ-MIQUEL, M. Alkanediol-based deep eutectic solvents for isolation of terpenoids from citrus essential oil: Experimental evaluation and COSMO-RS studies. **Separation and Purification Technology**, v. 227, n. June, p. 115707, 2019.

PAIVA, A. *et al.* Natural deep eutectic solvents – solvents for the 21st Century. **ACS Sustainable Chemistry & Engineering**, v. 2, p. 1063–1071, 2014.

PAL, M.; SINGH, R. K.; PANDEY, S. Evidence of Self-Aggregation of Cationic Surfactants in a Choline Chloride+Glycerol Deep Eutectic Solvent. **ChemPhysChem**, v.

16, n. 12, p. 2538–2542, 2015.

PALOMAR, J. *et al.* A COSMO-RS based guide to analyze/quantify the polarity of ionic liquids and their mixtures with organic cosolvents. **Physical Chemistry Chemical Physics**, v. 12, n. 8, p. 1991, 2010.

PANTELI, E.; VOUTSAS, E. Solubilities of cinnamic acid esters in binary mixtures of ionic liquids and organic solvents. **Fluid Phase Equilibria**, v. 295, n. 2, p. 208–214, 2010.

PARIENTE, S.; TANCHOUX, N.; FAJULA, F. Etherification of glycerol with ethanol over solid acid catalysts. **Green Chemistry**, v. 11, p. 1256–1261, 2008.

PARSA, J. B.; HAGHRO, M. F. Excess molar volume and viscosity deviation for binary mixtures of polyethylene glycol dimethyl ether 250 with 1,2-alkanediols (C3-C6) at T = (293.15 to 323.15) K. **Journal of Chemical Thermodynamics**, v. 40, n. 5, p. 782–788, 2008.

PASSOS, H.; FREIRE, M. G.; COUTINHO, J. A. P. Ionic liquid solutions as extractive solvents for value-added compounds from biomass. **Green Chemistry**, v. 16, p. 4786–4815, 2014.

PAUL, R.; CHATTARAJ, K. G.; PAUL, S. Role of Hydrotropes in Sparingly Soluble Drug Solubilization: Insight from a Molecular Dynamics Simulation and Experimental Perspectives. **Langmuir**, v. 37, n. 16, p. 4745–4762, abr. 2021.

PENA-PEREIRA, F.; NAMIEŚNIK, J. Ionic liquids and deep eutectic mixtures: Sustainable solvents for extraction processes. **ChemSusChem**, v. 7, n. 7, p. 1784–1800, 2014.

PERALES, E. *et al.* Ecotoxicity and QSAR studies of glycerol ethers in *Daphnia magna*. **Chemosphere**, v. 183, p. 277–285, set. 2017.

PERON, D. V.; FRAGA, S.; ANTELO, F. Thermal degradation kinetics of anthocyanins extracted from juçara (*Euterpe edulis* Martius) and “Italia” grapes (*Vitis vinifera* L.), and the effect of heating on the antioxidant capacity. **Food Chemistry**, v. 232, p. 836–840, 2017.

PINHO, S. P.; FERREIRA, O. Solubility of flavonoids in pure and mixed solvents.pdf. 2012.

PIRES, E. *et al.* Optimization of the synthesis of glycerol derived monoethers from glycidol by means of heterogeneous acid catalysis. **Molecules**, v. 23, p. 1–10, 2018.

PLAZA, M.; TURNER, C. Pressurized hot water extraction of bioactives. **TrAC -**

- Trends in Analytical Chemistry**, v. 71, p. 39–54, 2015.
- PRAUSNITZ, J. M.; LICHTENTHALER, R. N.; AZEVEDO, E. G. DE. **Molecular Thermodynamics of Fluid-Phase Equilibria**. 3rd. ed. [s.l.] Prentice Hall, 1998.
- QUEIMADA, A. *et al.* Solubilities of Biologically Active Phenolic Compounds: Measurements and Modeling Additions and corrections. **The Journal of Physical Chemistry B**, v. 113, n. 11, p. 3469–3476, 2009.
- QUESTE, S. *et al.* Short chain glycerol 1-monoethers - a new class of green solvo-surfactants. **Green Chemistry**, v. 8, p. 822–830, 2006a.
- QUESTE, S. *et al.* Short chain glycerol 1-monoethers—a new class of green solvo-surfactants. **Green Chem.**, v. 8, n. 9, p. 822–830, 2006b.
- QUESTE, S. *et al.* Thermophysical and bionotox properties of solvo-surfactants based on ethylene oxide, propylene oxide and glycerol. **Green Chemistry**, v. 9, p. 491–499, 2007.
- RAMBABU, K. *et al.* Date-fruit syrup waste extract as a natural additive for soap production with enhanced antioxidant and antibacterial activity. **Environmental Technology and Innovation**, v. 20, p. 101153, 2020.
- RE, R. *et al.* Antioxidant activity applying in improved ABTS radical cation decolorization assay. **Free Radical Biology & Medicine**, v. 26, p. 1231–1237, 1999.
- REICHARDT, C.; WELTON, T. Solute-Solvent Interactions. In: **Solvents and Solvent Effects in Organic Chemistry**. 4th. ed. Weinheim: Wiley-VCH, 2011a. p. 7–64.
- REICHARDT, C.; WELTON, T. Empirical Parameters of Solvent Polarity. In: **Solvents and Solvent Effects in Organic Chemistry**. 4th. ed. Weinheim: Wiley-VCH, 2011b. p. 425–508.
- REICHARDT, C.; WELTON, T. Solvent Effects on the Absorption Spectra of Organic Compounds. In: **Solvents and Solvent Effects in Organic Chemistry**. 4th. ed. Weinheim: Wiley-VCH, 2011c. p. 359–424.
- RENGSTL, D. *et al.* Effect of choline carboxylate ionic liquids on biological membranes. **Colloids Surf B Biointerfaces**, v. 1, n. 123, p. 575–581, 2014.
- RICCIARDI, A. M. *et al.* Glycidol, a valuable substrate for MAGEs synthesis: a simplified life cycle approach. **ChemSusChem**, v. 10, p. 2291–2300, 2017.
- ROCHA, M. A. A.; COUTINHO, J. A. P.; SANTOS, L. M. N. B. F. Vapor pressures of 1,3-dialkylimidazolium bis(trifluoromethylsulfonyl)imide ionic liquids with long alkyl chains. **The Journal of Chemical Physics**, v. 141, p. 134502, 2014.
- ROGERS, R. D.; SEDDON, K. R. Ionic liquids-solvents of the future? **Science**, v. 302,

p. 792–793, 2003.

ROSALES-CALDERON, O.; ARANTES, V. **A review on commercial-scale high-value products that can be produced alongside cellulosic ethanol.** [s.l.] BioMed Central, 2019. v. 12

RUESGAS-RAMÓN, M.; FIGUEROA-ESPINOZA, M. C.; DURAND, E. Application of Deep Eutectic Solvents (DES) for Phenolic Compounds Extraction: Overview, Challenges, and Opportunities. **Journal of Agricultural and Food Chemistry**, v. 65, n. 18, p. 3591–3601, 2017.

RUI, P. F. F.; ROCHA-SANTOS, T. A. P.; DUARTE, A. C. Supercritical fluid extraction of bioactive compounds. **Trends in Analytical Chemistry**, v. 76, p. 40–51, 2016.

SALES, I. *et al.* Enhancing Artemisinin Solubility in Aqueous Solutions: Searching for Hydrotropes based on Ionic Liquids. **Fluid Phase Equilibria**, v. 534, 2021.

SÁNCHEZ-CAMARGO, A. P. *et al.* New approaches for the selective extraction of bioactive compounds employing bio-based solvents and pressurized green processes. **The Journal of Supercritical Fluids**, v. 128, p. 112–120, 2017.

SANGHVI, R.; EVANS, D.; YALKOWSKY, S. H. Stacking complexation by nicotinamide: A useful way of enhancing drug solubility. **International Journal of Pharmaceutics**, v. 336, n. 1, p. 35–41, maio 2007.

SANTOS, P. H. *et al.* Extraction of bioactive compounds from feijoa (*Acca sellowiana* (O . Berg) Burret) peel by low and high-pressure techniques. **The Journal of Supercritical Fluids**, v. 145, p. 219–227, 2019.

SARKIC, A.; STAPPEN, I. Essential oils and their single compounds in cosmetics-a critical review. **Cosmetics**, v. 5, n. 1, p. 1–21, 2018.

SAVJANI, K. T.; GAJJAR, A. K.; SAVJANI, J. K. Drug Solubility: Importance and Enhancement Techniques. **ISRN Pharmaceutics**, v. 2012, n. 100 mL, p. 1–10, 2012.

SAXENA, R. K. *et al.* Microbial production and applications of 1,2-propanediol. **Indian Journal of Microbiology**, v. 50, n. 1, p. 2–11, 2010.

SCHULZ, M. *et al.* Juçara fruit (*Euterpe edulis* Mart.): sustainable exploitation of a source of bioactive compounds. **Food Research International**, v. 89, p. 14–26, 2016.

SETSCHENOW, J. Über die Konstitution der Salzlösungen auf Grund ihres Verhaltens zu Kohlensäure. **Phys. Chem.**, v. 4U, p. 117–125, 1989.

SHA, J. *et al.* Solubility determination, model evaluation, Hansen solubility parameter and thermodynamic properties of benorilate in six pure solvents and two binary solvent

mixtures. **Journal of Chemical Thermodynamics**, v. 153, 2021.

SHAKEEL, F. *et al.* Solubility and thermodynamic function of a bioactive compound bergenin in various pharmaceutically acceptable neat solvents at different temperatures. **Journal of Chemical Thermodynamics**, v. 101, p. 19–24, 2016.

SHEKAARI, H. *et al.* Effect of choline chloride/ethylene glycol or glycerol as deep eutectic solvents on the solubility and thermodynamic properties of acetaminophen. **Journal of Molecular Liquids**, v. 249, p. 1222–1235, 2018.

SHI, X. *et al.* Density, Viscosity, and Excess Properties of Binary Mixtures of 2-(Methylamino)ethanol with 2-Methoxyethanol, 2-Ethoxyethanol, and 2-Butoxyethanol from 293.15 to 353.15 K. **Journal of Chemical and Engineering Data**, v. 64, n. 9, p. 3960–3970, 2019.

SHIMIZU, S. Formulating rationally via statistical thermodynamics. **Current Opinion in Colloid & Interface Science**, v. 48, p. 53–64, ago. 2020.

SHIMIZU, S.; BOOTH, J. J.; ABBOTT, S. Hydrotropy: binding models vs. statistical thermodynamics. **Physical Chemistry Chemical Physics**, v. 15, n. 47, p. 20625–20632, 2013.

SHIMIZU, S.; MATUBAYASI, N. Hydrotropy: monomer-micelle equilibrium and minimum hydrotrope concentration. **Journal of Physical Chemistry B**, v. 118, n. 35, p. 10515–10524, 2014.

SHIMIZU, S.; MATUBAYASI, N. The origin of cooperative solubilisation by hydrotropes. **Phys. Chem. Chem. Phys.**, v. 18, n. 36, p. 25621–25628, 2016.

SHIMIZU, S.; MCLAREN, W. M.; MATUBAYASI, N. The Hofmeister series and protein-salt interactions. **The Journal of Chemical Physics**, v. 124, p. 5–8, 2006.

SHIRSATH, S. R.; SONAWANE, S. H.; GOGATE, P. R. Intensification of extraction of natural products using ultrasonic irradiations - a review of current status. **Chemical Engineering & Processing**, v. 53, p. 10–23, 2012.

SHULGIN, I.; RUCKENSTEIN, E. Kirkwood–Buff integrals in aqueous alcohol systems: aggregation, correlation volume, and local composition. **The Journal of Physical Chemistry B**, v. 103, n. 2, p. 872–877, 1999.

SILVA, R. P. F. F. DA; ROCHA-SANTOS, T. A. P.; DUARTE, A. C. Supercritical fluid extraction of bioactive compounds. **Trends in Analytical Chemistry journal**, v. 76, p. 40–51, 2016.

SINTRA, E. *et al.* Cholinium-based ionic liquids as bioinspired hydrotropes to tackle

solubility challenges in drug formulation. **European Journal of Pharmaceutics and Biopharmaceutics**, v. 164, p. 86–92, 2021.

SINTRA, T. E. *et al.* Enhanced dissolution of ibuprofen using ionic liquids as catanionic hydrotropes. **Physical Chemistry Chemical Physics**, v. 20, n. 3, p. 2094–2103, 2018.

SOARES, B. *et al.* Enhanced solubility of lignin monomeric model compounds and technical lignins in aqueous solutions of deep eutectic solvents. **ACS Sustainable Chemistry and Engineering**, v. 5, n. 5, p. 4056–4065, 2017.

SOARES, B. *et al.* Hydrotropy and Cosolvency in Lignin Solubilization with Deep Eutectic Solvents. **ACS Sustainable Chemistry and Engineering**, v. 7, n. 14, p. 12485–12493, 2019.

SOARES, B. P. *et al.* Glycerol Ethers as Hydrotropes and Their Use to Enhance the Solubility of Phenolic Acids in Water. **ACS Sustainable Chemistry and Engineering**, v. 8, n. 14, p. 5742–5749, 2020.

SOLOMONOV, B. N.; SEDOV, I. A. Quantitative description of the hydrophobic effect: The enthalpic contribution. **Journal of Physical Chemistry B**, v. 110, n. 18, p. 9298–9303, 2006.

SONG, Y. *et al.* Comparing a deep eutectic solvent (DES) to a hydrotrope for their ability to enhance the fractionation and enzymatic hydrolysis of willow and corn stover. **Sustainable Energy & Fuels**, v. 3, n. 5, p. 1329–1337, 2019.

SRINIVAS, V.; BALASUBRAMANIAN, D. When does the switch from hydrotropy to micellar behavior occur? **Langmuir**, v. 14, n. 23, p. 6658–6661, 1998.

SRINIVASULU, C. *et al.* Syringic acid (SA) – A Review of Its Occurrence, Biosynthesis, Pharmacological and Industrial Importance. **Biomedicine & Pharmacotherapy**, v. 108, p. 547–557, dez. 2018.

STANTON, K. *et al.* Environmental risk assessment of hydrotropes in the United States, Europe, and Australia. **Integrated Environmental Assessment and Management**, v. 6, n. 1, p. 155–163, 2009.

STARK, A. *et al.* The effect of hydrogen bond acceptor properties of ionic liquids on their cellulose solubility. **Science China Chemistry**, v. 55, n. 8, p. 1663–1670, ago. 2012.

SUBBARAO, C. V. *et al.* Functions of hydrotropes in solutions. **Chemical Engineering and Technology**, v. 35, n. 2, p. 225–237, 2012.

SUBRAMANIAN, D. *et al.* Mesoscale inhomogeneities in aqueous solutions of 3-methylpyridine and tertiary butyl alcohol. **Journal of Chemical & Engineering Data**, v.

56, p. 1238–1248, 2011.

SUNGPUD, C. *et al.* Ultrasonic-assisted virgin coconut oil based extraction for maximizing polyphenol recovery and bioactivities of mangosteen peels. **Journal of Food Science and Technology**, v. 57, n. 11, p. 4032–4043, 2020.

SUTTER, M. *et al.* Glycerol ether synthesis: a bench test for Green Chemistry concepts and technologies. **ACS Sustainable Chemistry & Engineering**, v. 115, p. 8609–8651, 2015.

SVÄRD, M.; GRACIN, S.; RASMUSON, Å. C. Oiling out or molten hydrate—liquid–liquid phase separation in the system vanillin–water. **Journal of Pharmaceutical Sciences**, v. 96, n. 9, p. 2390–2398, set. 2007.

THAKKER, M. R.; PARIKH, J. K.; DESAI, M. A. Ultrasound Assisted Hydrotropic Extraction: a greener approach for the isolation of geraniol from the leaves of *Cymbopogon martinii*. **ACS Sustainable Chemistry & Engineering**, v. 6, p. 3215–3224, 2018.

TOMMASI, E. *et al.* Enhanced and selective lipid extraction from the microalga *P. tricornutum* by dimethyl carbonate and supercritical CO₂ using deep eutectic solvents and microwaves as pretreatment. **ACS Sustainable Chemistry and Engineering**, v. 5, n. 9, p. 8316–8322, 2017.

TORSKANGER POLL, K.; ANDERSEN, Ø. M. Colour stability of anthocyanins in aqueous solutions at various pH values. **Food Chemistry**, v. 89, n. 3, p. 427–440, 2005.

TREVISAN, A. C. D. *et al.* Market for amazonian açai (*Euterpe oleraceae*) stimulates pulp production from Atlantic Forest juçara berries (*Euterpe edulis*). **Agroecology and Sustainable Food Systems**, v. 39, p. 762–781, 2015.

TURNER, C. Overview of Modern Extraction Techniques for Food and Agricultural Samples. In: **Modern Extraction Techniques**. [s.l: s.n.]. p. 3–19.

UMNAHANANT, P. *et al.* Vaporization Enthalpies of the α,ω -Alkanediols by Correlation Gas Chromatography. **Journal of Chemical & Engineering Data**, v. 51, n. 6, p. 2246–2254, nov. 2006.

VASILESCU, M. *et al.* Fluorescence probe study of the interactions between nonionic poly(oxyethylenic) surfactants and poly(acrylic acid) in aqueous solutions. **Langmuir**, v. 13, n. 26, p. 6951–6955, 1997.

VAZQUEZ-ROIG, P.; PICÓ, Y. Pressurized liquid extraction of organic contaminants in environmental and food samples. **Trends in Analytical Chemistry**, v. 71, p. 55–64,

2015.

VÁZQUEZ, G. *et al.* Response surface optimization of antioxidants extraction from chestnut (*Castanea sativa*) bur. **Industrial Crops & Products**, v. 35, p. 126–134, 2012.

VEIGA, P. M. *et al.* Acid zeolites for glycerol etherification with ethyl alcohol: Catalytic activity and catalyst properties. **Applied Catalysis A, General**, v. 548, p. 2–15, 2017.

VELEVA, V. R.; CUE, B. W.; TODOROVA, S. Benchmarking Green Chemistry Adoption by the Global Pharmaceutical Supply Chain. **ACS Sustainable Chemistry & Engineering**, v. 6, n. 1, p. 2–14, jan. 2018.

VEREVKIN, S. P. Determination of vapor pressures and enthalpies of vaporization of 1,2-alkanediols. **Fluid Phase Equilibria**, v. 224, n. 1, p. 23–29, set. 2004.

VIEIRA, G. S. *et al.* Chemical and economic evaluation of natural antioxidant extracts obtained by ultrasound-assisted and agitated bed extraction from jussara pulp (*Euterpe edulis*). **Journal of Food Engineering**, v. 119, n. 2, p. 196–204, 2013.

VIEIRA, G. S. *et al.* Determination of anthocyanins and non-anthocyanin polyphenols by ultra performance liquid chromatography/electrospray ionization mass spectrometry (UPLC/ESI–MS) in jussara (*Euterpe edulis*) extracts. **Journal of Food Science and Technology**, v. 54, p. 2135–2144, 2017.

VIEIRA, V. *et al.* Insights on the extraction performance of alkanediols and glycerol: using *Juglans regia* L. leaves as a source of bioactive compounds. **Molecules**, v. 25, n. 11, p. 2497, 2020.

VILAS-BOAS, S. M. *et al.* Solvent and temperature effects on the solubility of syringic, vanillic or veratric acids: Experimental, modeling and solid phase studies. **Journal of Molecular Liquids**, v. 289, 2019.

VINATORU, M. An overview of the ultrasonically assisted extraction of bioactive principles from herbs. **Ultrasonics Sonochemistry**, v. 8, n. 3, p. 303–313, jul. 2001.

VINATORU, M.; MASON, T. J.; CALINESCU, I. Ultrasonically assisted extraction (UAE) and microwave assisted extraction (MAE) of functional compounds from plant materials. **TrAC - Trends in Analytical Chemistry**, v. 97, p. 159–178, 2017.

WAGLE, V. B.; KOTHARI, P. S.; GAIKAR, V. G. Effect of temperature on aggregation behavior of aqueous solutions of sodium cumene sulfonate. **Journal of Molecular Liquids**, v. 133, n. 1–3, p. 68–76, 2007.

WAKISAKA, A.; KOMATSU, S.; USUI, Y. Solute-solvent and solvent-solvent

interactions evaluated through clusters isolated from solutions: Preferential solvation in water-alcohol mixtures. **Journal of Molecular Liquids**, v. 90, n. 1–3, p. 175–184, 2001.

WAN MAHMOOD, W. M. A. *et al.* Polyol-Based Deep Eutectic Solvents for Extraction of Natural Polyphenolic Antioxidants from *Chlorella vulgaris*. **ACS Sustainable Chemistry and Engineering**, v. 7, n. 5, p. 5018–5026, 2019.

WANG, F.; POTTEL, R.; KAATZE, U. Dielectric properties of alkanediol/water mixtures. **Journal of Physical Chemistry B**, v. 101, n. 6, p. 922–929, 1997.

WANG, H. *et al.* Recent advances in microwave-assisted extraction of trace organic pollutants from food and environmental samples. **Trends in Analytical Chemistry**, v. 75, p. 197–208, 2016.

WANG, H. *et al.* Insights into the Hydrogen Bond Interactions in Deep Eutectic Solvents Composed of Choline Chloride and Polyols. **ACS Sustainable Chemistry and Engineering**, v. 7, n. 8, p. 7760–7767, 2019.

WEN, L. *et al.* Combination of emerging technologies for the extraction of bioactive compounds. **Critical Reviews in Food Science and Nutrition**, v. 60, n. 11, p. 1826–1841, 2020.

WOJEICCHOWSKI, J. P. *et al.* Extraction of phenolic compounds from rosemary using choline chloride – based Deep Eutectic Solvents. **Separation and Purification Technology**, v. 258, p. 117975, 2021.

WOLFSON, A. *et al.* Glycerol derivatives as green reaction mediums. **Green Chemistry Letters and Reviews**, v. 5, n. 1, p. 7–12, 2012.

XU, S. *et al.* Review of Liquid-Liquid Phase Separation in Crystallization: From Fundamentals to Application. **Crystal Growth and Design**, v. 21, n. 12, p. 7306–7325, 2021.

YALKOWSKY, S. H.; HE, Y.; JAIN, P. **Handbook of Aqueous Solubility Data**. [s.l.] CRC Press, 2016. v. 44

YANG, H.; RASMUSON, Å. C. Ternary phase diagrams of ethyl paraben and propyl paraben in ethanol aqueous solvents. **Fluid Phase Equilibria**, v. 376, p. 69–75, 2014.

YIN, T.; CHEN, Y.; SHEN, W. Aggregation of an ionic-liquid type hydrotrope 1-Butyl-3-methylimidazolium p-toluenesulfonate in aqueous solution. **Colloids and Surfaces A: Physicochemical and Engineering Aspects**, v. 564, n. December 2018, p. 95–100, 2019.

YOO, I. K.; KIM, J. IL; KANG, Y. K. Conformational preferences and antimicrobial activities of alkanediols. **Computational and Theoretical Chemistry**, v. 1064, p. 15–

24, jul. 2015.

ZACHOVÁ, Z. *et al.* Combining high-pressure methods for extraction of stilbenes from grape cane. **The Journal of Supercritical Fluids**, v. 142, p. 38–44, 2018.

ZAINAL-ABIDIN, M. H. *et al.* New horizons in the extraction of bioactive compounds using deep eutectic solvents: a review. **Analytica Chimica Acta**, v. 979, p. 1–23, 2017.

ZANA, R.; ELJEBARI, M. J. Fluorescence probing investigation of the self-association of alcohols in aqueous solution. **Journal of Physical Chemistry**, v. 97, n. 42, p. 11134–11136, 1993.

ZEMB, T. N. *et al.* How to explain microemulsions formed by solvent mixtures without conventional surfactants. **Proceedings of the National Academy of Sciences**, v. 13, n. 16, p. 4260–4265, 2016.

ZHANG, H. *et al.* Insight into solid-liquid and liquid-liquid phase equilibrium behavior of vanillin and ethyl vanillin. **Journal of Molecular Liquids**, v. 365, p. 120059, 2022.

ZHANG, H.; YANG, X.; WANG, Y. Microwave assisted extraction of secondary metabolites from plants: current status and future directions. **Trends in Food Science & Technology**, v. 22, n. 12, p. 672–688, 2011.

ZHANG, Y. *et al.* Measurement and Correlation of the Solubility of Vanillic Acid in Eight Pure and Water + Ethanol Mixed Solvents at Temperatures from (293.15 to 323.15) K. **Journal of Chemical and Engineering Data**, v. 61, n. 1, p. 420–429, 2016.

ZHAO, H. *et al.* Solution crystallization of vanillin in the presence of a liquid-liquid phase separation. **Industrial and Engineering Chemistry Research**, v. 51, n. 45, p. 14646–14652, 2012.

APPENDIX A

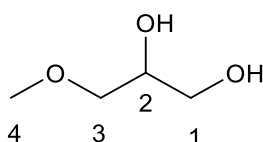
S1. PRODUCT CHARACTERIZATION

The water content of the synthesised monoalkyl glycerol ethers was measured using a Metrohm 831 Karl Fisher coulometer (results reported in Table S1).

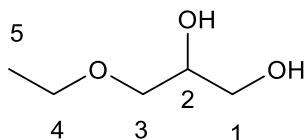
Table S1. Water content (mass percentage) and standard deviation (s) of the glycerol ethers synthesised in this work.

Substance	wt % H ₂ O $\pm$ s
[1.0.0]	1.1884 $\pm$ 0.0061
[2.0.0]	0.6230 $\pm$ 0.0030
[3.0.0]	0.6133 $\pm$ 0.0254
[4.0.0]	0.8893 $\pm$ 0.0092
[5.0.0]	0.3026 $\pm$ 0.0010
[6.0.0]	0.2139 $\pm$ 0.0163
[1.0.1]	0.0786 $\pm$ 0.0021
[2.0.2]	0.0468 $\pm$ 0.0017

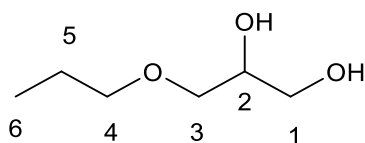
Gas chromatography was carried out in a Hewlett Packard 7890 series II Gas Chromatograph using a column of phenyl silicone 5.5% (Zebron Inferno 30 m x 0.25 mm x 0.25 μ m) and Helium as carrier gas. The conditions and method used in the equipment were: 553.2 K injector temperature, 523.2 K detector temperature. Temperature program: 80 °C for 3 min., temperature gradient 5 K min⁻¹ until 383.2 K, temperature gradient 20 K min⁻¹ until 503.2 K, 503.2 K isotherm for 5 min. The ¹H- and ¹³C-NMR spectra ([d₆]DMSO, δ ppm, *J* Hz) were obtained using a Bruker AV-400 instrument with tetramethylsilane (TMS) as standard.



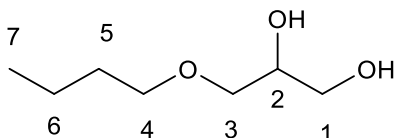
3-Methoxypropan-1,2-diol [1.0.0]: ¹H RMN (400 MHz, [d₆]DMSO): δ 4.61 (d, 1H, *J* = 5.1 Hz, OH₂), 4.46 (t, 1H, *J* = 5.7 Hz, OH₁), 3.56 (sext, 1H, *J* = 5.1 Hz, H₂), 3.18-3.36 (m, 4H, H₁, H₃), 3.23 (s, 3H, H₄). ¹³C RMN (100 MHz, [d₆]DMSO): δ 74.2 (CH₂, C₃), 70.3 (CH, C₂), 62.9 (CH₂, C₁), 58.3 (CH₃, C₄).



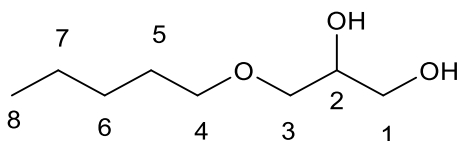
3-ethoxypropan-1,2-diol [2.0.0]: ¹H RMN (400 MHz, [d₆]DMSO): δ 4.59 (d, 1H, *J* = 5.1 Hz, OH₂), 4.45 (t, 1H, *J* = 5.7 Hz, OH₁), 3.55 (sext, 1H, *J* = 5.3 Hz, H₂), 3.42 (q, 2H, *J* = 7.0 Hz, H₄), 3.21-3.37 (m, 4H, H₁, H₃), 1.10 (t, 3H, *J* = 7.0 Hz, H₅). ¹³C RMN (100 MHz, [d₆]DMSO): δ 72.3 (CH₂, C₃), 70.8 (CH, C₂), 66.0 (CH₂, C₄), 63.4 (CH₂, C₁), 15.4 (CH₃, C₅).



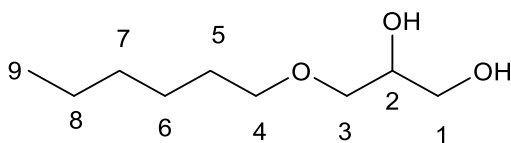
3-propoxypropan-1,2-diol [3.0.0]: ^1H RMN (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.57 (d, 1H, $J = 5.1$ Hz, OH_2), 4.44 (t, 1H, $J = 5.7$ Hz, OH_1), 3.55 (sext, 1H, $J = 5.3$ Hz, H_2), 3.22-3.38 (m, 6H, H_1 , H_3H_4), 1.48 (sext, 2H, $J = 6.7$ Hz, H_5), 0.85 (t, 3H, $J = 7.4$ Hz, H_6). ^{13}C RMN (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 72.2 (CH_2 , C_3) 72.1 (CH_2 , C_4), 70.5 (CH , C_2), 63.2 (CH_2 , C_1), 22.4 (CH_2 , C_5), 10.5 (CH_3 , C_6).



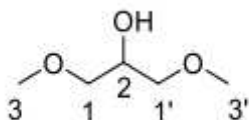
3-butoxypropan-1,2-diol [4.0.0]: ^1H RMN (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.54 (d, 1H, $J = 5.1$ Hz, OH_2), 4.41 (t, 1H, $J = 5.7$ Hz, OH_1), 3.53 (sext, 1H, $J = 5.2$ Hz, H_2), 3.34 (t, 2H, $J = 6.7$ Hz, H_4), 3.19-3.32 (m, 4H, H_1 , H_3), 1.44 (quint, 2H, $J = 7.6$ Hz, H_5), 1.29 (sext, 2H, $J = 7.3$ Hz, H_6), 0.85 (t, 3H, $J = 7.4$ Hz, H_7). ^{13}C RMN (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 72.3 (CH_2 , C_3), 70.6 (CH , C_2), 70.2 (CH_2 , C_4), 63.2 (CH_2 , C_1), 31.4 (CH_2 , C_5), 18.9 (CH_2 , C_6), 13.8 (CH_3 , C_7).



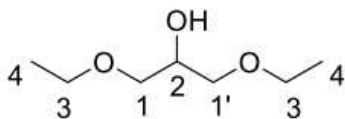
3-pentiloxopropan-1,2-diol [5.0.0]: ^1H RMN (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.56 (d, 1H, $J = 5.1$ Hz, OH_2), 4.43 (t, 1H, $J = 5.7$ Hz, OH_1), 3.55 (sext, 1H, $J = 5.3$ Hz, H_2), 3.35 (t, 2H, $J = 6.6$ Hz, H_4), 3.25-3.37 (m, 3H, H_1 , H_{3a}), 3.24 (dd, 1H, $J = 9.8$ Hz, $J = 6.0$ Hz, H_{3b}), 1.48 (quint, 2H, $J = 7.0$ Hz, H_5), 1.22-1.32 (m, 4H, H_6 , H_7), 0.86 (t, 3H, $J = 7.0$ Hz, H_8). ^{13}C RMN (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 72.3 (CH_2 , C_3), 70.6 (CH , C_2), 70.5 (CH_2 , C_4), 63.2 (CH_2 , C_1), 28.9 (CH_2 , C_5), 27.9 (CH_2 , C_6), 22.0 (CH_2 , C_7), 13.9 (CH_3 , C_8).



3-hexyloxypropan-1,2-diol [6.0.0]: ^1H RMN (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.56 (d, 1H, $J = 5.0$ Hz, OH_2), 4.43 (t, 1H, $J = 5.7$ Hz, OH_1), 3.55 (sext, 1H, $J = 5.3$ Hz, H_2), 3.36 (t, 2H, $J = 6.6$ Hz, H_4), 3.22-3.36 (m, 3H, H_1 , H_{3a}), 3.24 (dd, 1H, $J = 9.8$ Hz, $J = 6.0$ Hz, H_{3b}), 1.43-1.52 (m, 2H, H_5), 1.20-1.33 (m, 6H, H_6 , H_7 , H_8), 0.85 (t, 3H, $J = 7.0$ Hz, H_9). ^{13}C RMN (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 77.3 (CH_2 , C_3), 70.6 (CH , C_2), 70.6 (CH_2 , C_4), 63.2 (CH_2 , C_1), 31.2 (CH_2 , C_5), 29.3 (CH_2 , C_6), 25.4 (CH_2 , C_7), 22.1 (CH_2 , C_8), 13.9 (CH_3 , C_9).



1,3-dimethoxypropan-2-ol [1.0.1]: ^1H NMR (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.79 (d, 1H, $J = 5.2$ Hz, OH_2), 3.70 (sext, 1H, $J = 5.1$ Hz, H_2), 3.28 (dd, 2H, $J = 9.8$ Hz, $J = 5.0$ Hz, H_{1a} , $\text{H}_{1'a}$), 3.23 (dd, 2H, $J = 9.9$ Hz, $J = 5.6$ Hz, H_{1b} , $\text{H}_{1'b}$), 3.24 (s, 6H, H_3 , $\text{H}_{3'}$). ^{13}C NMR (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 74.2 (CH_2 , C_1), 68.2 (CH , C_2), 58.4 (CH_3 , C_3).



1,3-diethoxypropan-2-ol [2.0.2]: ^1H NMR (400 MHz, $[\text{d}_6]\text{DMSO}$): δ 4.72 (d, 1H, $J = 5.2$ Hz, OH_2), 3.67 (sext, 1H, $J = 5.1$ Hz, H_2), 3.42 (c, 4H, $J = 7.0$ Hz, H_3), 3.32 (dd, 2H, $J = 9.8$ Hz, $J = 5.0$ Hz, H_{1a} , $\text{H}_{1'a}$), 3.26 (dd, 2H, $J = 9.9$ Hz, $J = 5.9$ Hz, H_{1b} , $\text{H}_{1'b}$), 1.09 (t, 6H, $J = 7.0$ Hz, H_4). ^{13}C NMR (100 MHz, $[\text{d}_6]\text{DMSO}$): δ 72.2 (CH_2 , C_1), 68.6 (CH , C_2), 65.8 (CH_2 , C_3), 15.1 (CH_3 , C_4).

Figure S1. ^1H -RMN and ^{13}C -RMN (APT) of 3-methoxypropan-1,2-diol [1.0.0].

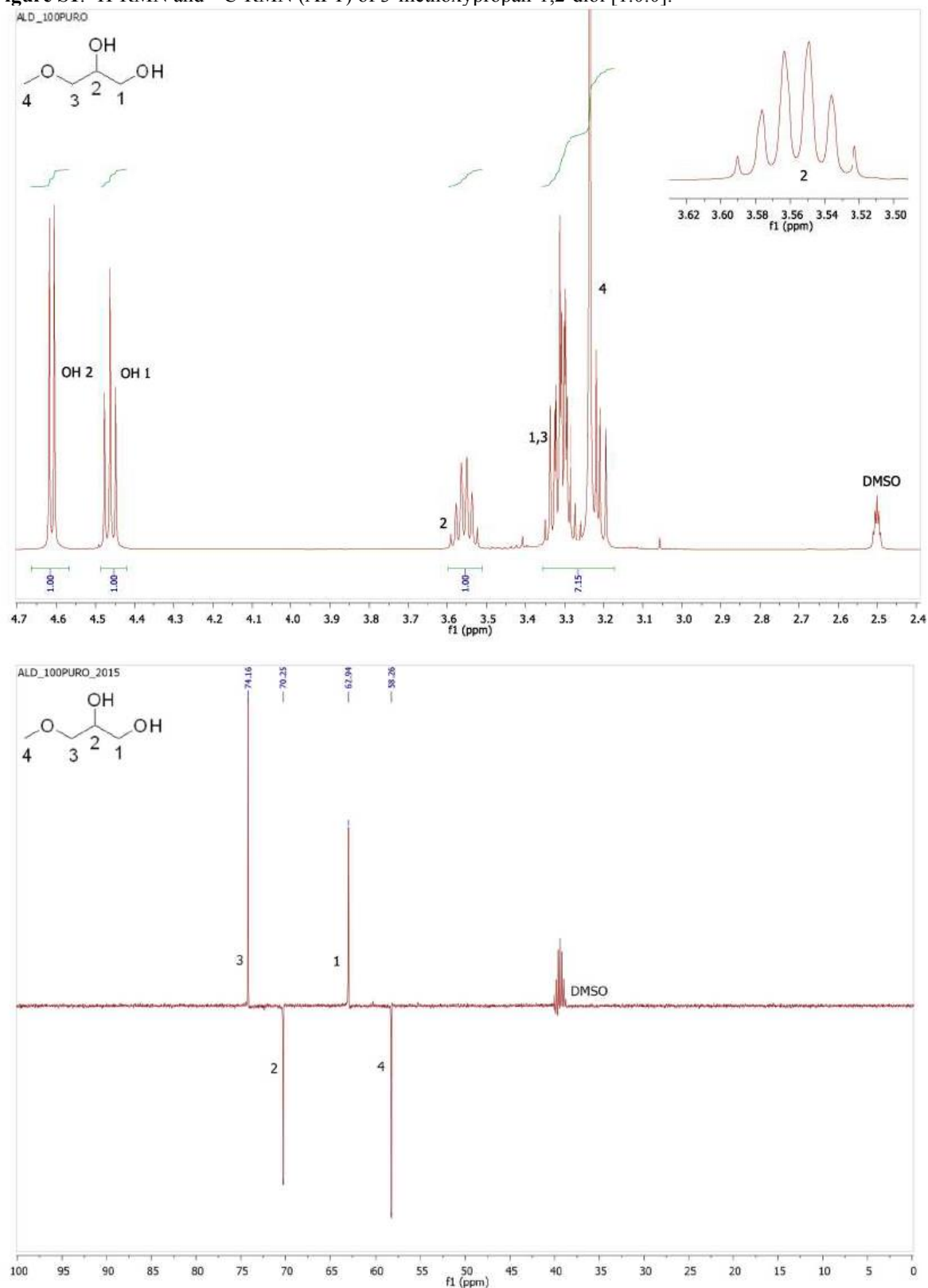


Figure S2. ^1H -RMN and ^{13}C -RMN (APT) of 3-ethoxypropan-1,2-diol [2.0.0].

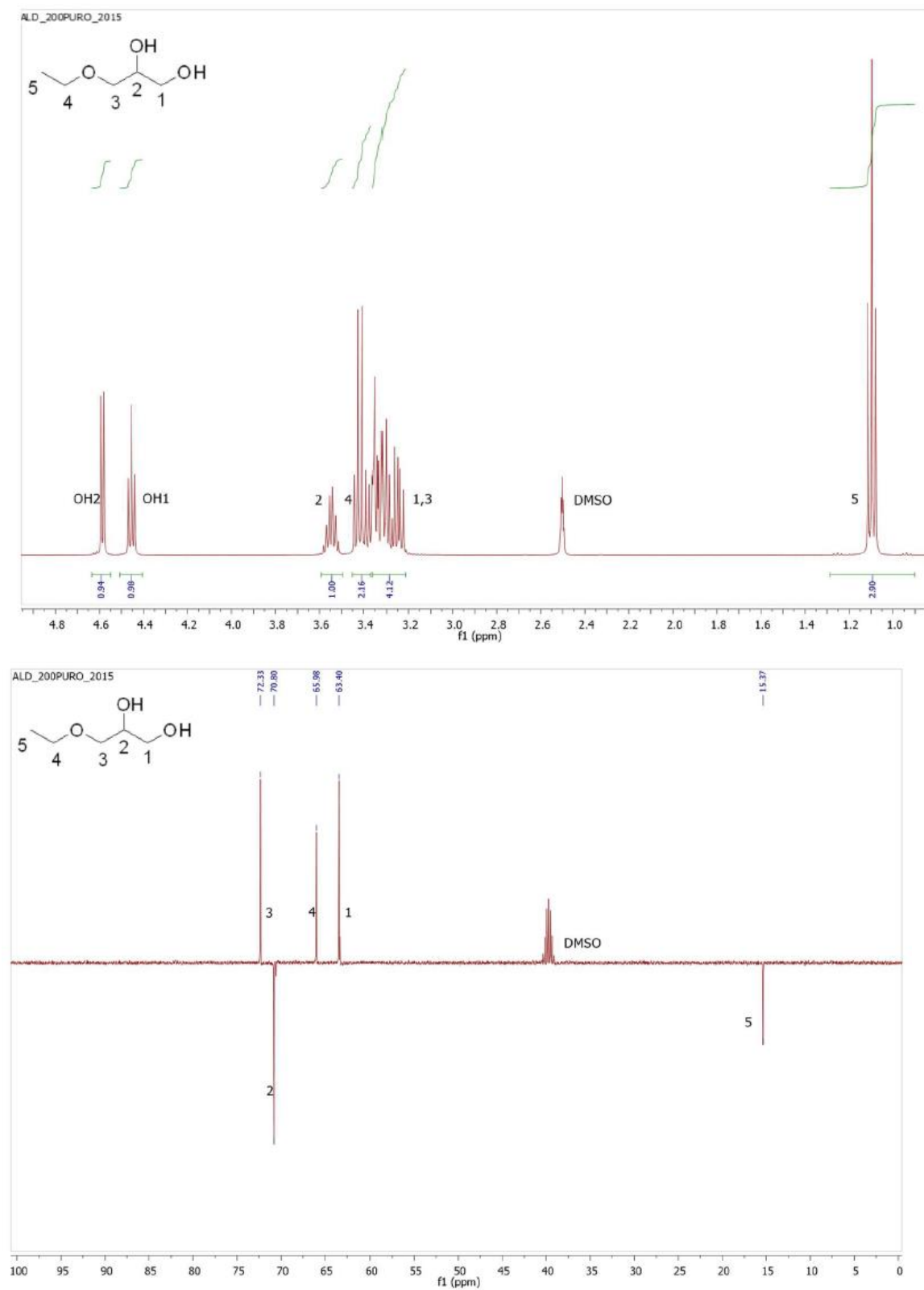


Figure S3. ^1H -RMN and ^{13}C -RMN (APT) of 3-propoxypropan-1,2-diol [3.0.0].

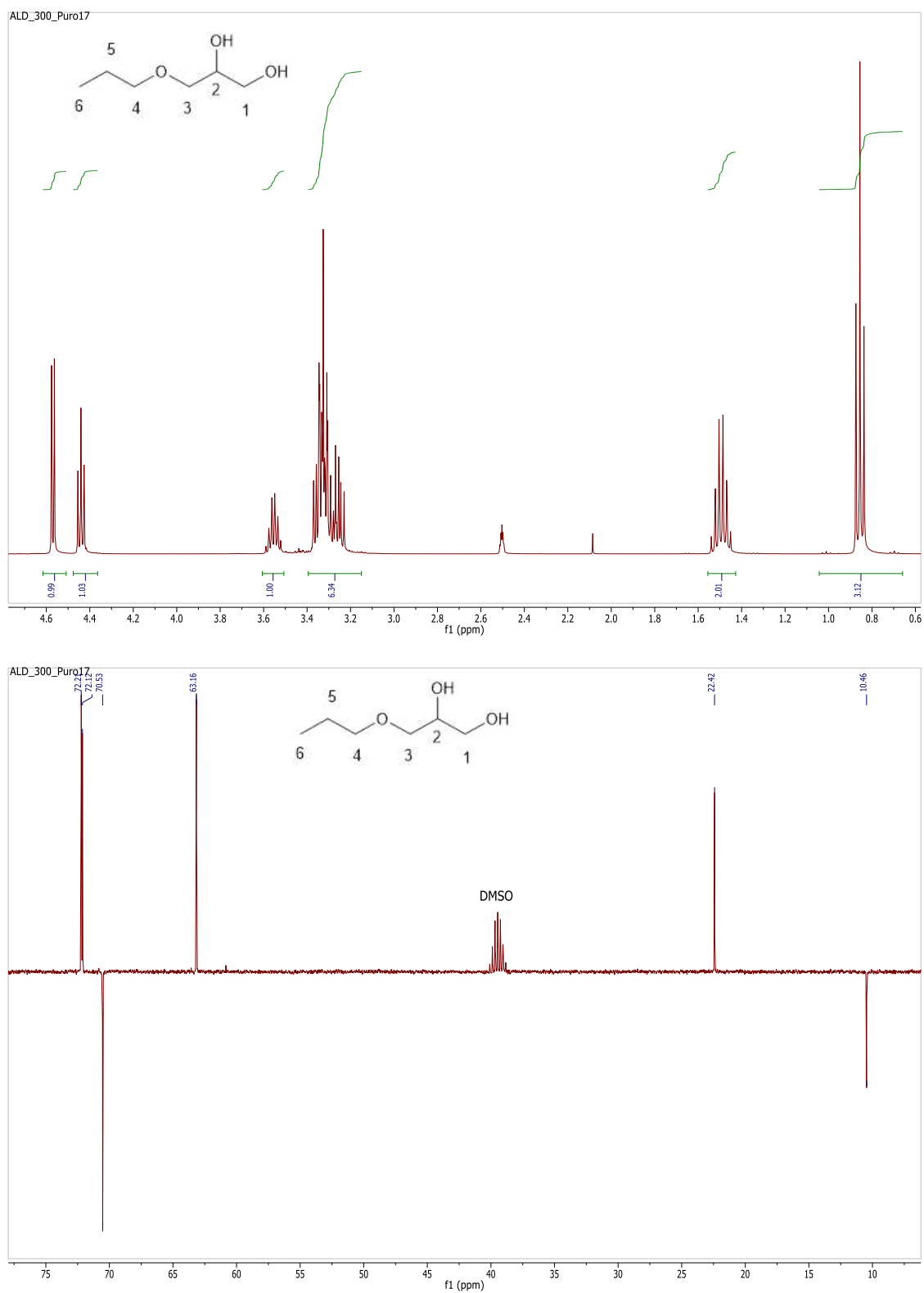


Figure S4. ^1H -RMN and ^{13}C -RMN (APT) of 3-butoxypropan-1,2-diol [4.0.0].

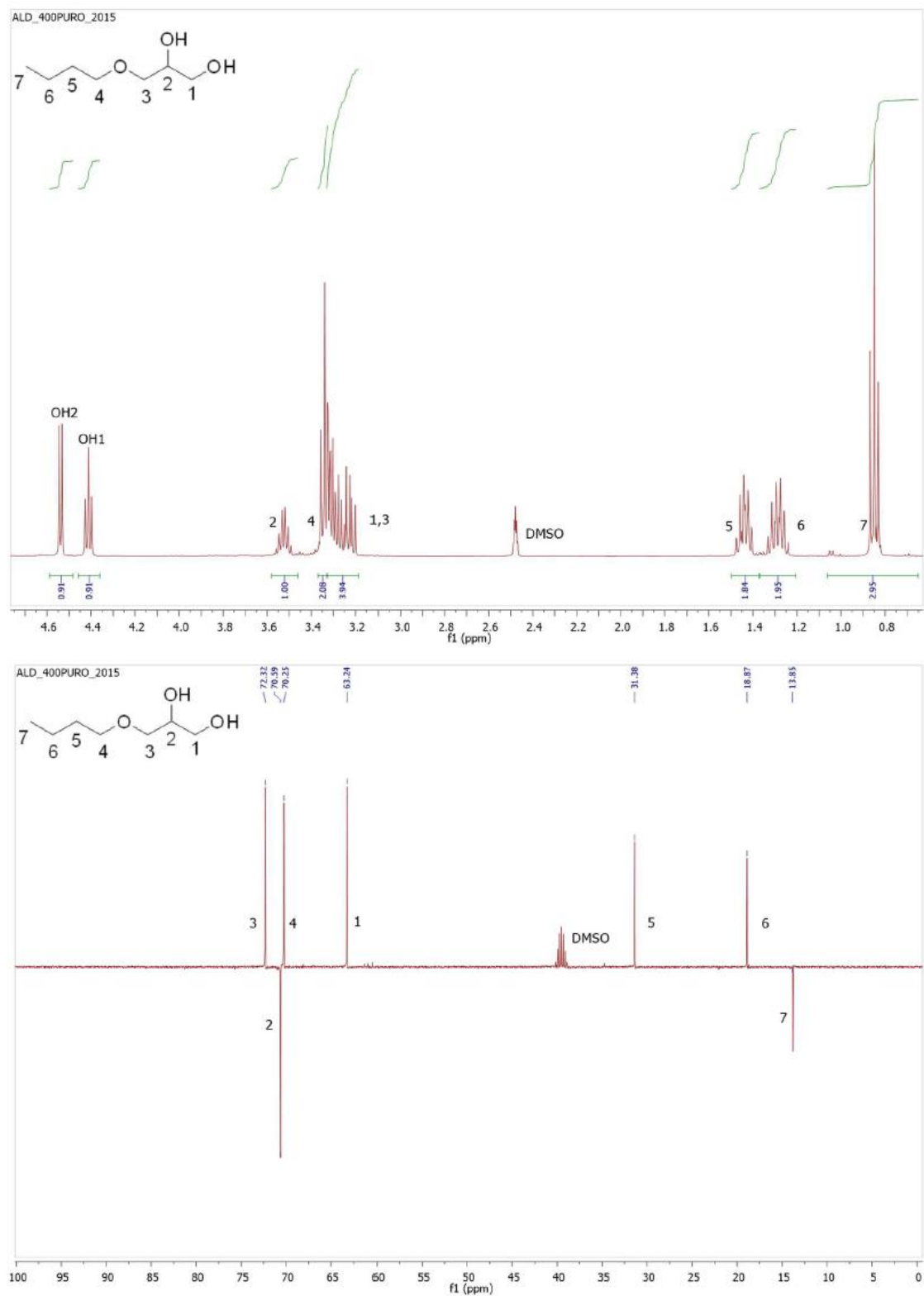


Figure S5. ^1H -RMN and ^{13}C -RMN (APT) of 3-pentoxypropan-1,2-diol [5.0.0].

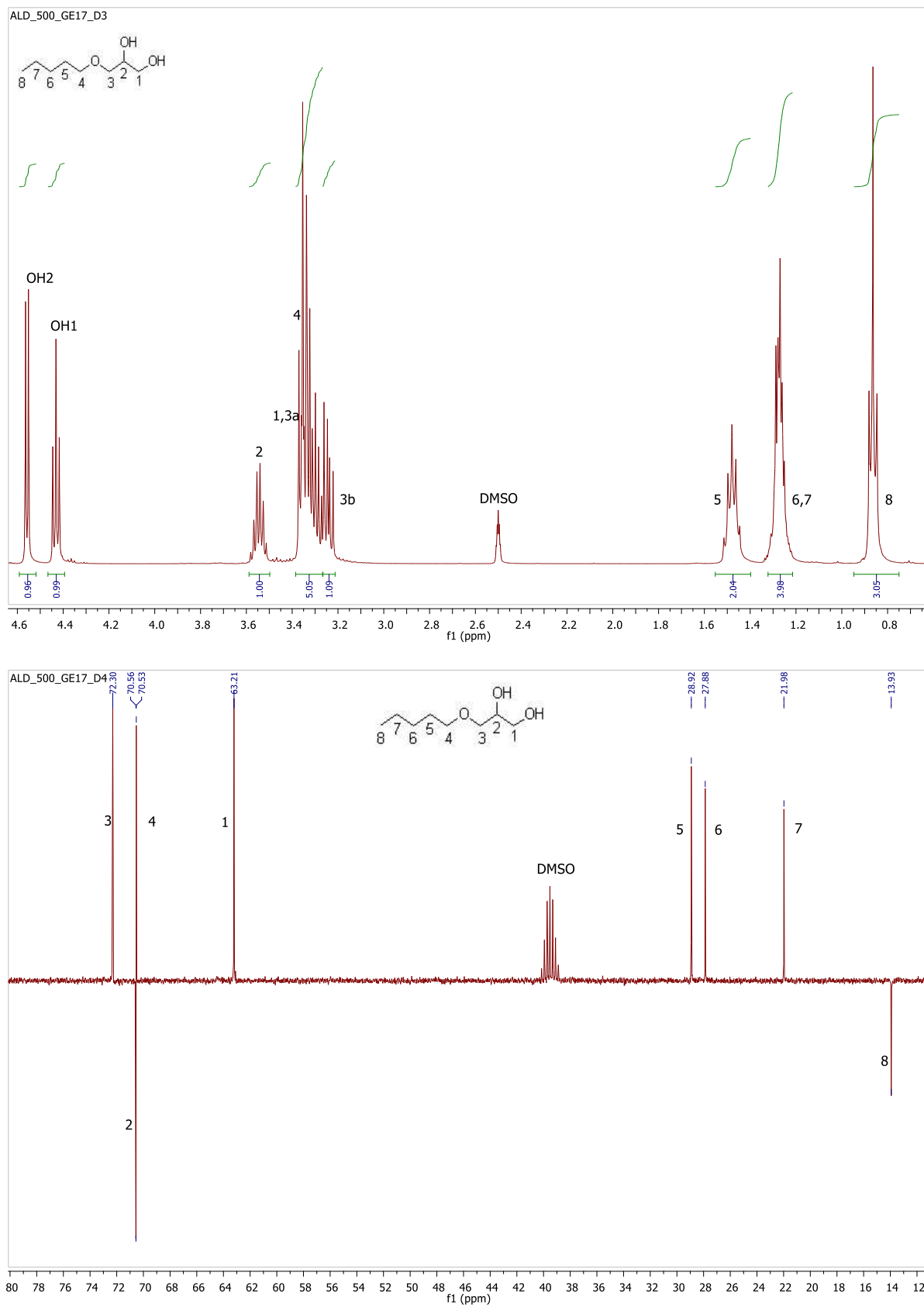


Figure S6. ^1H -RMN and ^{13}C -RMN (APT) of 3-hexyloxypropan-1,2-diol [6.0.0].

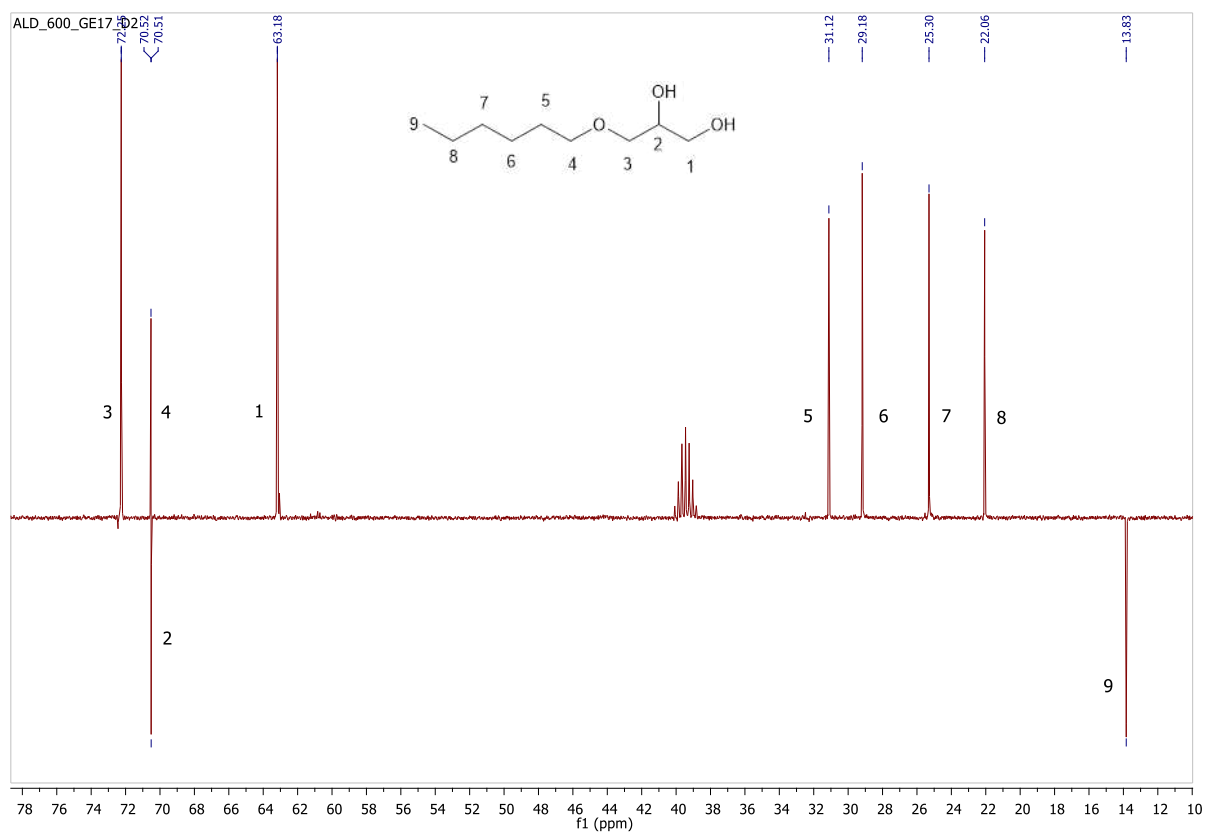
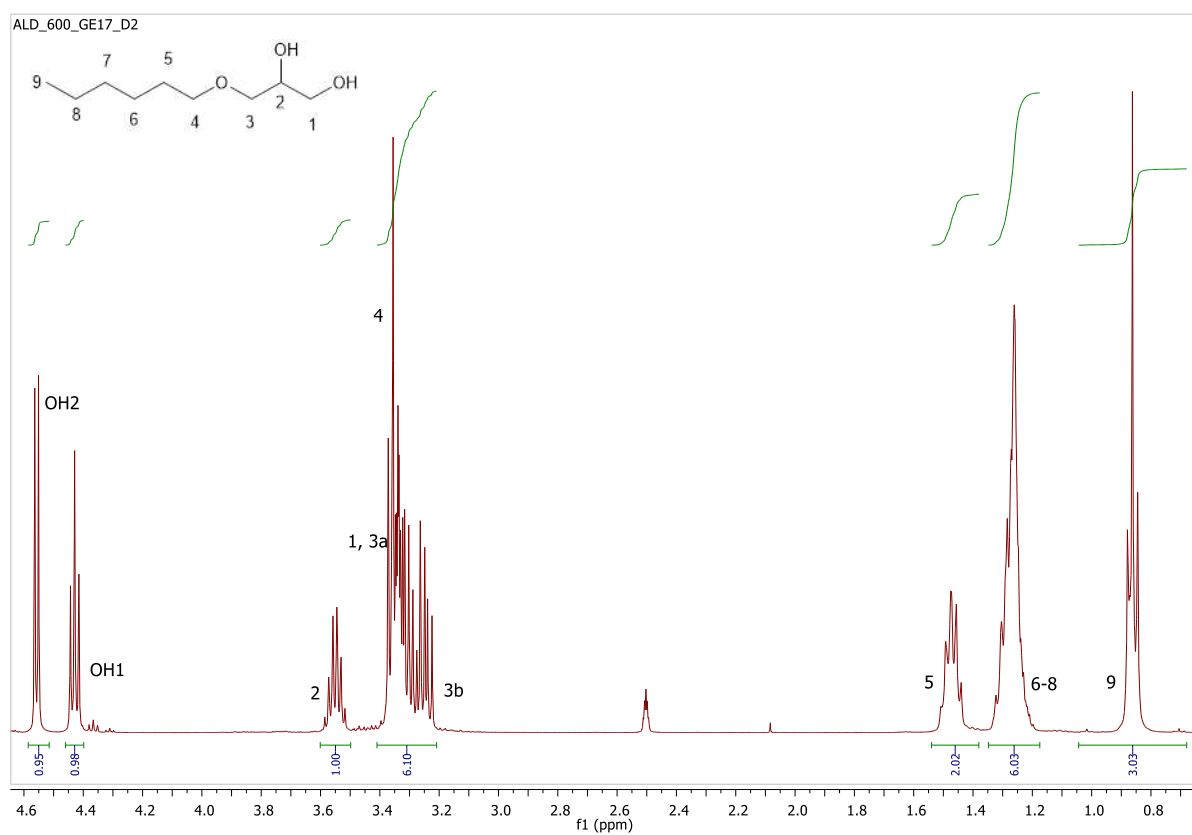


Figure S7. ^1H -RMN and ^{13}C -RMN (APT) of 1,3-Dimethoxypropan-2-ol [1.0.1].

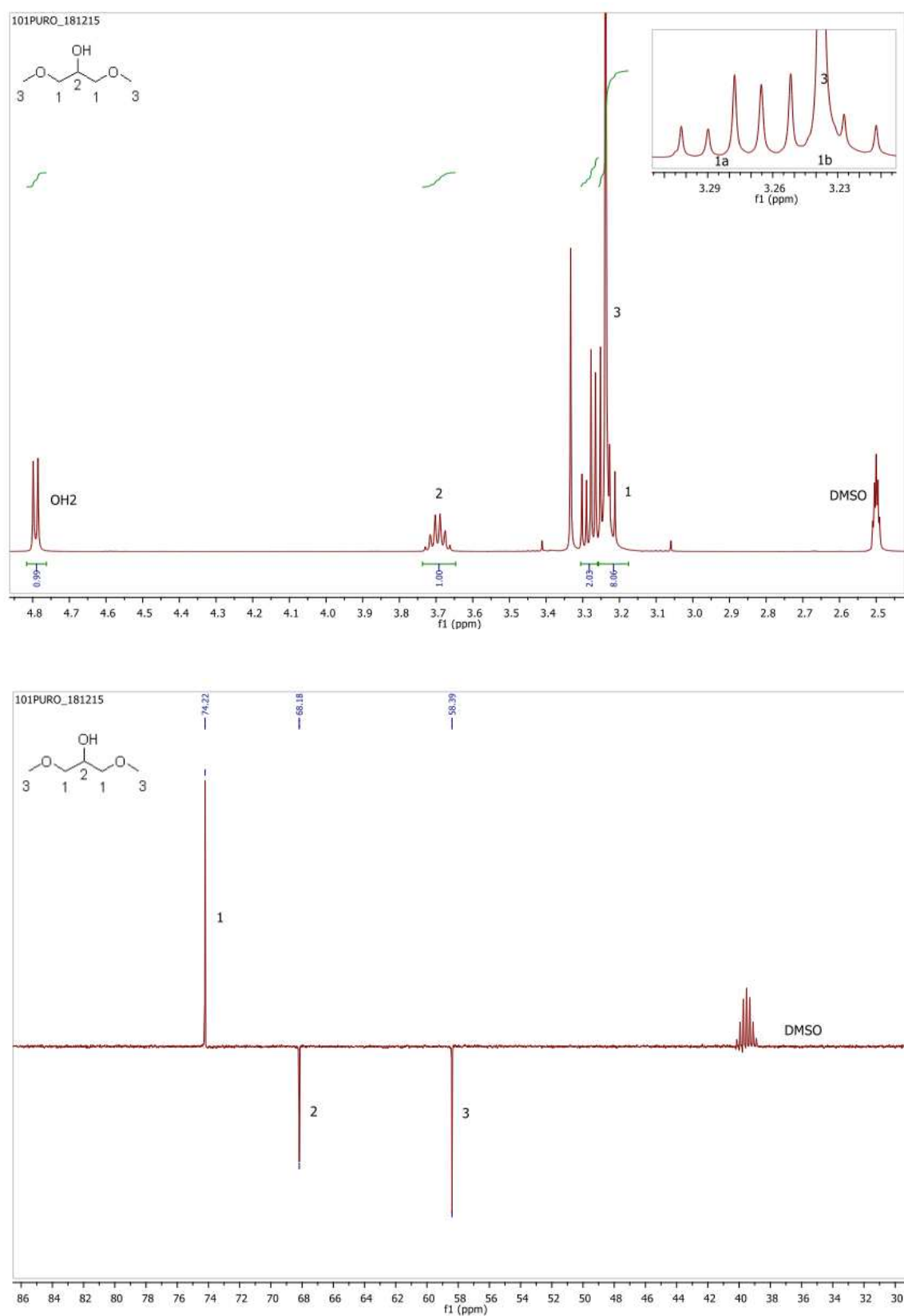


Figure S8. ^1H -RMN and ^{13}C -RMN (APT) of 1,3-Diethoxypropan-2-ol [2.0.2].

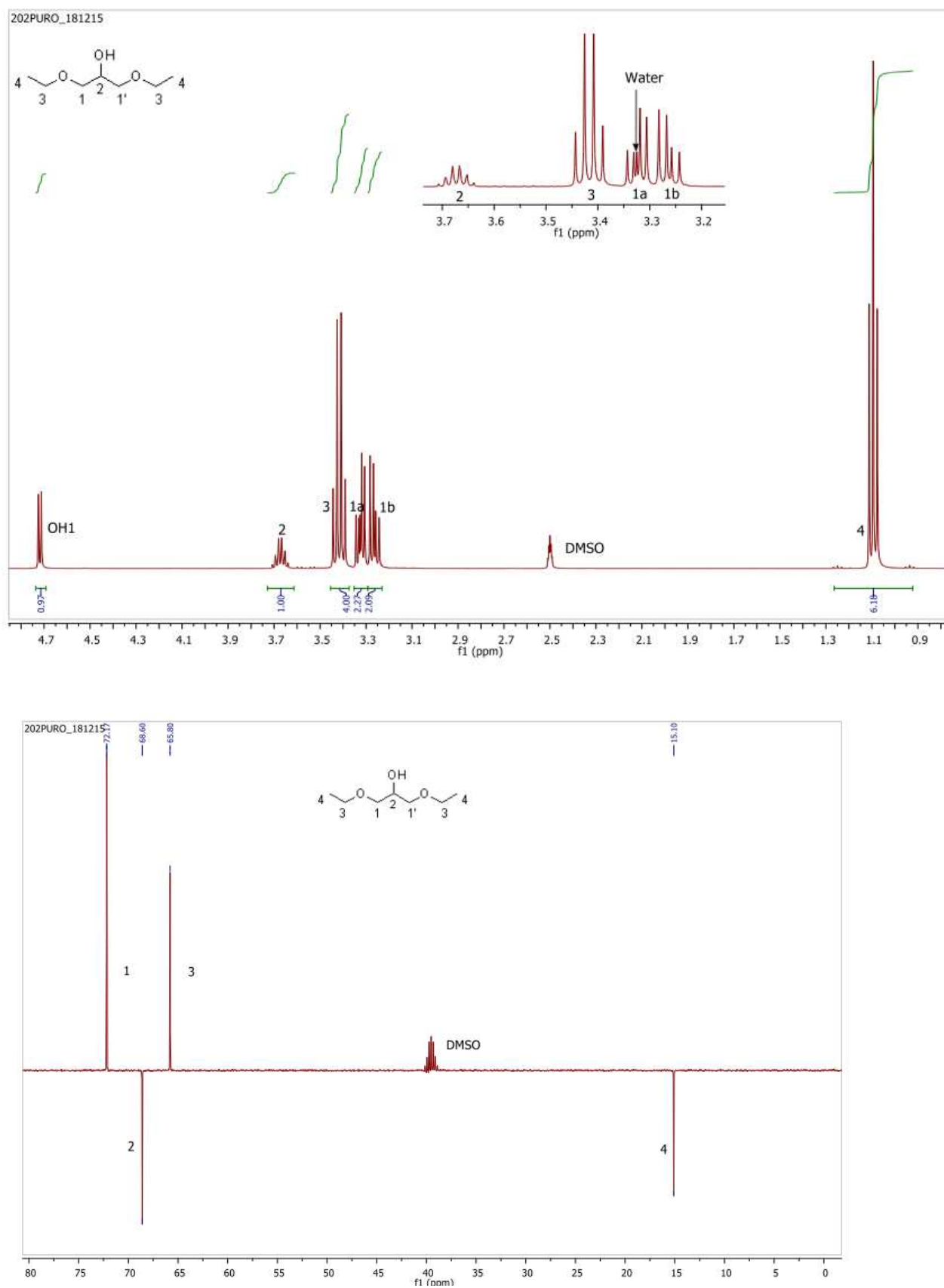
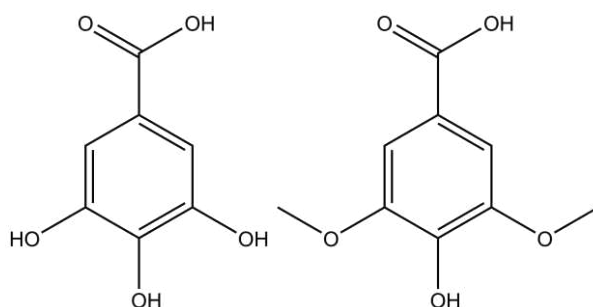


Figure S9. Chemical structure of gallic acid (left) and syringic acid (right).



S2. SOLUBILITY DATA

Gallic Acid

Table S2. Gallic acid solubility in water and aqueous solutions of [0.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
0	0.0919	0.0045
1.02	0.0937	0.0009
2.19	0.0989	0.0016
3.06	0.0967	0.0009
5.03	0.1020	0.0088
10.12	0.1261	0.0031
20.04	0.1572	0.0052
30.12	0.2003	0.0106
40.04	0.2686	0.0108
59.96	0.4711	0.0065
79.40	0.7119	0.0416
99.63	1.3762	0.0260

Table S3. Gallic acid solubility in aqueous solutions of [1.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.00	0.0975	0.0002
3.01	0.1091	0.0005
5.04	0.1343	0.0055
9.8	0.1823	0.0026
20.7	0.3315	0.0173
29.56	0.5211	0.0282
49.15	0.8818	0.0106
59.65	1.1346	0.0328
71.99	1.4183	0.0293
78.98	1.5740	0.0234
89.19	1.8729	0.0259
98.81	1.9177	0.0200

Table S4. Gallic acid solubility in aqueous solutions of [2.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.02	0.0975	0.0023
1.94	0.1093	0.0033
3.05	0.1158	0.0013
4.88	0.1269	0.0006
9.86	0.1716	0.0053
19.56	0.2358	0.0175
29.65	0.4716	0.0251
39.18	0.6334	0.0341
58.85	1.0318	0.0456
78.50	1.2626	0.0603
99.38	1.6179	0.0373

Table S5. Gallic acid solubility in aqueous solutions of [3.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.04	0.0988	0.0022
1.98	0.1100	0.0057
3.04	0.1212	0.0032
4.95	0.1326	0.0120
10.22	0.2018	0.0044
20.18	0.3192	0.0063
30.10	0.5457	0.0157
39.62	0.7886	0.0150
59.82	1.0213	0.0290
78.72	1.0803	0.0269
99.39	1.2681	0.0516

Table S6. Gallic acid solubility in aqueous solutions of [4.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.04	0.1009	0.0019
2.03	0.1144	0.0025
3.02	0.1237	0.0050
5.02	0.1466	0.0019
9.98	0.2185	0.0046
19.98	0.3877	0.0113
29.80	0.5311	0.0230
39.60	0.7103	0.0347
49.48	0.8006	0.0127
60.04	0.8433	0.0057
70.28	0.8748	0.0052
79.88	0.8685	0.0088
89.93	0.9110	0.0019
99.11	1.0273	0.0521

Table S7. Gallic acid solubility in aqueous solutions of [5.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.02	0.1016	0.0003
2.09	0.1204	0.0052
3.08	0.1328	0.0009
5.03	0.1632	0.0015
9.92	0.2070	0.0033
19.94	0.2808	0.0129
29.99	0.3424	0.0236
39.60	0.4334	0.0072
50.67	0.5985	0.0033
59.37	0.6123	0.0065
79.48	0.6270	0.0345
99.70	0.8001	0.0098

Table S8. Gallic acid solubility in aqueous solutions of [6.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
38.24	0.3454	0.0055
44.92	0.3901	0.0146
49.26	0.4569	0.0187
59.53	0.5373	0.0288
64.89	0.4949	0.0183
69.00	0.5113	0.0259
73.88	0.4980	0.0017
79.98	0.4814	0.0243
83.67	0.5309	0.0019
99.79	0.6272	0.0068

Table S9. Gallic acid solubility in aqueous solutions of [1.0.1] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.00	0.1051	0.0006
3.01	0.1238	0.0014
5.02	0.1434	0.0023
10.19	0.2180	0.0020
20.36	0.4297	0.0192
29.88	0.6336	0.0022
39.99	0.9839	0.0393
50.17	1.1893	0.0281
59.89	1.4312	0.0177
69.39	1.5821	0.0088
79.96	1.8102	0.0016
99.92	1.8623	0.0902

Table S10. Gallic acid solubility in aqueous solutions of [2.0.2] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.06	0.1039	0.0013
3.06	0.1265	0.0003
5.01	0.1502	0.0062
10.03	0.2289	0.0015
20.21	0.4602	0.0102
30.05	0.6837	0.0299
39.97	0.9669	0.0071
50.06	1.1384	0.0131
59.26	1.3351	0.0163
69.85	1.4111	0.0313
79.97	1.4256	0.0400
99.95	1.3826	0.0810

Syringic Acid

Table S11. Syringic acid solubility in water and aqueous solutions of [0.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
0	0.0085	0.00002
1.04	0.0086	0.0003
3.03	0.0090	0.00001
2.02	0.0091	0.0001
5.11	0.0236	0.0006
10.05	0.0110	0.0002
19.90	0.0119	0.0007
29.74	0.0154	0.0003
39.77	0.0183	0.0002
49.70	0.0206	0.0002
59.91	0.0232	0.0002
69.43	0.0254	0.0005
79.10	0.0291	0.0000
89.40	0.0327	0.0002
99.63	0.0386	0.0026

Table S12. Syringic acid solubility in aqueous solutions of [1.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.00	0.0089	0.0002
3.01	0.0112	0.0000
6.19	0.0147	0.0006
10.14	0.0187	0.0022
19.95	0.0345	0.0003
29.62	0.0552	0.0004
39.83	0.0842	0.0007
59.62	0.1824	0.0006
69.52	0.2534	0.0018
79.06	0.3343	0.0008
98.81	0.4367	0.0021

Table S13. Syringic acid solubility in aqueous solutions of [2.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.02	0.0091	0.0006
3.05	0.0103	0.0004
5.46	0.0126	0.0001
10.21	0.0160	0.0008
19.78	0.0309	0.0002
29.81	0.0607	0.0019
39.68	0.1083	0.0028
59.64	0.2629	0.0036
69.44	0.3358	0.0009
80.33	0.4023	0.0038
99.38	0.3234	0.0064

Table S14. Syringic acid solubility in aqueous solutions of [3.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.04	0.0091	0.0001
3.04	0.0110	0.0001
5.16	0.0125	0.0001
10.29	0.0216	0.0002
20.14	0.0552	0.0003
30.20	0.1214	0.0037
39.77	0.1952	0.0016
57.22	0.3222	0.0014
69.45	0.3843	0.0014
79.43	0.3911	0.0007
99.39	0.2428	0.0085

Table S15. Syringic acid solubility in aqueous solutions of [4.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.04	0.0091	0.00003
3.02	0.0117	0.0004
5.02	0.0171	0.0001
10.03	0.0301	0.0003
20.19	0.0888	0.0004
30.02	0.1489	0.0008
40.48	0.2127	0.0080
59.64	0.3054	0.0046
68.33	0.3281	0.0050
78.87	0.3356	0.0022
99.11	0.1971	0.0010

Table S16. Syringic acid solubility in aqueous solutions of [5.0.0] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.02	0.0093	0.0003
3.08	0.0134	0.0001
5.07	0.0201	0.0008
9.96	0.0373	0.0012
19.96	0.0727	0.0020
29.77	0.1088	0.0029
39.35	0.1449	0.0011
59.90	0.2199	0.0022
70.11	0.2435	0.0053
79.92	0.2548	0.0028
99.70	0.1554	0.0017

Table S17. Syringic acid solubility in aqueous solutions of [1.0.1] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.00	0.0093	0.0002
3.01	0.0120	0.0004
5.02	0.0126	0.0002
10.09	0.0185	0.0006
20.36	0.0453	0.0014
29.88	0.0839	0.0029
39.99	0.1691	0.0033
49.72	0.2930	0.0093
59.89	0.4393	0.0070
69.84	0.5407	0.0043
79.96	0.6568	0.0175
99.92	0.4597	0.0228

Table S18. Syringic acid solubility in aqueous solutions of [2.0.2] at 303.2 K.

wt% Hydrotrope	Solubility /M	s /M
1.06	0.0094	0.0003
3.06	0.0120	0.0004
5.01	0.0145	0.0005
10.08	0.0236	0.0006
20.21	0.0702	0.0043
30.13	0.1585	0.0035
39.97	0.2999	0.0109
50.04	0.3744	0.0085
59.26	0.5044	0.0101
69.96	0.5293	0.0157
79.97	0.5299	0.0068
99.95	0.1863	0.0094

S3. COOPERATIVE HYDROTROPY MODEL

In order to use the cooperative hydrotropy model to fit the experimental data, it was necessary to convert the concentration of hydrotrope in the solute (free-solute basis) to its mole fraction in the ternary system ($x_{Hydrotrope}$), as well as the solubility of the solute to mole fraction in the ternary system (x_{Solute}). It was assumed that the density of the system is equal to that of water. Hence, Equations S1 and S2 were used:

$$x_{Hydrotrope} = \frac{\frac{(1000-S_3) \cdot w_2}{M_2}}{\frac{(1000-S_3) \cdot w_1}{M_1} + \frac{(1000-S_3) \cdot w_2}{M_2} + \frac{S_3}{M_3}} \quad (S1)$$

$$x_{Solute} = \frac{\frac{S_3}{M_3}}{\frac{(1000-S_3) \cdot w_1}{M_1} + \frac{(1000-S_3) \cdot w_2}{M_2} + \frac{S_3}{M_3}} \quad (S2)$$

where w_1 and w_2 are the mass fractions of water and hydrotrope in the solvent (free-solute basis), respectively, M_1 , M_2 and M_3 are the molar masses of water, hydrotrope and solute, respectively, and S_3 is the solubility of the solute in the hydrotropy system (mol/L).

Figure S10. Experimental solubility data, $\blacklozenge$, for gallic acid in aqueous solutions of a) [0.0.0], b) [1.0.0], c) [2.0.0], d) [3.0.0], e) [4.0.0], f) [5.0.0], g) [1.0.1] and h) [2.0.2], fitted using the cooperative hydrotrope model (---). The x-axis represents the mole fraction of the hydrotrope in the ternary system (as opposed to its mole fraction in the solvent free of solute).

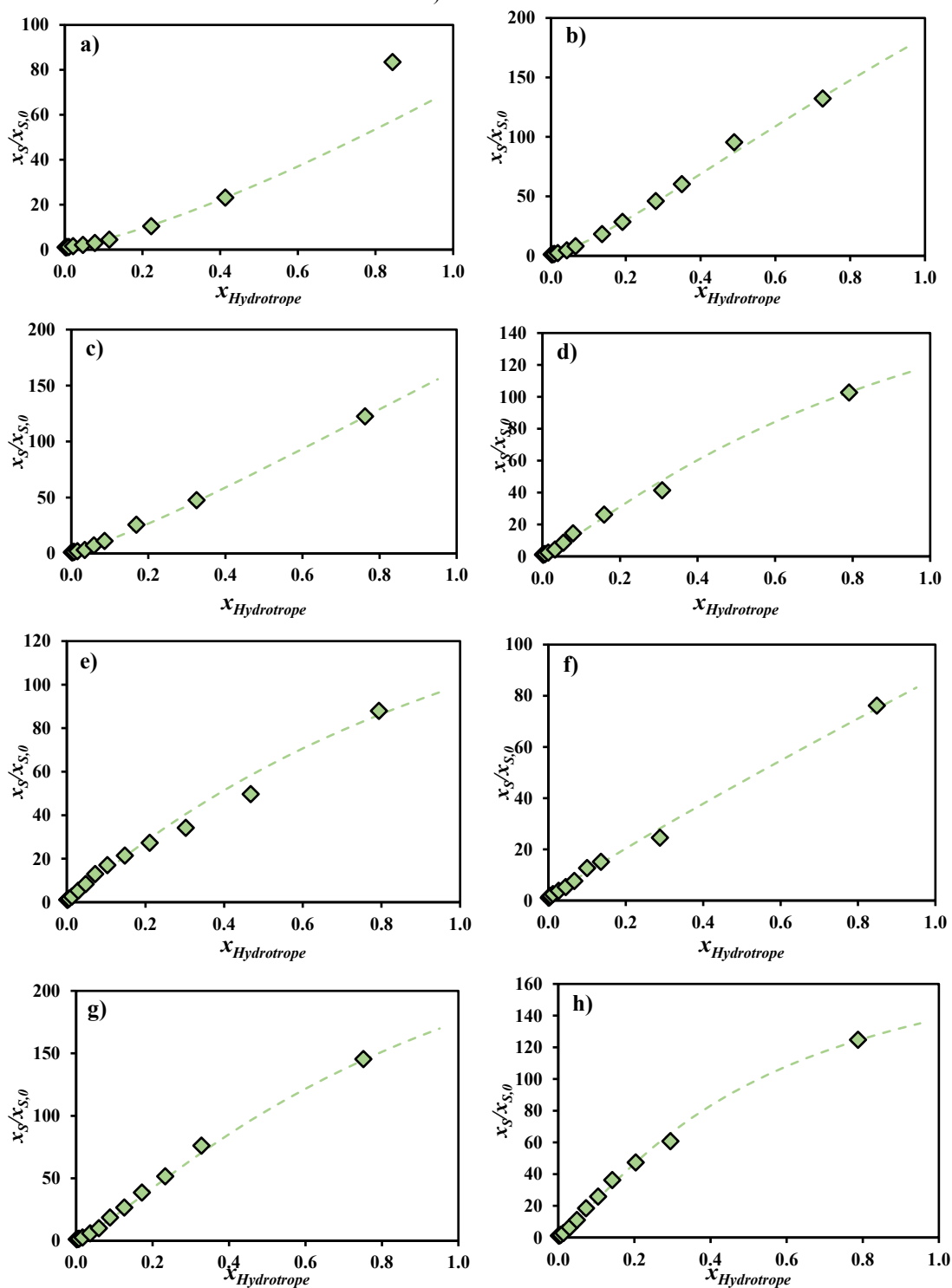


Figure S11. Experimental solubility data, $\blacklozenge$, for syringic acid in aqueous solutions of a) [0.0.0], b) [1.0.0], c) [2.0.0], d) [3.0.0], e) [4.0.0], f) [5.0.0], g) [1.0.1] and h) [2.0.2], fitted using the cooperative hydrotrope model (---). The x-axis represents the mole fraction of the hydrotrope in the ternary system (as opposed to its mole fraction in the solvent free of solute).

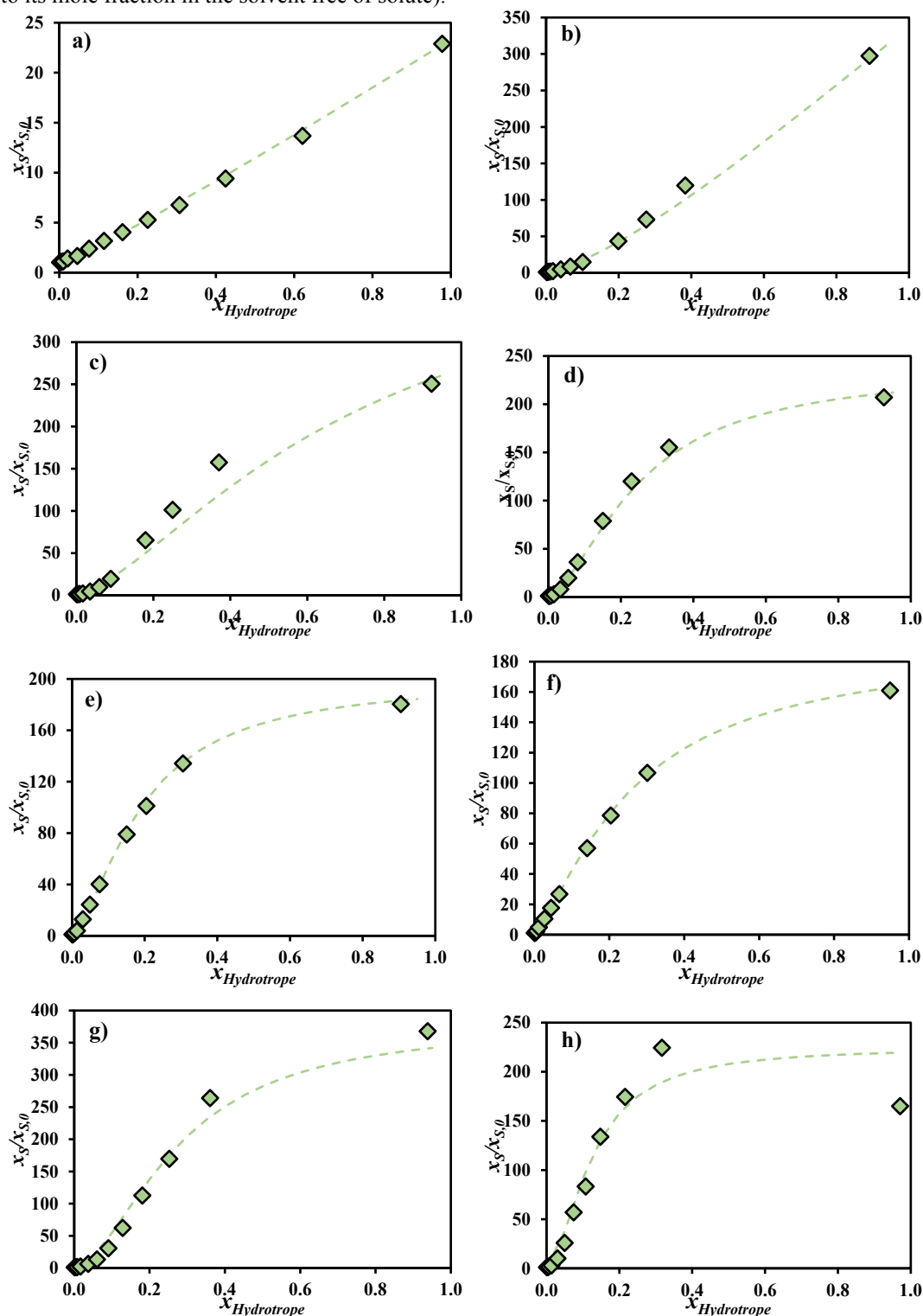


Table S19. Parameters of the cooperative hydrotrophy model for the solubility of gallic and syringic acids in glycerol ether-based hydrotrope solutions.

	Hydrotrope	<i>max</i>	<i>m</i>	<i>b</i>
Gallic acid	[0.0.0]	10000 ^{a)}	1.317	-4.949
	[1.0.0]	675	1.313	-0.988
	[2.0.0]	1606	1.208	-2.178
	[3.0.0]	236	1.200	0.013
	[4.0.0]	228	1.084	-0.259
	[5.0.0]	10000 ^{a)}	0.932	-4.746
	[1.0.1]	347	1.252	0.014
	[2.0.2]	201	1.235	0.769
Syringic acid	[0.0.0]	562	1.126	-3.183
	[1.0.0]	2371	1.382	-1.801
	[2.0.0]	432	1.482	0.490
	[3.0.0]	236	1.628	2.260
	[4.0.0]	199	1.561	2.593
	[5.0.0]	192	1.330	1.782
	[1.0.1]	379	1.782	2.299
	[2.0.2]	225	1.789	3.729

a) unbounded optimization: increasing the value *ad infinitum* decreases the quadratic error sum.

S4. SOLUTE RECOVERY

The feasibility of adding water to recover dissolved solute from the hydrotropic system was preliminarily studied in this work using the solubility curves fitted with the cooperative hydrotropy model. The concept relies on calculating the mass fraction of solute that is recovered by the addition of a given volume of water. The algorithm used to calculate the recovery curves is as follow. The hydrotropic system, with total mole number n_t^0 , is selected (solute-hydrotrope pair and hydrotrope mole fraction).

1. Select fraction of water to be added (n_{H_2O}/n_t^0).
2. Arbitrate the fraction of solute that precipitates (n_p/n_t^0).
3. Calculate increase of the new system (n_t/n_t^0):

$$\frac{n_t}{n_t^0} = 1 + \frac{n_{H_2O}}{n_t^0} - \frac{n_p}{n_t^0}$$

4. Calculate hypothetical composition of the new system (x_1, x_2, x_3) from the composition of the original system (x_1^0, x_2^0, x_3^0):

$$x_1 = \frac{x_1^0 + n_{H_2O}/n_t^0}{n_t/n_t^0}$$

$$x_2 = \frac{x_2^0}{n_t/n_t^0}$$

$$x_3 = \frac{x_3^0 - n_p/n_t^0}{n_t/n_t^0}$$

5. Calculate saturation mole fraction of solute (x_3') for the hypothetical mole fraction of hydrotrope obtained in step 3 (Equation 3.3 of the main text with the parameters of Table S19).
6. If $x_3 = x_3'$, stop iteration. If $x_3 > x_3'$, return to step 1 and arbitrate higher n_p . If $x_3 < x_3'$, return to step 1 and arbitrate lower n_p .

Figure S12. Estimated fraction of gallic acid (w_{Solute}) recovered from hydrotrope solution by the addition of water (V_{Water} is the volumetric reason between added water and initial system), with an initial hydrotrope mole fraction of 0.01 —, 0.05 —, 0.1 —, 0.2 —, 0.4 —, 0.6 — and 0.8 —. A negative value indicates that no precipitation happens, with the system being able to dissolve further solute.

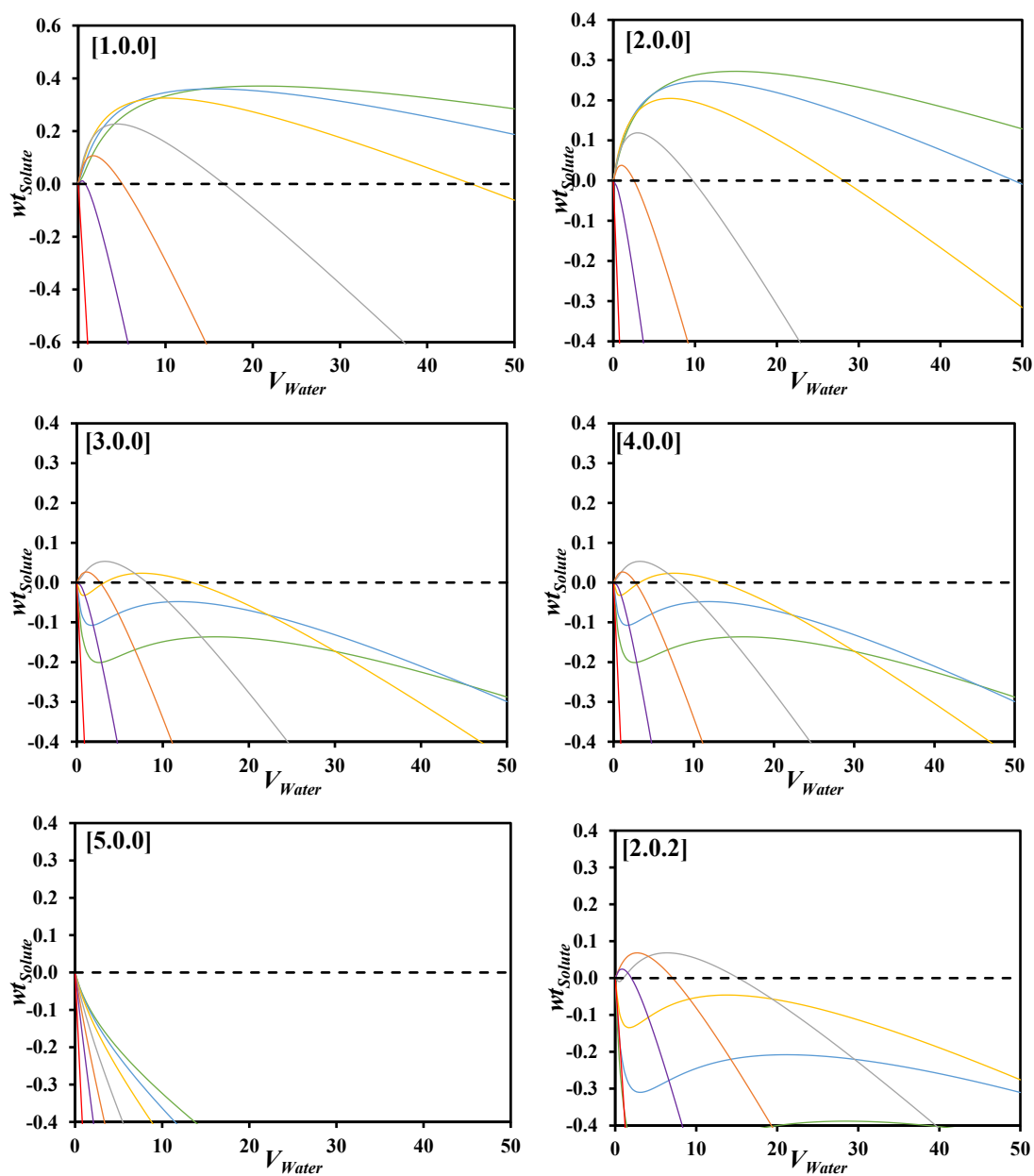
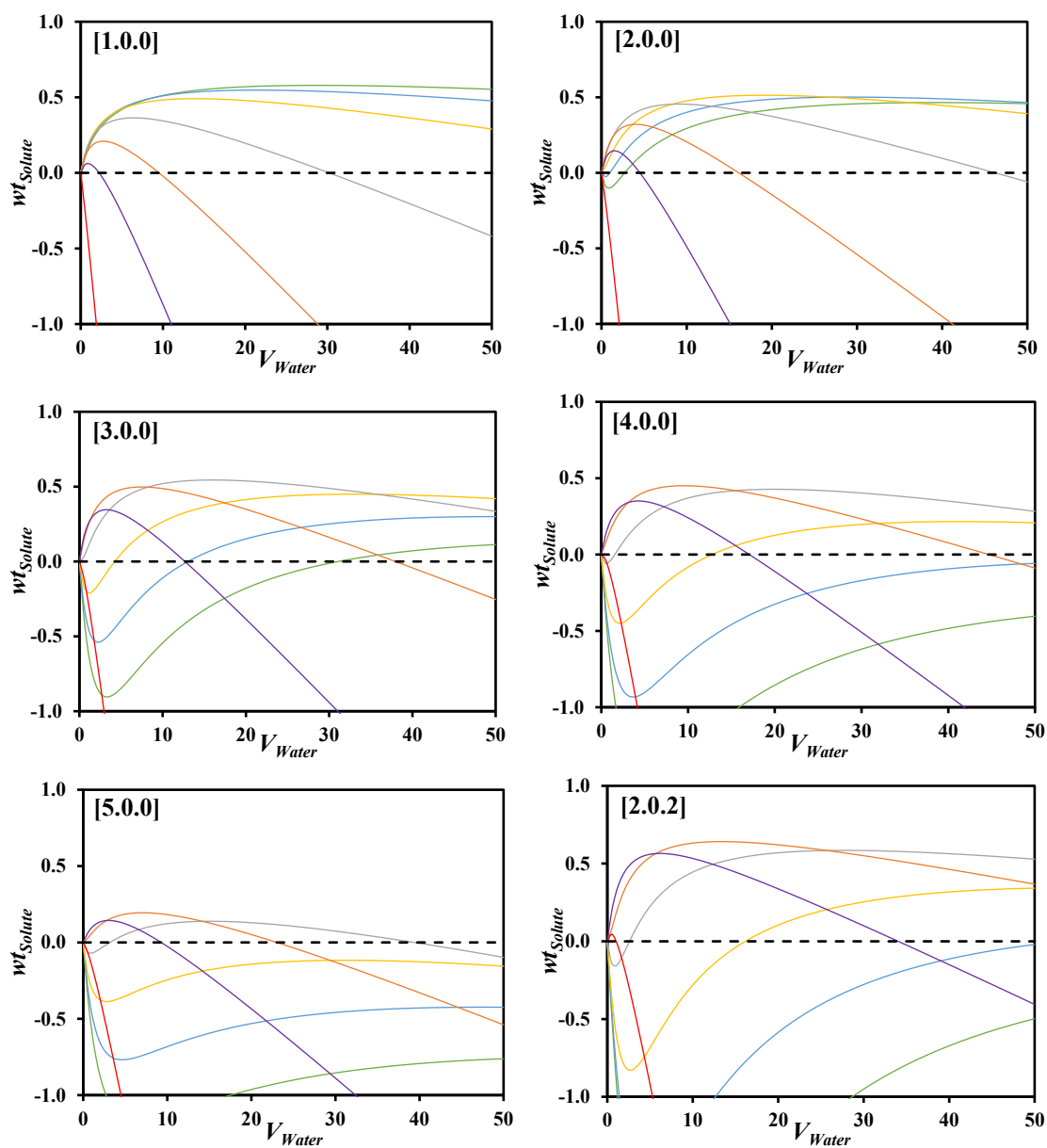


Figure S13. Estimated fraction of syringic acid (w_{Solute}) recovered from hydrotrope solution by the addition of water (V_{Water} is the volumetric reason between added water and initial system), with an initial hydrotrope mole fraction of 0.01 —, 0.05 —, 0.1 —, 0.2 —, 0.4 —, 0.6 — and 0.8 —. A negative value indicates that no precipitation happens, with the system being able to dissolve further solute.



APPENDIX B

S1. EXPERIMENTAL DETAILS

Table S1. Solute concentration in the hydrotropic systems (0.4 M of hydrotrope) whose ^1H -NMR spectra was measured in this work.

Hydrotrope	Gallic Acid Concentration /mol·kg ⁻¹	Syringic Acid Concentration /mol·kg ⁻¹
[1.0.0]	0	0
	0.06	0.005
	0.12	0.010
[2.0.0]	0	0
	0.06	0.005
	0.12	0.010
[3.0.0]	0	0
	0.06	0.005
	0.12	0.010
[4.0.0]	0	0
	0.06	0.008
	0.12	0.013
[5.0.0]	0	0
	0.06	0.010
	0.12	0.018

σ -Profile

Each molecule (either solute or hydrotrope) was optimized using TURBOMOLE, through its interface TmoleX, by adopting the TZVP template available in the software package. This template uses a def-TZVP basis set for all atoms, a DFT with the B-P86 functional and the COSMO solvation model (continuum with infinite permittivity). From the molecule file so obtained, the σ -profile of the molecule was computed using the COSMOtherm software with the BP_TZVP_C30_1701 parametrization. For chloride-based ionic liquids, the cation was optimized separately from the chloride anion. The apolar factor of the ionic liquid is obtained by summing the apolar factors of the cation and of the anion.

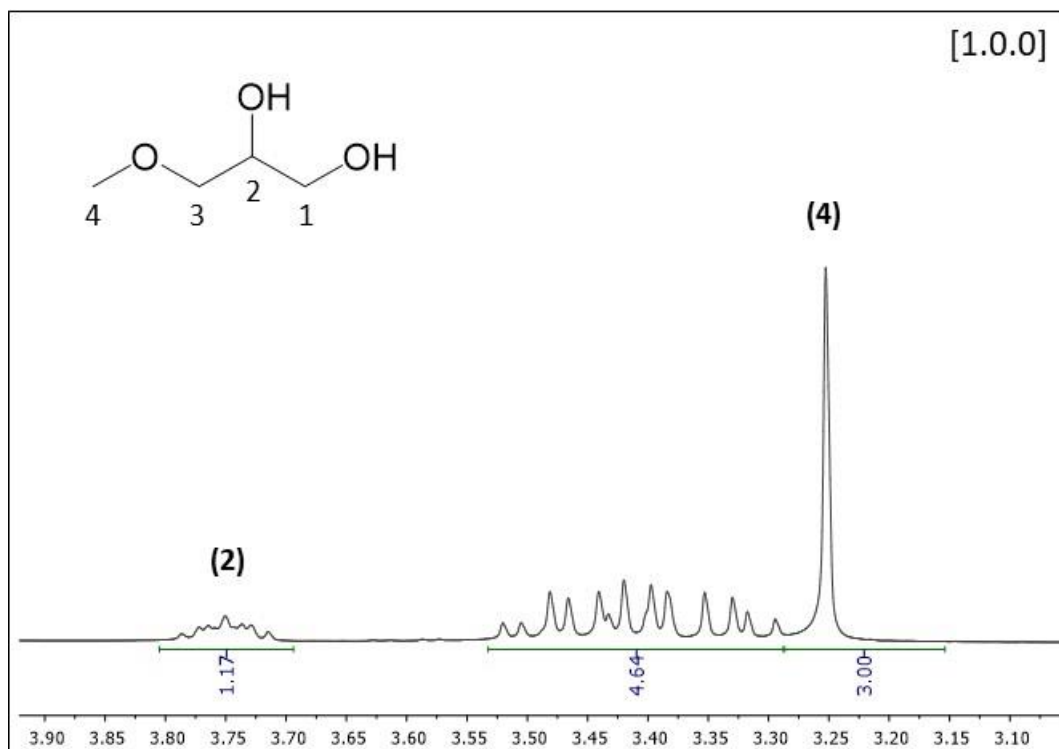
Note that, unlike for the glycerol ether hydrotrope series, the hydrotropy extent of some chloride-based ionic liquids does not correlate well with the corresponding apolar factor (Figure S4). This is most likely due to the role of the chloride ion, which may stay solvated or accumulate around the solute-cation cluster for charge stabilization.

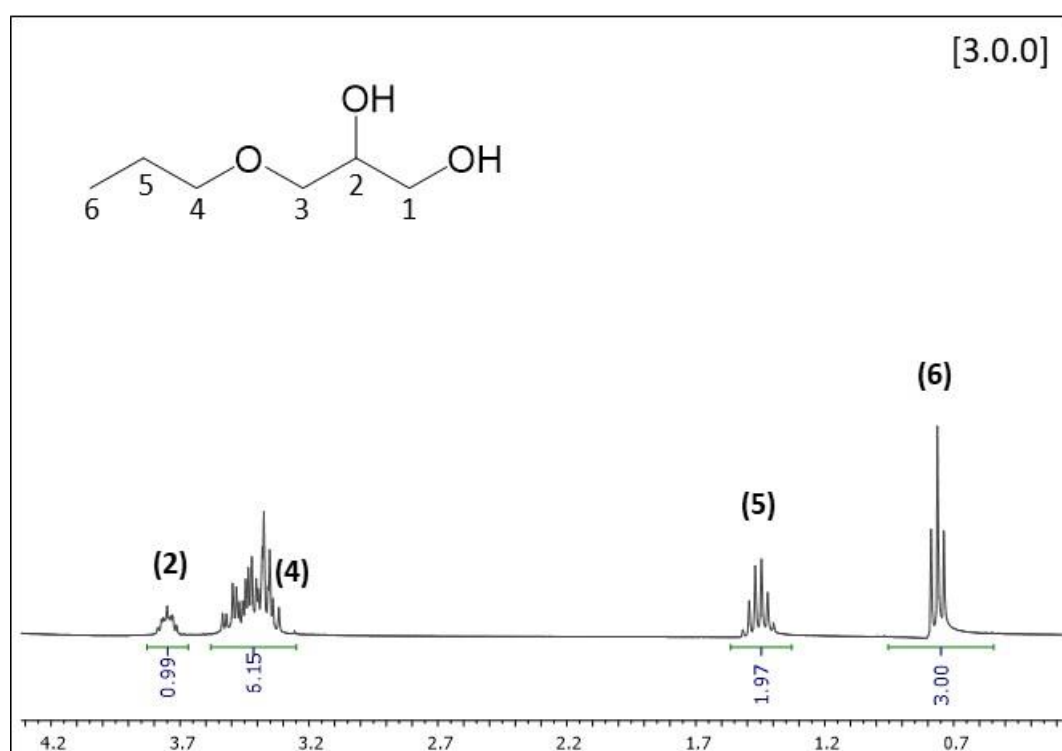
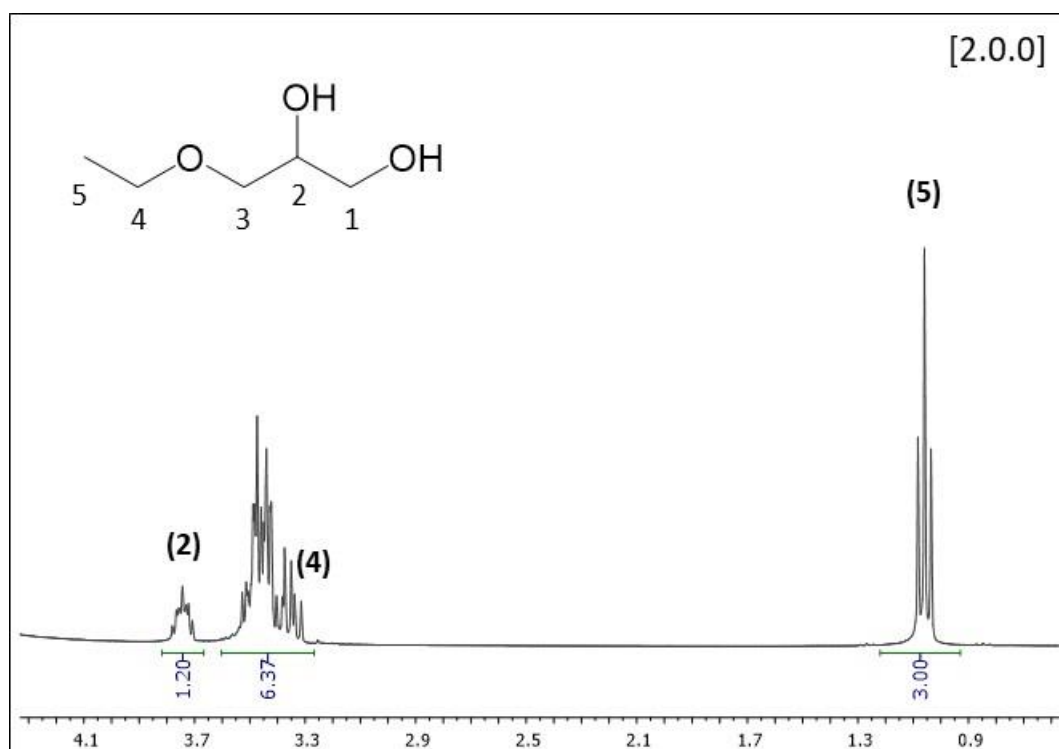
Table S2. Apolar Factor of substances and ions used in the literature as hydrotropes.

Substance/Ion	Apolar Factor ($\cdot 10^3$)	Substance/Ion	Apolar Factor ($\cdot 10^3$)
1-ethyl-3-methylimidazolium	0.4755	Chloride	0
1-butyl-3-methylimidazolium	0.7120	Gallic Acid	0.4981
1-hexyl-3-methylimidazolium	0.9684	Syringic Acid	0.7235
1-octyl-3-methylimidazolium	1.2264	[0.0.0]	0.3352
1-butyl-3-methylpyridinium	0.8495	[1.0.0]	0.5041
1-butyl-1-methylpiperidinium	0.7764	[2.0.0]	0.6481
1-butyl-1-methylpyrrolidinium	0.6958	[3.0.0]	0.7820
Tetrabutylammonium	1.6684	[4.0.0]	0.9117
Tetrabutylphosphonium	1.7036	[5.0.0]	1.0406
Cholinium	0.2170	[1.0.1]	0.6828
Sodium	0	[2.0.2]	0.9636

S2. COMPLEMENTARY FIGURES

Figure S1. ^1H -NMR spectra for aqueous solutions of monoalkylglycerol ethers (0.4 mol/kg concentration) in the chemical shift window used in this work. From top to bottom, [1.0.0], [2.0.0], [3.0.0], [4.0.0] and [5.0.0], with the structure of each substance reported as inset. Signaled peaks are those used in this work, with the number matching the number sequence of the inset structure. Chemical shifts (x-axis) are reported in ppm.





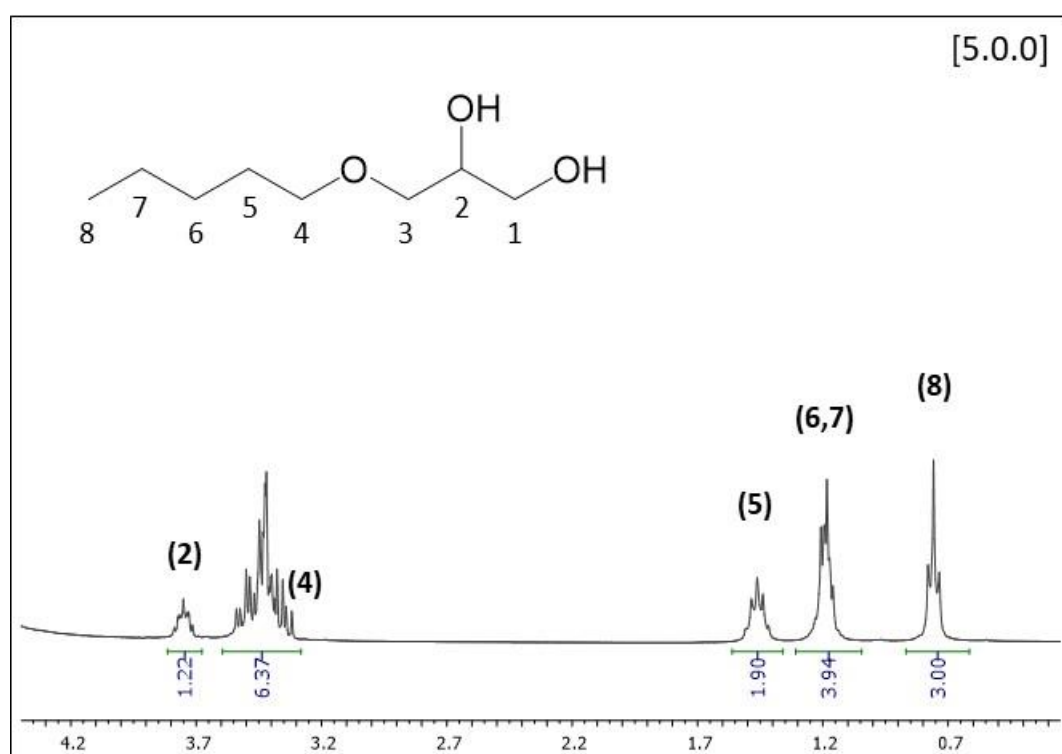
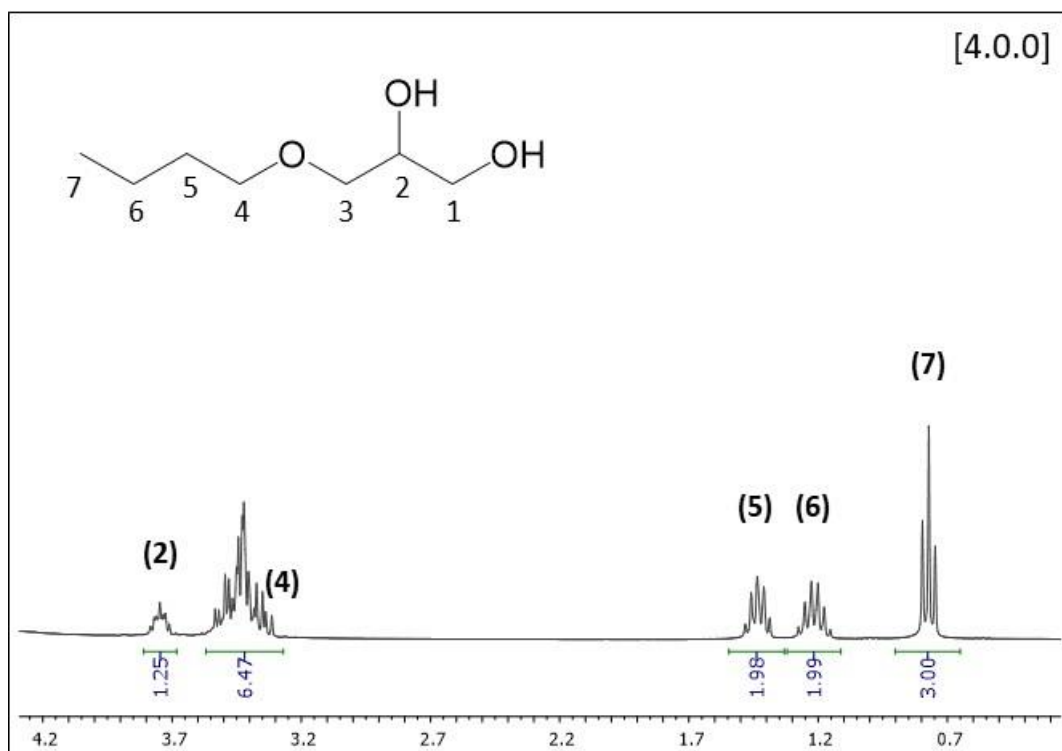


Figure S2. Change in chemical shift of the protons ($-\Delta\delta_H$) associated to water and several methyl groups of a) [1.0.0], b) [2.0.0], c) [3.0.0] and d) [5.0.0] dissolved in water (0.4 mol/kg) as a function of gallic acid concentration. Similar data for [4.0.0] is reported in the main text (Figure 3.7). Legend: $\circ$ - water; $\diamond$ - 2nd carbon; $\blacklozenge$ - 4th carbon; $\blacklozenge$ - 5th carbon; $\blacklozenge$ - 6th carbon; $\blacklozenge$ - 7th carbon; $\blacklozenge$ - 8th carbon. For [5.0.0] the peaks from the hydrogens of the 6th and 7th carbon are undistinguishable and are represented as $\blacklozenge$.

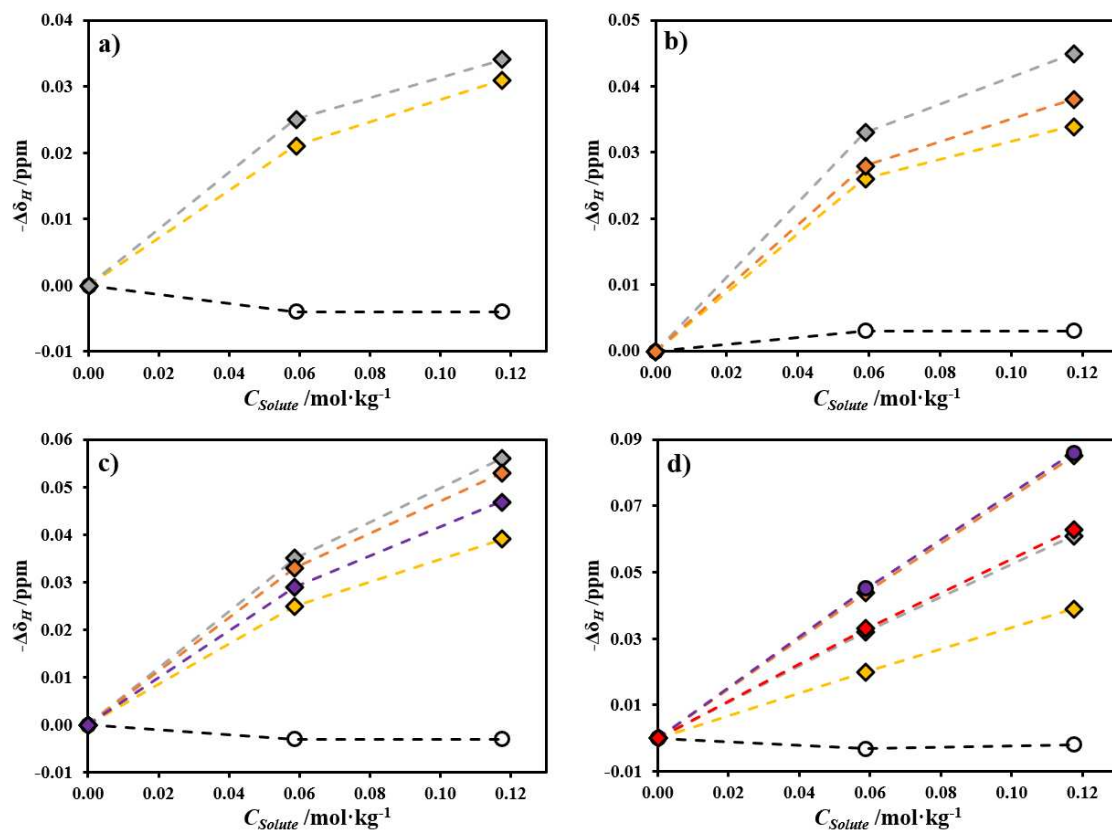


Figure S3. Change in chemical shift of the protons ($-\Delta\delta_H$) associated to water and several methyl groups of a) [1.0.0], b) [2.0.0], c) [3.0.0] and d) [5.0.0] dissolved in water (0.4 mol/kg) as a function of syringic acid concentration. Similar data for [4.0.0] is reported in the main text (Figure 3.7). Legend: $\circ$ - water; $\diamond$ - 2nd carbon; $\diamond$ - 4th carbon; $\diamond$ - 5th carbon; $\diamond$ - 6th carbon; $\diamond$ - 7th carbon; $\diamond$ - 8th carbon. For [5.0.0] the peaks from the hydrogens of the 6th and 7th carbon are undistinguishable and are represented as $\bullet$ -.

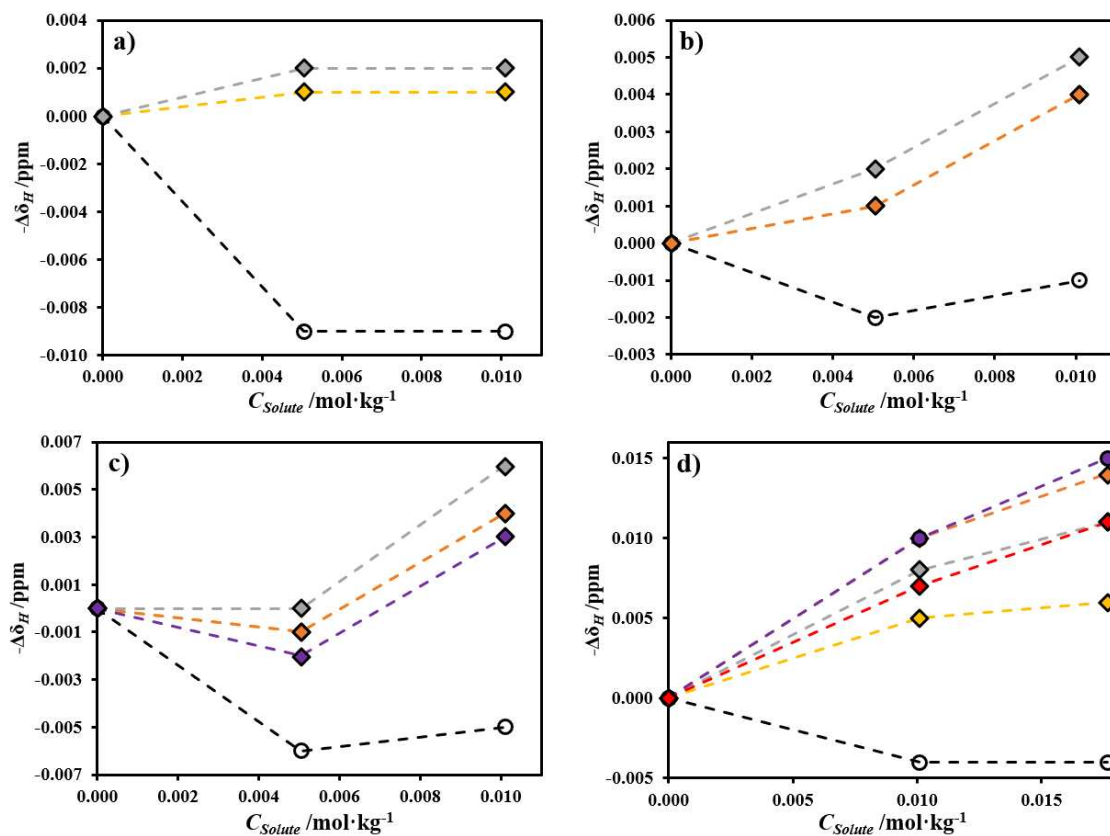


Figure S4. Setschenow constants of a) vanillin (CLÁUDIO *et al.*, 2015) b) gallic acid (CLÁUDIO *et al.*, 2015) and c) ibuprofen (SINTRA *et al.*, 2018) for chloride-based ionic liquid hydrotropic systems as a function of the apolar factor of the ionic liquid. The dashed lines are the straight lines fitted to the data using the least squares method.

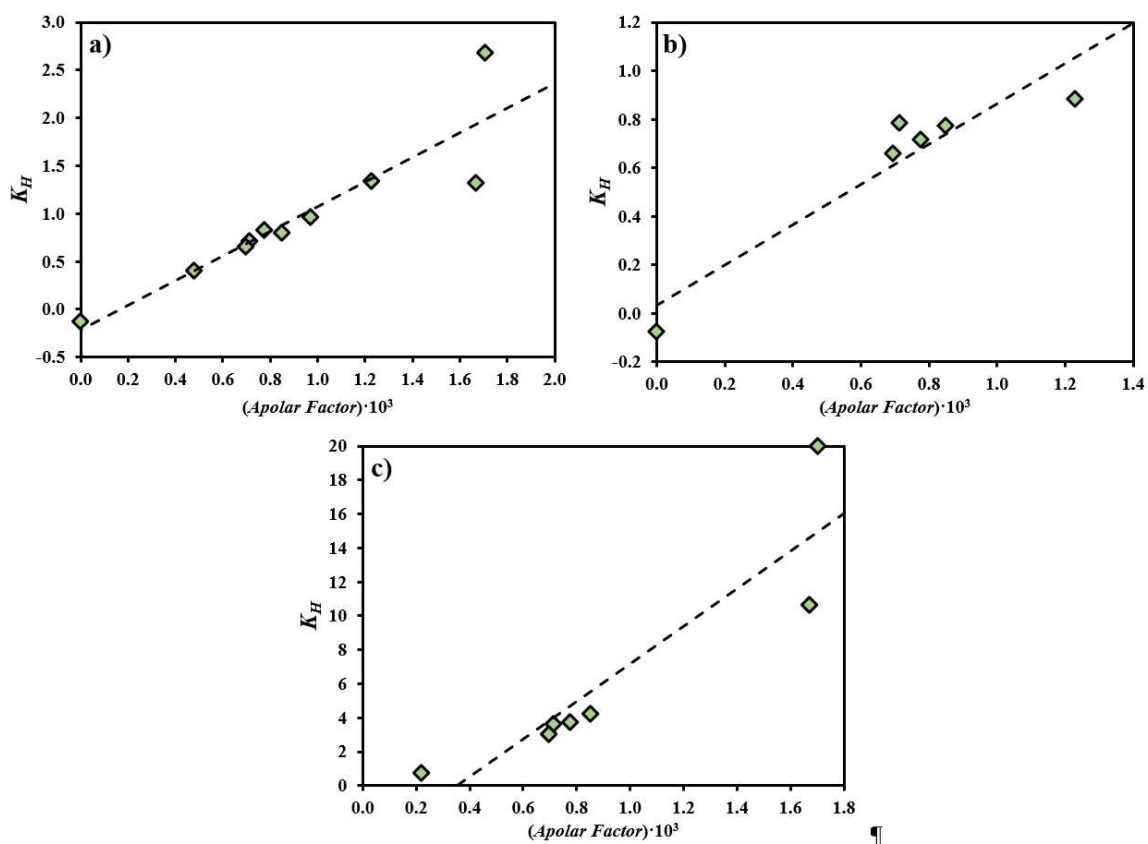
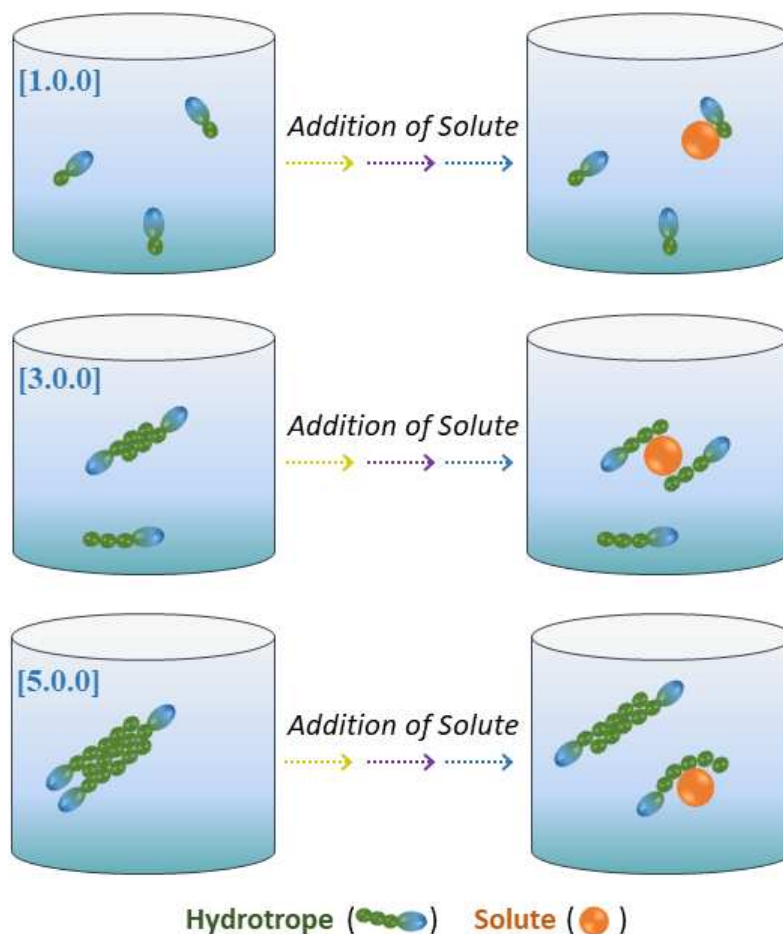


Figure S5. Schematic illustration of the hydrotropy mechanism, evidencing the main findings in this work. In the first case, apolarity of the hydrotrope is small and the driving force for aggregation is low; in the second case, apolarity of the hydrotrope is equal to that of the solute and the driving force for aggregation is maximized, resulting in more hydrotrope molecules aggregated around the solute; in the third case, apolarity of the hydrotrope is greater than that of the solute and, thus, driving force for hydrotrope-hydrotrope aggregation is larger than that of hydrotrope-solute aggregation.



S3. REFERENCES

TURBOMOLE V7.1 2016, a development of University of Karlsruhe and Forschungszentrum Karlsruhe GmbH, 1989-2007, TURBOMOLE GmbH, since 2007; available from <http://www.turbomole.com>.

COSMOtherm, Release 19; COSMOlogic GmbH & Co. KG, <http://www.cosmologic.de>.

CLÁUDIO, A. F. M. *et al.* The magic of aqueous solutions of ionic liquids: ionic liquids as a powerful class of catanionic hydrotropes. **Green Chemistry**, v. 17, n. 7, p. 3948–3963, 2015.

SINTRA, T. E. *et al.* Enhanced dissolution of ibuprofen using ionic liquids as catanionic hydrotropes. **Physical Chemistry Chemical Physics**, v. 20, n. 3, p. 2094–2103, 2018.

APPENDIX C

S1. COMPLEMENTARY TABLES

Table S1. Experimental solubility of syringic acid (g/L of solvent) in glycol ethers aqueous mixtures at 303.15K at different weight fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

Compound / w_H	2-ethoxyethanol	$\pm s$	1-propoxy-2-propanol	$\pm s$	2- butoxyethanol	$\pm s$
0.00	1.462	0.004	-	-	-	-
0.01	1.63	0.06	1.724	0.003	-	-
0.02	1.84	0.01	2.07	0.05	-	-
0.03	2.098	0.005	2.43	0.02	-	-
0.04	-	-	2.753	0.002	-	-
0.05	2.57	0.04	3.24	0.07	-	-
0.10	3.97	0.07	6.5	0.1	3.5	0.2
0.20	10.18	0.06	21.6	0.1	9.046	0.008
0.30	23.8	0.5	37.4	0.4	25.0	0.2
0.40	45	1	53.6	0.5	41.161	0.07
0.50	75.02	0.7	76	3	58	1
0.60	103	1	85	1	72.1	0.6
0.70	121.6	0.8	88	0.2	86.9	0.3
0.80	127	3	81.4	0.5	96	3
0.90	118	1	63.7	0.6	92	1
1.00	89.8	0.5	31.6	0.2	84	2

Table S2. Experimental solubility of vanillic acid (g/L of solvent) in 2-ethoxyethanol aqueous mixtures at 303.15K at different weight fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

Compound / w_H	2-ethoxyethanol	$\pm s$
0.00	1.44	0.01
0.05	3.06	0.08
0.10	4.81	0.04
0.20	12.0	0.3
0.30	26.9	0.9
0.40	56.0	0.6
0.50	92.8	0.6
0.60	131	1
0.70	147.0	0.2
0.80	165	1
0.90	170	2
1.00	159	4

Table S3. Experimental solubility of vanillic acid (g/L of solvent) in 1-propoxy-2-propanol aqueous mixtures at 303.15K at different weight fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

w_H	Homogeneous system solubility (g/L)	Two-phase system solubility (g/L)	
0.01	1.66 (0.02)		
0.02	2.16 (0.07)		
0.03	2.48 (0.02)		
0.05	3.75 (0.13)		
0.10	6.72 (0.05)		
0.20	22.7 (0.3)	Bottom layer	Top layer
0.30	-		
0.35	-	w_H 24.1 – 32.4 (0.5)	w_H 48.5 – 94 (6)
0.40	-		
0.50	103 (1)		
0.60	115 (1)		
0.70	128.5 (0.8)		
0.80	130 (4)		
0.90	116 (3)		
1.00	70 (1)		

Table S4. Experimental solubility of vanillic acid (g/L of solvent) in 2- butoxyethanol aqueous mixtures at 303.15K at different weight fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

w_H	Homogeneous system solubility (g/L)	Two-phase system solubility (g/L)	
0.05	3.05 (0.06)		
0.10	9.0 (0.2)	Bottom layer	Top layer
0.20	-		
0.30	-	w_H 11.8 – 13.6 (0.7)	w_H 48.3 – 78 (4)
0.40	-		
0.50	83 (1)		
0.60	103.1 (0.2)		
0.70	113.6 (0.4)		
0.80	122 (1)		
0.90	117 (1)		
1.00	76 (1)		

Table S5. Parameters of the cooperative model of hydrotropy obtained from the experimental solubility data measured in this work for syringic acid (SA) and vannilic acid (VA) in alkanediols aqueous solutions at (303.2 ± 0.5) K.

Hydrotrope	2-ethoxyethanol		2-butoxyethanol		1-propoxy -2-propanol	
solute	SA	VA	SA	VA	SA	VA
$(S/S_0)_{\max}$	101.11	121.96	31.25	92.65	75.77	103.26
m	1.66	2.00	1.59	1.63	1.53	1.59
b	5.54	7.19	6.71	6.69	6.54	6.74

APPENDIX D

S1. EXPERIMENTAL SOLUBILITY DATA

Figure S1. Chemical structure of the alkanediols used in this work as hydrotropes.

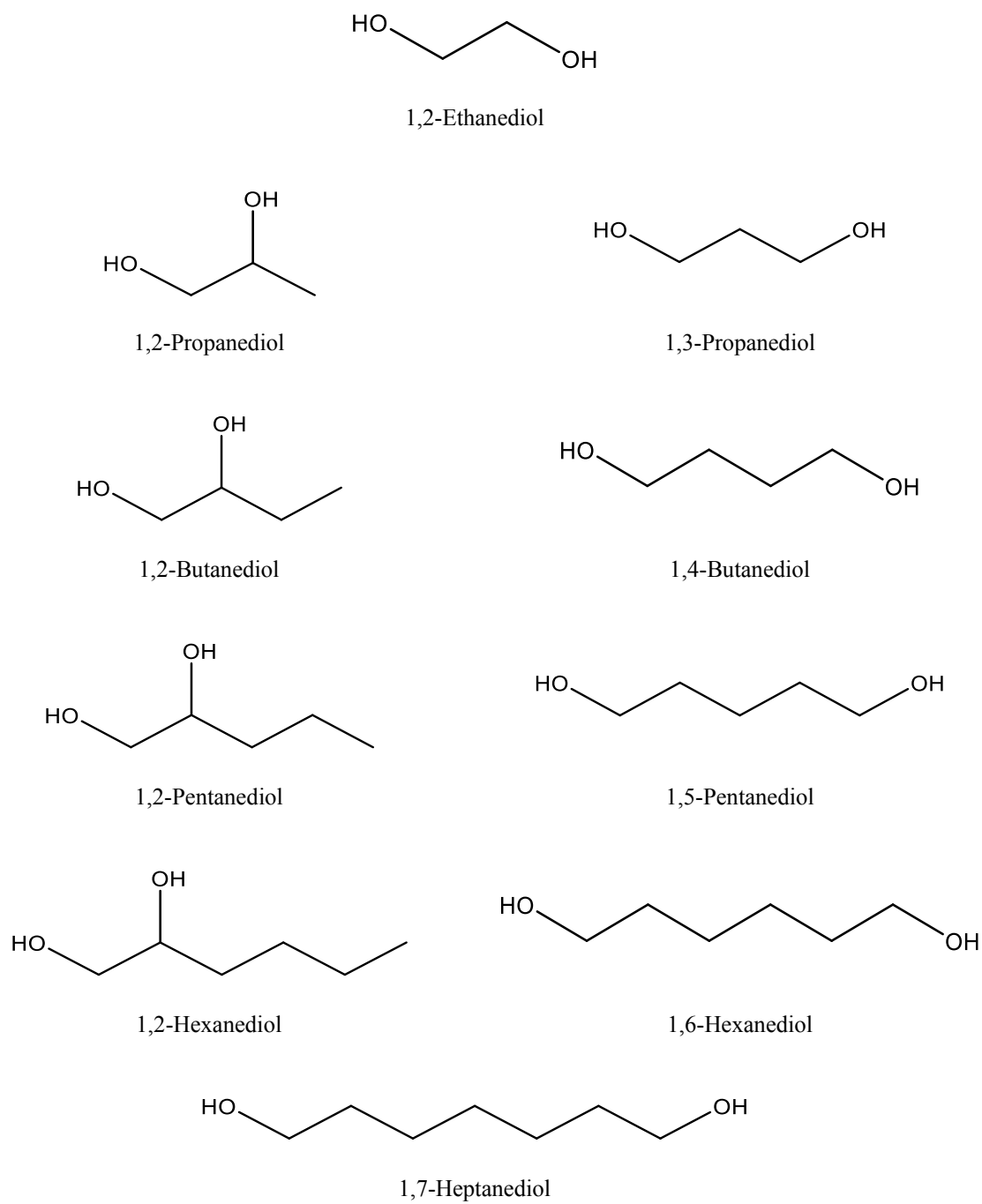


Table S1. Solubility of syringic acid (S) in aqueous solutions of 1,2-alkanediols (wt % is the weight percentage of alkanediol in water without solute), measured at (303.2 ± 0.5) K, along with the standard uncertainty of each measurement (s, obtained from three independent replicas).

1,2-Ethanediol			1,2-Propanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$	wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.08	1.47	0.03	5.00	1.89	0.05
10.01	1.72	0.01	10.03	2.35	0.004
20.37	2.44	0.01	20.03	3.78	0.04
30.13	3.62	0.06	29.97	6.32	0.07
39.97	5.41	0.03	40.00	10.79	0.07
49.84	8.30	0.25	50.02	17.35	0.03
59.65	12.04	0.13	59.85	27.25	0.57
69.70	17.93	0.13	69.83	36.19	0.76
79.62	25.76	0.02	79.91	44.69	0.60
89.55	35.87	0.67	89.96	48.39	0.32
99.90	49.38	0.36	100.00	48.62	0.89

1,2-Butanediol			1,2-Pentanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$	wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.19	2.02	0.03	5.03	2.75	0.15
10.30	2.95	0.09	10.33	4.88	0.08
20.13	6.71	0.03	20.12	14.78	0.01
30.15	13.69	0.20	29.96	29.61	0.16
40.03	25.20	0.24	40.12	43.69	0.26
49.82	37.69	0.02	50.05	55.80	0.13
59.87	48.89	0.13	59.62	64.73	0.04
69.95	56.70	0.12	69.85	73.68	0.87
79.62	58.61	0.60	79.94	72.11	0.24
89.86	52.27	0.28	89.75	60.77	1.19
100.00	39.22	0.16	100.00	41.08	1.11

1,2-Hexanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.06	3.39	0.04
10.18	7.60	0.24
20.02	15.82	0.06
30.02	23.91	0.06
39.98	32.05	0.29
50.05	40.08	0.12
60.02	47.41	0.19
69.72	52.27	0.31
79.69	53.05	0.21
89.57	46.70	1.81
100.00	35.30	0.69

Table S2. Solubility of syringic acid (S) in aqueous solutions of 1,n-alkanediols (wt % is the weight percentage of alkanediol in water without solute), measured at (303.2 ± 0.5) K, along with the standard uncertainty of each measurement (s, obtained from three independent replicas).

1,3-Propanediol			1,4-Butanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$	wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.12	1.79	0.02	5.14	2.10	0.05
10.06	2.42	0.01	10.21	3.12	0.04
20.32	3.99	0.04	19.92	6.26	0.03
30.06	6.32	0.02	30.12	11.33	0.31
39.76	9.35	0.11	39.87	17.80	0.00
49.88	14.05	0.05	50.07	26.72	0.40
59.76	20.18	0.21	59.73	36.60	0.33
69.49	27.42	0.05	69.59	45.92	0.19
80.05	35.18	0.30	79.10	52.38	0.20
89.82	42.63	0.29	89.37	54.32	0.48
100.00	50.20	0.10	100.00	52.30	0.32

1,5-Pentanediol			1,6-Hexanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$	wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.03	2.90	0.10	5.03	3.22	0.03
10.16	5.06	0.06	10.15	7.14	0.08
20.21	11.96	0.03	20.02	18.96	0.13
30.11	22.24	0.22	30.05	35.07	0.59
39.99	35.60	0.76	39.82	51.77	0.21
49.97	49.48	0.07	49.84	68.03	0.35
60.04	62.09	0.19	59.99	79.71	0.91
70.01	70.06	0.44	69.55	86.14	1.83
79.89	74.86	0.48	79.87	80.13	0.32
89.95	63.97	1.90	89.34	65.32	0.93
100.00	47.19	0.56			

1,7-Heptanediol		
wt %	$S / \text{g} \cdot \text{L}^{-1}$	$s / \text{g} \cdot \text{L}^{-1}$
5.05	4.31	0.02
10.10	10.09	0.10
20.03	26.70	0.17
30.08	45.10	0.93
39.99	60.76	0.29
49.78	77.47	0.28
59.87	95.23	1.35
69.11	96.52	0.32
78.02	93.26	1.11
89.46	76.54	2.54
100.00	38.05	1.43

Table S3. Solubility of pyrene (S) in aqueous solutions of 1,2-pentanediol or 1,5-pentanediol (wt % is the weight percentage of alkanediol in water without solute), measured at (303.2 ± 0.5) K, along with the standard uncertainty of each measurement (s, obtained from three independent replicas).

1,2-Pentanediol			1,5-Pentanediol		
wt %	$S/\text{g}\cdot\text{L}^{-1}$	$s/\text{g}\cdot\text{L}^{-1}$	wt %	$S/\text{g}\cdot\text{L}^{-1}$	$s/\text{g}\cdot\text{L}^{-1}$
20.02	0.021	0.001	20.05	0.0019	0.0001
30.05	0.194	0.003	30.04	0.031	0.004
39.99	0.573	0.017	40.11	0.126	0.009
49.98	1.200	0.010	49.87	0.331	0.004
59.96	2.003	0.011	59.84	0.744	0.019
69.92	3.436	0.003	69.83	1.514	0.021
79.82	5.393	0.054	79.99	2.824	0.154
89.90	8.007	0.190	89.86	5.90	0.093
100.00	13.091	0.394	100.00	10.141	0.760

Table S4. Solubility of Reichardt's dye (S) in aqueous solutions of 1,2-pentanediol or 1,5-pentanediol (wt % is the weight percentage of alkanediol in water without solute), measured at (303.2 ± 0.5) K, along with the standard uncertainty of each measurement (s, obtained from three independent replicas).

1,2-Pentanediol			1,5-Pentanediol		
wt %	$S/\text{g}\cdot\text{L}^{-1}$	$s/\text{g}\cdot\text{L}^{-1}$	wt %	$S/\text{g}\cdot\text{L}^{-1}$	$s/\text{g}\cdot\text{L}^{-1}$
20.03	0.39	0.02	20.08	0.089	0.002
29.87	1.34	0.06	30.02	0.301	0.005
40.00	3.80	0.07	39.99	0.503	0.03
49.97	9.47	0.20	50.06	1.028	0.05
79.98	21.21	0.83	59.95	2.148	0.036
100.00	45.58	2.96	69.77	5.404	0.264
			79.97	10.468	0.63
			89.96	16.887	0.849
			100.00	41.439	1.58

S2. SETSCHENOW CONSTANTS

Table S5. Setschenow constants for syringic acid and the alkanediols studied in this work obtained from fitting solubility data measured at (303.2 ± 0.5) K (wt % is the weight percentage range of alkanediol in water without solute used in the linear fitting).

1,2-Alkanediols			1,n-Alkanediols		
Hydrotrope	$K_S/\text{kg}\cdot\text{mol}^{-1}$	wt %	Hydrotrope	$K_S/\text{kg}\cdot\text{mol}^{-1}$	wt %
1,2-Ethanediol	0.138	0-20	1,3-Propanediol	0.374	0-20
1,2-Propanediol	0.363	0-20	1,4-Butanediol	0.659	0-20
1,2-Butanediol	0.663	0-20	1,5-Pentanediol	1.300	0-10
1,2-Pentanediol	1.206	0-20	1,6-Hexanediol	1.846	0-10
1,2-Hexanediol	1.923	0-10	1,7-Heptanediol	2.589	0-10

S3. COOPERATIVE MODEL OF HYDROTROPY

Figure S2. Left-hand-side of Equation 3.2 as a function of the natural logarithm of hydrotrope mole fraction in the water/hydrotrope/solute ternary mixture for syringic acid and the 1,2-alkanediols studied in this work at (303.2 ± 0.5) K. Dashed lines represent the best linear fitting obtained using the method of least squares. The slope and intercept of the linear fitting correspond to parameters m and b of the cooperative model of hydrotropy.

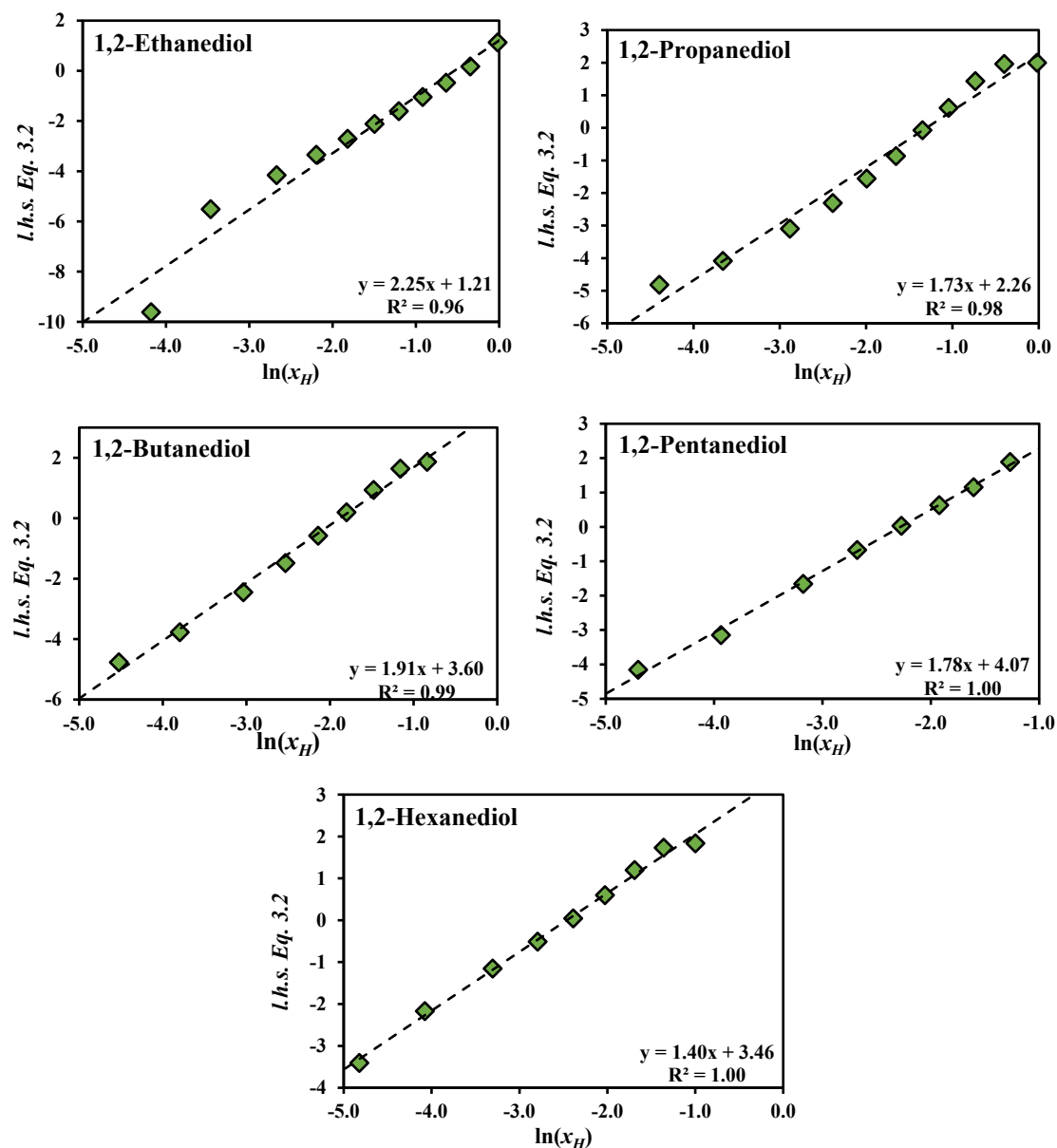


Figure S3. Left-hand-side of Equation 3.2 as a function of the natural logarithm of hydrotrope mole fraction in the water/hydrotrope/solute ternary mixture for syringic acid and the 1,n-alkanediols studied in this work at (303.2 ± 0.5) K. Dashed lines represent the best linear fitting obtained using the method of least squares. The slope and intercept of the linear fitting correspond to parameters m and b of the cooperative model of hydrotropy.

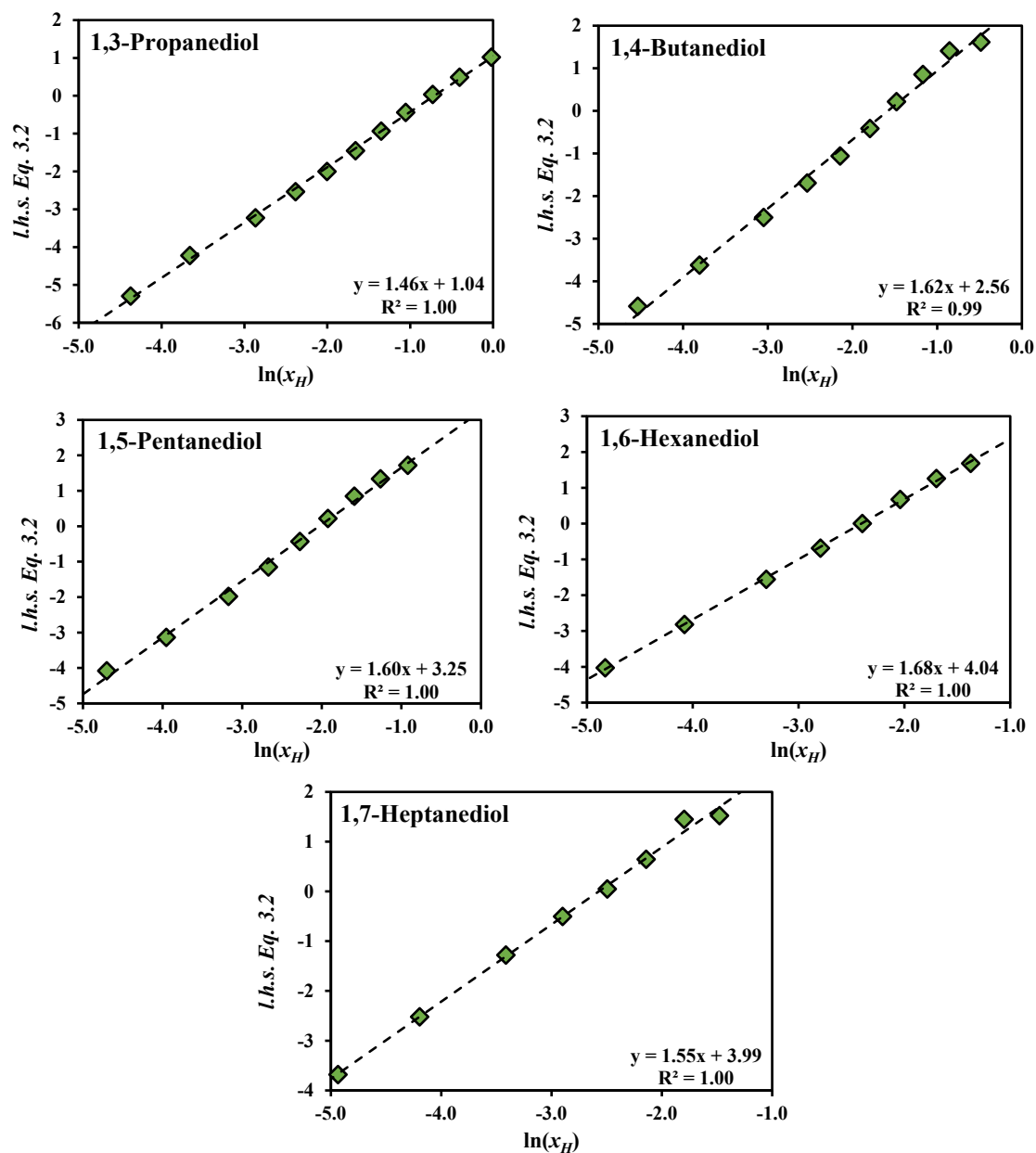


Table S6. Parameters of the cooperative model of hydrotrophy obtained by fitting Equation 3.2 to the experimental solubility data measured in this work for syringic acid and alkanediols at (303.2 ± 0.5) K.

1,2-Alkanediols				1,n-Alkanediols			
Hydrotrope	m	b	$(S/S_0)_{max}$	Hydrotrope	m	b	$(S/S_0)_{max}$
1,2-Ethenediol	2.25	1.21	44	1,3-Propanediol	1.46	1.04	46
1,2-Propanediol	1.73	2.26	38	1,4-Butanediol	1.62	2.56	44
1,2-Butanediol	1.91	3.60	46	1,5-Pentanediol	1.60	3.25	60
1,2-Pentanediol	1.78	4.07	58	1,6-Hexanediol	1.68	4.04	70
1,2-Hexanediol	1.40	3.46	42	1,7-Heptanediol	1.55	3.99	80

S4. POLARITY SCALES

Table S7. Kamlet-Taft solvatochromic parameter π^* for aqueous solutions of 1,2-propanediol and of 1,5-propanediol, experimentally measured in this work at (298.2 ± 0.5) K (wt % is the weight percentage of alkanediol in water).

1,2-Pentanediol		1,5-Pentanediol	
wt %	π^*	wt %	π^*
0.00	1.28	0.00	1.28
5.02	1.28	5.08	1.28
10.04	1.28	10.08	1.28
20.03	1.20	20.08	1.28
29.87	1.06	30.02	1.27
40.00	1.04	39.99	1.19
49.97	0.95	50.06	1.14
59.54	0.95	59.95	1.11
70.01	0.89	69.77	1.07
79.98	0.84	79.97	0.98
89.86	0.79	89.96	0.94
100.00	0.65	100.00	0.79

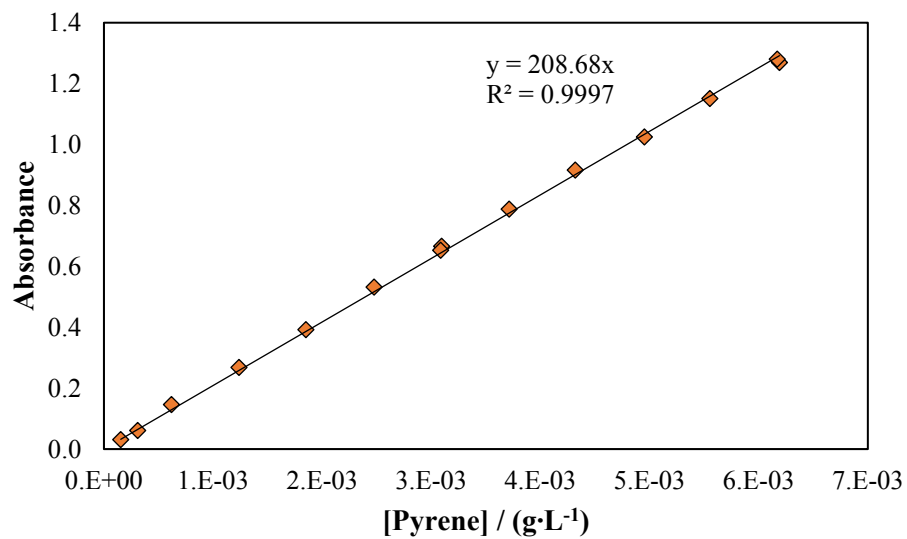
Table S8. Py scale parameter I_1/I_3 for aqueous solutions of 1,2-propanediol and of 1,5-propanediol, experimentally measured in this work at (298.2 ± 0.5) K (wt % is the weight percentage of alkanediol in water).

1,2-Pentanediol		1,5-Pentanediol	
wt %	I_1/I_3	wt %	I_1/I_3
0.00	1.892	0.00	1.892
9.95	1.752	19.99	1.581
19.97	1.400	29.96	1.479
30.07	1.316	40.06	1.494
40.07	1.274	49.95	1.462
50.06	1.251	59.96	1.493
59.92	1.265	69.86	1.480
69.93	1.247	79.67	1.494
79.89	1.239	89.75	1.458
89.78	1.233	100.00	1.384
100.00	1.203		

APPENDIX E

S1. COMPLEMENTARY FIGURE

Figure S1. Calibration curve of pyrene for UV spectra for two independent pyrene initial stock solutions at 335 nm.



S2. COMPLEMENTARY TABLES

Table S1. Experimental solubility of pyrene (g/L of solvent) in 1,2-alkanediols aqueous mixtures at 303.2 K at different mass fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

Compound/ w_H	1,2-ethanediol	$\pm s$	1,2- propanediol	$\pm s$	1,2-butanediol	$\pm s$	1,2- pentanediol	$\pm s$	1,2-hexanediol	$\pm s$
0.10	-	-	-	-	-	-	-	-	4.2E-02	1E-03
0.20	-	-	-	-	-	-	2.1E-02	1E-03	4.34E-01	8E-03
0.30	-	-	1.3E-03	2E-04	8.9E-03	6E-04	1.94E-01	3E-03	0.997	3E-03
0.40	-	-	5.54E-03	7E-05	6.6E-02	4E-03	5.7E-01	2E-02	1.70	0.03
0.50	1.433E-03	7E-06	2.4E-02	3E-03	2.44E-01	4E-03	1.20	0.01	2.54	0.01
0.60	1.84E-02	4E-04	9.1E-02	7E-03	6.2E-01	1E-02	2.00	0.01	3.69	0.02
0.70	7.1E-02	6E-03	0.268	4.E-03	1.353	5E-03	3.436	3E-03	4.96	0.06
0.80	0.222	6E-03	0.68	0.02	2.58	0.06	5.39	0.05	7.1	0.2
0.90	0.648	2E-03	1.6	0.1	5.09	0.02	8.0	0.2	10.0	0.4
0.95	1.08	0.01	2.44	0.01	6.3	0.1	10.0	0.2	13.6	0.4
1.00	1.91	0.04	4.09	0.06	9.57	0.05	13.1	0.4	15.8	0.3

Table S2. Experimental solubility of pyrene (g/L of solvent) in 1,n-alkanediols aqueous mixtures at 303.2 K at different mass fractions (w_H) of hydrotrope under 0.101 MPa and their correspondent standard deviation s (g/L of solvent).

Compound/ w_H	1,3- propanediol	$\pm s$	1,4-butanediol	$\pm s$	1,5- pentanediol	$\pm s$	1,6-hexanediol	$\pm s$	1,7- heptanediol	$\pm s$
0.125	-	-	-	-	-	-	-	-	2.98E-02	1E-04
0.20	-	-	-	-	1.93E-03	9E-05	3.06E-02	1E-04	0.221	8E-03
0.30	1.39E-03	3E-05	-	-	0.033	1E-03	0.145	1E-03	0.65	0.02
0.40	5.3E-03	2E-04	2.06E-03	1E-05	0.126	9E-03	0.41	0.01	1.38	0.05
0.50	0.0114	2E-04	0.0142	7E-04	0.331	4E-03	0.86	0.04	2.38	0.05
0.60	0.0504	2E-04	0.0577	2E-04	0.74	0.02	1.66	0.03	3.71	0.05
0.70	0.17	0.01	0.27	0.01	1.51	0.02	2.9	0.1	5.74	0.07
0.80	0.47	0.01	0.91	0.04	2.8	0.2	4.1	0.1	9.5	0.3
0.90	1.18	0.01	2.45	0.02	5.90	0.09	7.7	0.3	15.1	0.4
0.95	2.050	0.03	4.1	0.10	8.1	0.1	-	-	16.8	0.3
1.00	3.48	4E-03	6.709	6E-03	10.1	0.8	-	-	21.4	0.6

Table S3. Parameters of the cooperative model of hydrotropy obtained from the experimental solubility data measured in this work for pyrene in alkanediols aqueous solutions at (303.2 ± 0.5) K.

1,2-Alkanediols				1,n-Alkanediols			
Hydrotrope	m	b	$(S/S_0)_{max}$	Hydrotrope	m	b	$(S/S_0)_{max}$
1,2-Ethanediol	2.65	1.06	15698.88	1,3-Propanediol	2.70	2.37	22077.30
1,2-Propanediol	2.71	3.03	24060.50	1,4-Butanediol	2.66	1.53	42358.75
1,2-Butanediol	2.29	2.01	56287.30	1,5-Pentanediol	2.05	3.65	59618.62
1,2-Pentanediol	1.44	2.78	76961.89	1,6-Hexanediol	1.82	4.39	58581.51
1,2-Hexanediol	1.18	2.74	93143.08	1,7-Heptanediol	1.48	3.68	125681.53

Table S4. Kamlet-Taft solvatochromic parameter π^* for aqueous solutions of 1,2-alkanediols, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,2-ethanediol	1,2-propanediol	1,2-butanediol	1,2-pentanediol	1,2-hexanediol
0.00	1.28				
0.10	1.27	1.28	1.28	1.28	1.06
0.20	1.27	1.28	1.28	1.19	0.93
0.30	1.27	1.28	1.27	1.06	0.91
0.40	1.25	1.23	1.19	1.03	0.90
0.50	1.22	1.20	1.10	0.98	0.88
0.60	1.18	1.15	1.06	0.94	0.86
0.70	1.15	1.10	1.01	0.91	0.84
0.80	1.10	1.03	0.95	0.86	0.80
0.90	1.04	0.95	0.87	0.80	0.75
0.95	1.01	0.86	0.82	0.73	0.68
1.00	0.99	0.82	0.75	0.68	0.62

Table S5. Kamlet-Taft solvatochromic parameter π^* for aqueous solutions of 1,n-alkanediols, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,3-propanediol	1,4-butanediol	1,5-pentanediol	1,6-hexanediol	1,7-heptanediol
0.10	1.27	1.28	1.28	1.27	1.24
0.20	1.27	1.25	1.28	1.23	1.14
0.30	1.27	1.25	1.23	1.18	1.11
0.40	1.23	1.23	1.17	1.14	1.06
0.50	1.22	1.18	1.15	1.10	1.04
0.60	1.18	1.15	1.11	1.08	1.03
0.70	1.13	1.10	1.06	1.04	0.99
0.80	1.08	1.05	1.03	1.00	0.96
0.90	1.03	0.98	0.95	0.93	0.91
0.95	0.97	0.93	0.90	0.84	0.83
1.00	0.92	0.88	0.80	-	0.72

Table S6. *Py* scale parameter I_1/I_3 for aqueous solutions of 1,2-alkanediol, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,2-ethanediol	1,2-propanediol	1,2-butanediol	1,2-pentanediol	1,2-hexanediol
0.00	1.94				
0.05	-	-	-	1.86	-
0.08	-	-	-	-	1.30
0.10	-	-	-	1.78	1.14
0.20	-	-	-	1.48	1.12
0.30	-	-	1.54	1.33	1.12
0.40	-	1.73	1.49	1.30	1.11
0.50	1.83	1.67	1.44	1.27	1.13
0.60	1.79	1.64	1.41	1.27	1.13
0.70	1.77	1.57	1.38	1.26	1.14
0.80	1.70	1.50	1.36	1.26	1.16
0.90	1.66	1.45	1.33	1.24	1.16
0.95	1.64	1.42	1.33	1.24	1.15
1.00	1.60	1.42	1.30	1.23	1.14

Table S7. *Py* scale parameter I_1/I_3 for aqueous solutions of 1,n-alkanediol, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,3-propanediol	1,4-butanediol	1,5-pentanediol	1,6-hexanediol
0.10	-	-	-	1.66
0.20	1.89	-	1.63	1.57
0.30	1.84	1.75	1.58	1.53
0.40	1.81	1.69	1.56	1.50
0.50	1.75	1.65	1.54	1.48
0.60	1.71	1.62	1.52	1.46
0.70	1.68	1.60	1.51	1.43
0.80	1.62	1.54	1.48	1.40
0.90	1.57	1.50	1.45	1.34
0.95	-	1.45	1.41	1.31
1.00	1.54	1.44	1.38	1.27

Table S8. Local composition (x_L) form Py scale for aqueous solutions of 1,2-alkanediol, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,2-ethanediol	1,2-propanediol	1,2-butanediol	1,2-pentanediol	1,2-hexanediol
0.05	-	-	-	0.08	-
0.08	-	-	-	-	0.79
0.10	-	-	-	0.17	1.00
0.20	-	-	-	0.57	1.02
0.30	-	-	0.47	0.81	1.02
0.40	-	0.24	0.56	0.87	1.03
0.50	0.23	0.33	0.66	0.91	1.01
0.60	0.32	0.39	0.73	0.92	1.01
0.70	0.39	0.52	0.80	0.93	1.00
0.80	0.61	0.71	0.84	0.94	0.98
0.90	0.72	0.88	0.90	0.96	0.98
0.95	0.83	0.99	0.92	0.98	0.99
1.00	1.00	1.00	1.00	1.00	1.00

Table S9. Local composition (x_L) form Py scale for aqueous solutions of 1,n-alkanediol, at different mass fractions (w_H), measured in this work at (298.2 ± 0.5) K.

Compound/ w_H	1,3-propanediol	1,4-butanediol	1,5-pentanediol	1,6-hexanediol
0.10	-	-	-	0.33
0.20	0.07	-	0.39	0.47
0.30	0.15	0.22	0.47	0.52
0.40	0.21	0.31	0.51	0.58
0.50	0.33	0.39	0.56	0.61
0.60	0.42	0.45	0.61	0.65
0.70	0.51	0.50	0.64	0.69
0.80	0.70	0.65	0.70	0.75
0.90	0.87	0.76	0.79	0.86
0.95	-	0.95	0.90	0.93
1.00	1.00	1.00	1.00	1.00

APPENDIX F

S1. COMPLEMENTARY FIGURES

Figure S1. Sigma surface and total energy for different conformers of 1,2-ethanediol obtained from CosmoThermX using BP_TZVP_21 parametrization.

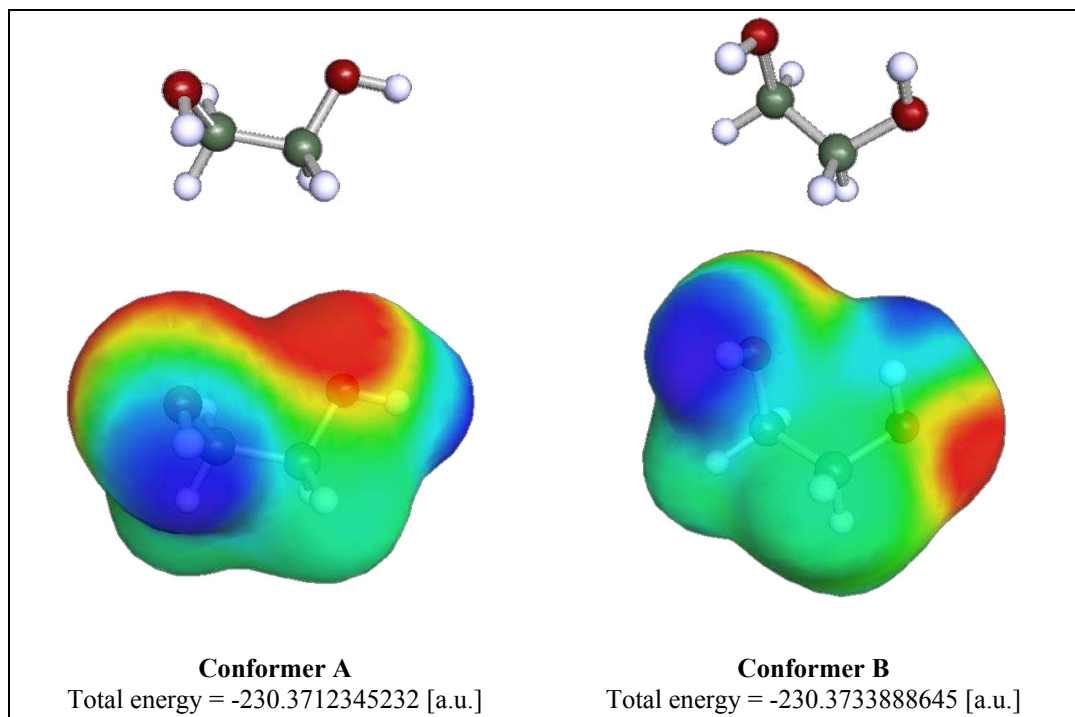


Figure S2. Sigma surface and total energy for different conformers of 1,2-propnaediol obtained from CosmoThermX using BP_TZVP_21 parametrization.

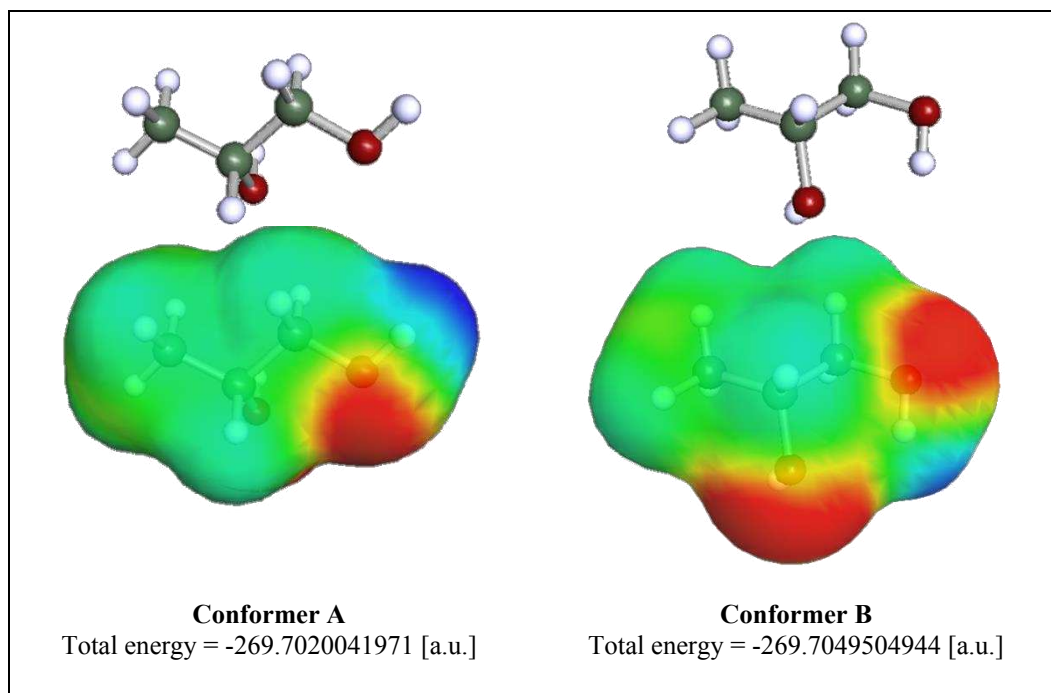


Figure S3. Sigma surface and total energy for different conformers of 1,2-pentanediol obtained from CosmoThermX using BP_TZVP_21 parametrization.

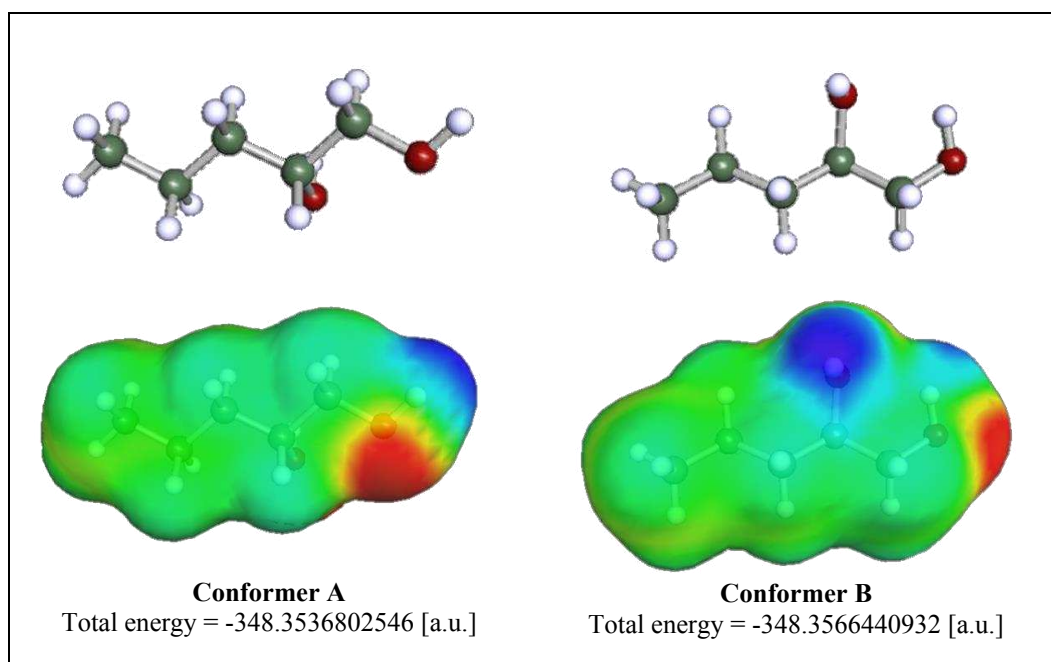


Figure S4. Sigma surface and total energy for different conformers of 1,2-hexanediol obtained from CosmoThermX using BP_TZVP_21 parametrization.

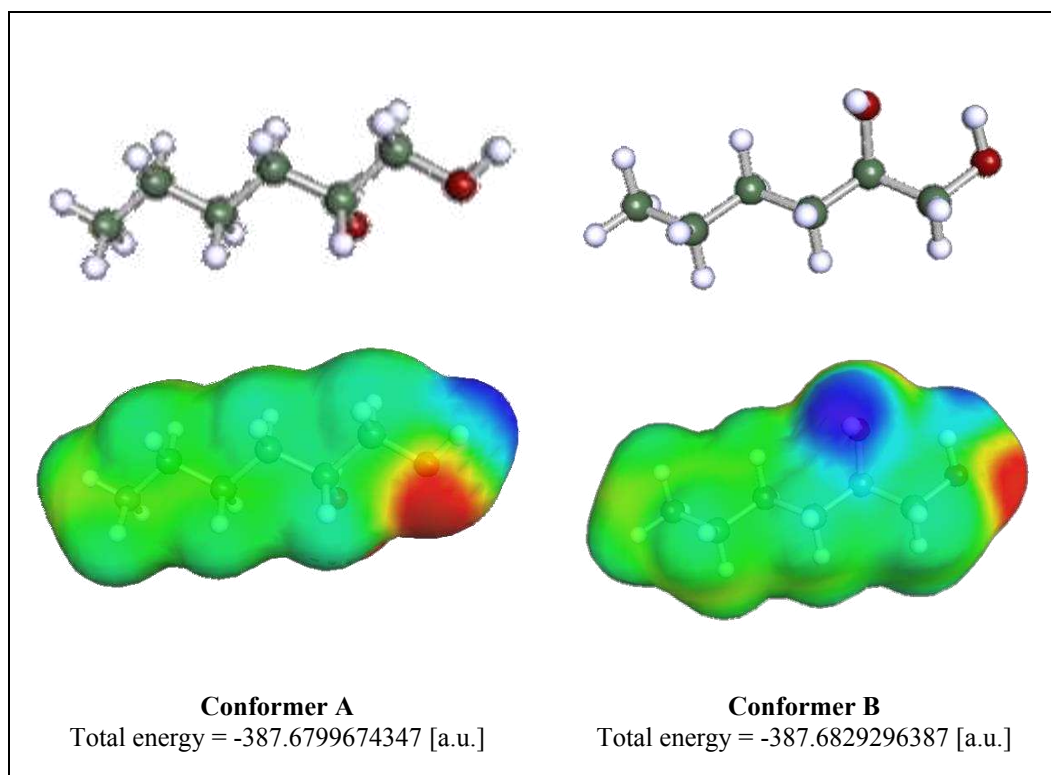
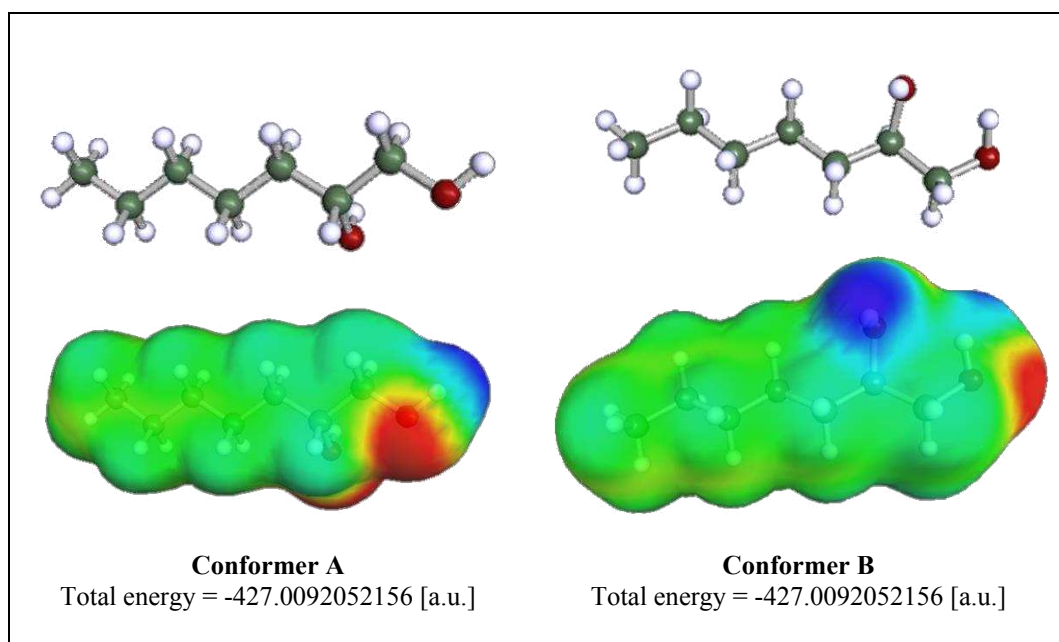


Figure S5. Sigma surface and total energy for different conformers of 1,2-heptanediol obtained from CosmoThermX using BP_TZVP_21 parametrization.



APPENDIX G

S1. COMPLEMENTARY FIGURES

Figure S1. Calibration curve of syringic acid for UV spectra for two independent syringic acid initial stock solutions.

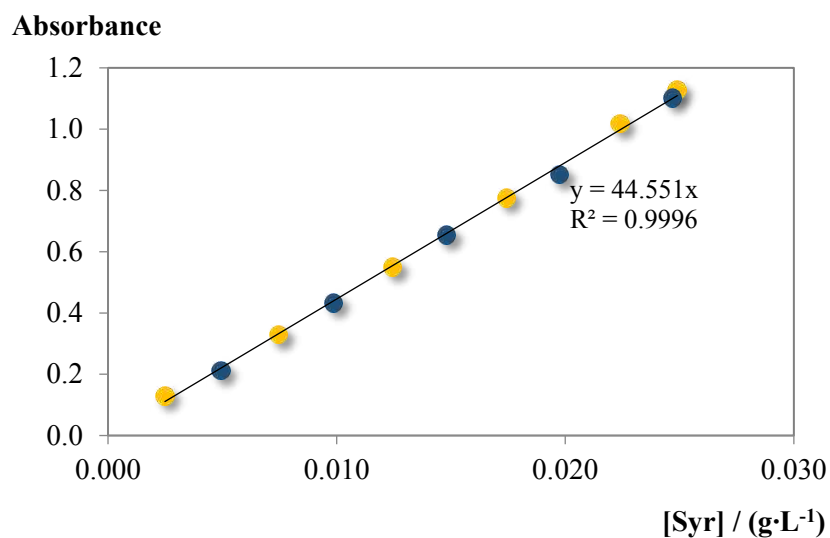


Figure S2. $\ln x_1$ vs. $(1/T - 1/T_{mh})$ for syringic acid in aqueous solutions of 1,2-propanediol at 0.0 $\blacklozenge$, 0.05 $\blacklozenge$, 0.1 $\blacklozenge$, 0.2 $\blacklozenge$, 0.3 $\blacklozenge$, 0.4 $\blacklozenge$, 0.5 $\blacklozenge$, 0.6 $\blacklozenge$, 0.7 $\blacklozenge$, 0.8 $\blacklozenge$, 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.

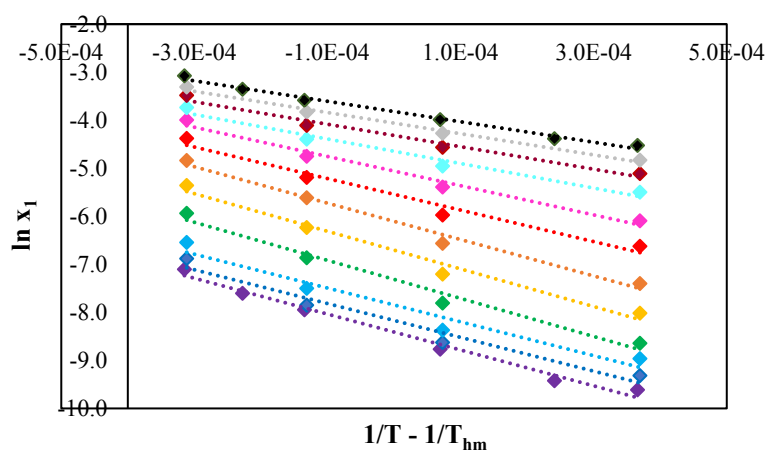


Figure S3. $\ln x_1$ vs. $(1/T-1/T_{mh})$ for syringic acid in aqueous solutions of 1,3-butanediol at 0.0 $\blacklozenge$; 0.05 $\blacklozenge$; 0.1 $\blacklozenge$; 0.2 $\blacklozenge$; 0.3 $\blacklozenge$; 0.4 $\blacklozenge$; 0.5 $\blacklozenge$; 0.6 $\blacklozenge$; 0.7 $\blacklozenge$; 0.8 $\blacklozenge$; 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.

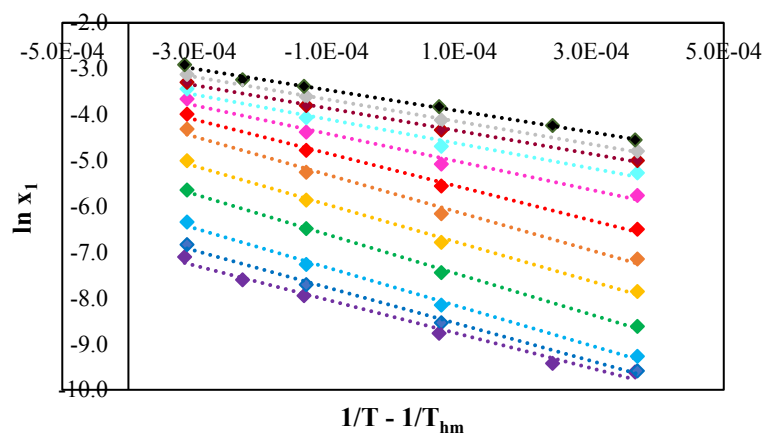


Figure S4. $\ln x_1$ vs. $(1/T-1/T_{mh})$ for syringic acid in aqueous solutions of 1,2-pentanediol at 0.0 $\blacklozenge$; 0.05 $\blacklozenge$; 0.1 $\blacklozenge$; 0.2 $\blacklozenge$; 0.3 $\blacklozenge$; 0.4 $\blacklozenge$; 0.5 $\blacklozenge$; 0.6 $\blacklozenge$; 0.7 $\blacklozenge$; 0.8 $\blacklozenge$; 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.

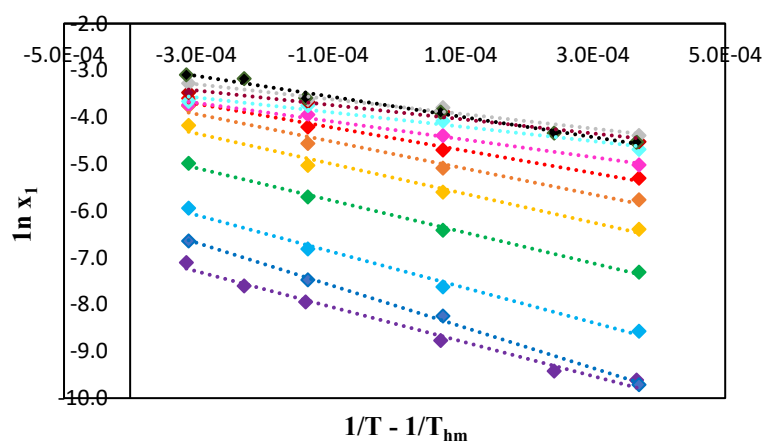


Figure S5. $\ln x_1$ vs. $(1/T-1/T_{mh})$ for syringic acid in aqueous solutions of [1.0.0] at 0.0 $\blacklozenge$; 0.05 $\blacklozenge$; 0.1 $\blacklozenge$; 0.2 $\blacklozenge$; 0.3 $\blacklozenge$; 0.4 $\blacklozenge$; 0.5 $\blacklozenge$; 0.6 $\blacklozenge$; 0.7 $\blacklozenge$; 0.8 $\blacklozenge$; 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.

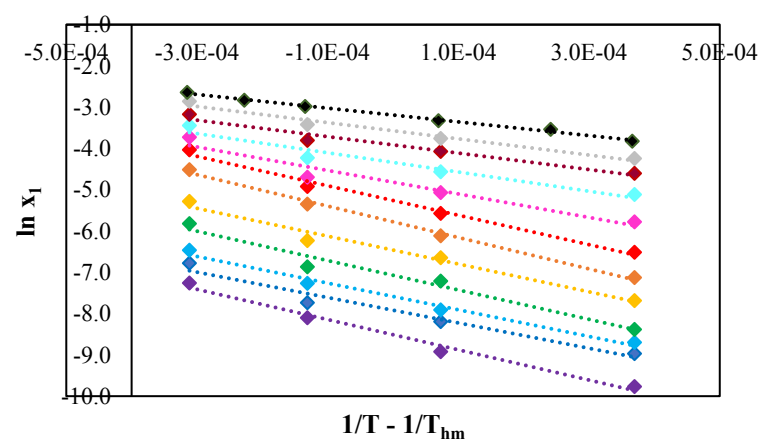


Figure S6. $\ln x_1$ vs. $(1/T-1/T_{mh})$ for syringic acid in aqueous solutions of 2-ethoxyethanol at 0.0 $\blacklozenge$; 0.05 $\blacklozenge$; 0.1 $\blacklozenge$; 0.2 $\blacklozenge$; 0.3 $\blacklozenge$; 0.4 $\blacklozenge$; 0.5 $\blacklozenge$; 0.6 $\blacklozenge$; 0.7 $\blacklozenge$; 0.8 $\blacklozenge$; 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.

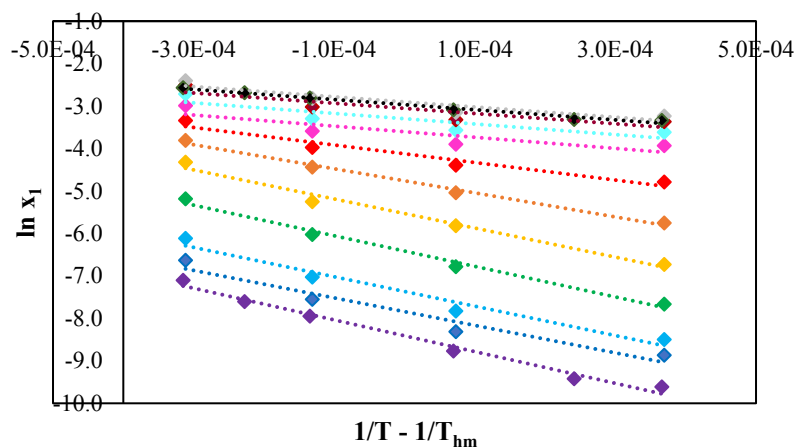
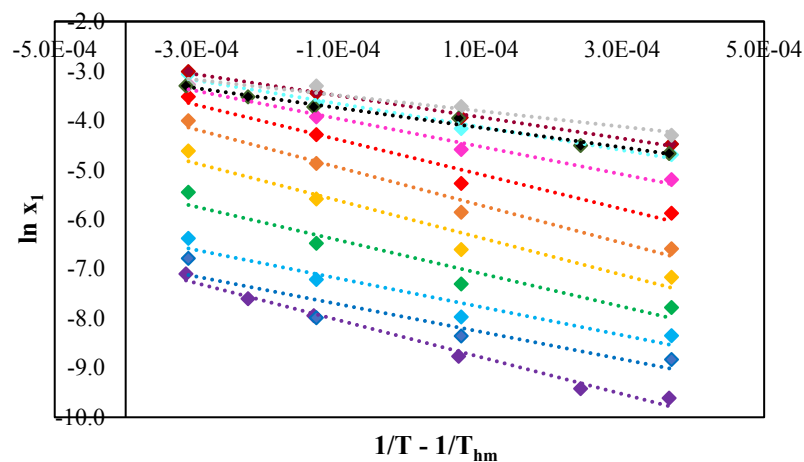


Figure S7. $\ln x_1$ vs. $(1/T-1/T_{mh})$ for syringic acid in aqueous solutions of cyrene at 0.0 $\blacklozenge$; 0.05 $\blacklozenge$; 0.1 $\blacklozenge$; 0.2 $\blacklozenge$; 0.3 $\blacklozenge$; 0.4 $\blacklozenge$; 0.5 $\blacklozenge$; 0.6 $\blacklozenge$; 0.7 $\blacklozenge$; 0.8 $\blacklozenge$; 0.9 $\blacklozenge$ and 1.0 $\blacklozenge$ weight fractions.



S2. COMPLEMENTARY TABLES

Table S1. Experimental solubility (g/L of solvent) of syringic acid in the various hydrotrope solutions at different temperatures and hydrotrope weight fractions (w_H) at 0.101 MPa

Temperature / K	1,2- propanediol	1,3-butanediol	1,2-pentanediol	[1.0.0]	2- ethoxyethanol	cyrene
$w_H = 0$	g/L					
278.2	0.632 (0.018)					
288.2	0.763 (0.003)					
303.2	1.462 (0.004)					
323.2	3.297 (0.020)					
333.2	4.616 (0.045)					
343.2	7.555 (0.059)					
$w_H = 0.05$	g/L					
278.2	0.953 (0.038)	0.723 (0.110)	0.636 (0.000)	1.350 (0.050)	1.480 (0.050)	1.550 (0.010)
303.2	1.891 (0.052)	2.058 (0.018)	2.745 (0.23)	2.918 (0.042)	2.568 (0.037)	2.480 (0.059)
323.2	4.074 (0.095)	4.676 (0.034)	5.881 (0.008)	4.570 (0.20)	5.430 (0.130)	3.560 (0.450)
343.2	10.586 (0.195)	11.021 (0.397)	13.322 (0.206)	11.720 (0.140)	13.450 (0.210)	11.650 (0.320)
$w_H = 0.1$	g/L					
278.2	1.306 (0.268)	0.954 (0.044)	1.905 (0.147)	1.700 (0.120)	2.050 (0.010)	2.420 (0.020)
303.2	2.348 (0.004)	2.896 (0.012)	4.885 (0.075)	3.711 (0.010)	3.975 (0.067)	3.520 (0.079)
323.2	5.539 (0.056)	6.956 (0.060)	10.830 (0.270)	7.010 (0.110)	8.750 (0.210)	7.410 (0.520)
343.2	14.194 (0.135)	17.194 (0.127)	25.443 (0.524)	15.420 (0.070)	21.410 (0.20)	16.820 (0.650)
$w_H = 0.2$	g/L					
278.2	1.658 (0.000)	1.671 (0.022)	6.105 (0.179)	2.140 (0.130)	4.270 (0.320)	3.960 (0.010)
303.2	3.785 (0.036)	5.355 (0.026)	14.775 (0.008)	6.846 (0.118)	10.185 (0.063)	6.320 (0.198)
323.2	9.629 (0.190)	13.810 (0.690)	29.472 (0.698)	9.550 (0.230)	21.350 (0.350)	14.170 (0.590)

343.2	23.793 (0.675)	31.335 (0.032)	58.753 (2.952)	26.720 (1.040)	48.280 (0.130)	39.030 (1.230)
w_H = 0.3	g/L					
278.2	2.828 (0.103)	3.237 (0.030)	13.670 (0.889)	3.920 (0.240)	9.750 (0.030)	6.680 (0.190)
303.2	6.316 (0.067)	9.363 (0.044)	29.607 (0.29)	10.939 (0.442)	23.830 (0.454)	11.550 (0.595)
323.2	16.431 (0.127)	23.120 (0.381)	51.295 (1.881)	16.340 (0.360)	41.100 (1.850)	31.500 (0.330)
343.2	38.473 (0.190)	53.040 (1.873)	115.037 (0.794)	41.360 (0.080)	99.480 (1.760)	80.360 (1.900)
w_H = 0.4	g/L					
278.2	4.749 (0.248)	5.870 (0.325)	22.682 (0.429)	6.20 (0.520)	22.880 (0.430)	10.730 (0.060)
303.2	10.791 (0.071)	15.589 (0.127)	43.686 (0.262)	16.678 (0.053)	45.491 (0.989)	22.033 (0.040)
323.2	27.392 (1.278)	37.592 (0.214)	72.007 (2.920)	35.241 (1.340)	80.340 (4.630)	57.350 (1.450)
343.2	57.771 (0.040)	92.366 (2.063)	151.568 (4.841)	78.430 (0.950)	144.070 (2.080)	130.190 (1.590)
w_H = 0.5	g/L					
278.2	9.183 (0.242)	9.916 (0.000)	31.256 (2.047)	10.040 (0.140)	51.900 (0.660)	19.460 (0.020)
303.2	17.351 (0.032)	25.028 (0.032)	55.801 (0.127)	25.340 (0.085)	75.015 (0.697)	34.790 (0.396)
323.2	37.148 (0.349)	53.23 (0.571)	88.438 (0.238)	47.350 (0.808)	110.450 (3.190)	89.780 (2.090)
343.2	80.694 (0.29)	111.782 (1.270)	155.440 (30.355)	110.320 (0.020)	195.640 (0.320)	182.600 (5.870)
w_H = 0.6	g/L					
278.2	13.748 (0.286)	17.923 (2.539)	35.588 (1.476)	18.170 (1.630)	102.523 (0.730)	33.150 (0.130)
303.2	27.250 (0.571)	34.881 (0.238)	64.729 (0.040)	36.138 (0.147)	104.150 (1.010)	59.310 (0.793)
323.2	50.437 (0.032)	68.265 (0.000)	98.314 (0.635)	51.342 (0.613)	137.340 (1.120)	110.340 (1.870)
343.2	103.309 (1.508)	133.779 (0.000)	118.544 (11.309)	128.840 (0.820)	234.010 (1.130)	200.890 (3.170)
w_H = 0.7	g/L					
278.2	21.481 (0.524)	24.730 (2.452)	41.301 (3.746)	29.620 (1.520)	118.110 (0.640)	46.100 (0.150)
303.2	36.195 (0.762)	43.344 (0.476)	73.680 (0.873)	50.220 (0.119)	121.621 (0.827)	75.760 (1.387)
323.2	61.952 (1.841)	78.730 (0.397)	100.054 (1.190)	68.798 (0.476)	154.340 (0.366)	120.310 (3.490)
343.2	114.756 (2.143)	140.850 (0.794)	107.330 (7.574)	143.430 (1.590)	258.360 (4.470)	208.690 (0.950)
w_H = 0.8	g/L					

278.2	26.515 (0.881)	26.481 (0.595)	39.258 (0.984)	39.970 (3.070)	123.340 (1.340)	45.890 (0.100)
303.2	44.690 (0.603)	50.644 (0.595)	72.108 (0.238)	66.254 (0.366)	126.522 (2.886)	76.260 (1.585)
323.2	68.887 (1.936)	83.949 (0.476)	87.652 (2.222)	84.660 (0.850)	165.420 (0.160)	121.560 (3.240)
343.2	123.959 (0.238)	134.116 (1.428)	104.431 (7.380)	152.450 (0.100)	260.120 (1.280)	180.250 (1.430)
w_H = 0.9	g/L					
278.2	28.350 (1.333)	25.572 (0.310)	34.410 (3.016)	43.810 (3.230)	110.430 (1.590)	41.040 (0.970)
303.2	48.394 (0.317)	49.354 (0.198)	60.773 (1.190)	69.210 (0.162)	117.917 (1.296)	71.218 (0.343)
323.2	73.511 (0.286)	80.470 (0.000)	71.098 (0.397)	94.798 (3.115)	158.580 (1.920)	105.349 (4.284)
343.2	119.863 (0.000)	126.148 (0.635)	98.090 (1.428)	160.450 (0.970)	230.250 (7.030)	106.170 (1.380)
w_H = 1.0	g/L					
278.2	29.158 (1.333)	23.217 (0.577)	19.932 (0.190)	45.290 (0.159)	71.977 (1.130)	18.282 (0.429)
288.2	33.220 (0.842)	31.571 (0.447)	24.030 (0.343)	59.763 (0.238)	73.090 (0.992)	21.277 (0.518)
303.2	48.618 (0.889)	46.997 (0.516)	37.110 (1.111)	72.669 (0.421)	89.785 (0.489)	36.420 (1.387)
323.2	71.110 (1.270)	71.547 (0.397)	49.157 (1.203)	100.222 (0.959)	115.458 (1.190)	44.910 (0.340)
333.2	88.214 (0.405)	81.667 (1.465)	73.510 (0.284)	115.317 (2.790)	129.140 (0.648)	54.488 (0.361)
343.2	114.532 (0.397)	111.221 (0.159)	79.067 (2.619)	137.202 (0.959)	142.814 (1.440)	67.409 (0.099)

Table S2. Experimental solubility of vanillic acid (g/L of solvent) in 2-ethoxyethanol and cyrene/water mixtures at 303.2K and different weight fractions (w_H) of hydrotrope under 0.101 MPa.

Compound	2-ethoxyethanol		Cyrene	
w _H	Solubility (g/L)	σ (g/L)	Solubility (g/L)	σ (g/L)
0.00	1.435	0.012	1.435	0.012
0.05	3.058	0.079	2.315	0.018
0.10	4.810	0.039	3.344	0.055
0.20	11.985	0.265	5.419	0.203
0.30	26.897	0.888	9.051	0.123
0.40	56.015	0.576	16.768	0.234
0.50	92.753	0.593	30.431	0.469
0.60	130.881	1.199	54.777	0.617
0.70	147.013	0.247	78.895	1.480
0.80	165.091	1.121	97.695	0.049
0.90	169.509	1.846	87.998	0.247
1.00	159.017	4.494	31.739	0.296

Table S3. Density temperature correlations $\rho(T)$ (in g.L⁻¹ and T in Kelvin) for the six selected hydrotropes, and the corresponding determination coefficient (R^2).

Compound	Reference	Correlation $\rho(T)$ /g.L ⁻¹	R^2
1,2-propanediol	(GEORGE; SASTRY, 2003; LI; SU; WANG, 2007)	$\rho = -1.10 \cdot 10^{-3} T^2 - 6.89 \cdot 10^{-3} T + 1.15 \cdot 10^3$	0.999
1,3-butanediol	(GEORGE; SASTRY, 2003; NAIN, 2007)	$\rho = -4.74 \cdot 10^{-3} T^2 + 2.31 T + 7.34 \cdot 10^2$	0.997
1,2-pentanediol	(PARSA; HAGHRO, 2008)	$\rho = -6.25 \cdot 10^{-4} T^2 - 3.56 \cdot 10^{-1} T + 1.13 \cdot 10^3$	0.999
[1.0.0]	(LEAL-DUASO <i>et al.</i> , 2017)	$\rho = -8.08 \cdot 10^{-1} T + 1.35 \cdot 10^3$	1.000
2-ethoxyethanol	(SHI <i>et al.</i> , 2019)	$\rho = -8.53 \cdot 10^{-4} T^2 - 3.93 \cdot 10^{-1} T + 1.12 \cdot 10^3$	1.000
cyrene	(BAIRD <i>et al.</i> , 2019)	$\rho = -6.30 \cdot 10^{-5} T^2 - 9.39 \cdot 10^{-1} T + 1.53 \cdot 10^3$	1.000

Table S4. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in 1,2-propanediol aqueous solutions ($p = 101.3$ kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.00	0.9866	22.04 (0.38)	31.04 (5.03)	29.04 (14.99)	0.78	0.22
0.05	0.9634	21.05 (1.36)	29.26 (17.35)	26.46 (51.57)	0.78	0.22
0.10	0.9518	20.04 (1.57)	29.26 (20.04)	29.17 (59.57)	0.76	0.24
0.20	0.9792	18.85 (1.15)	32.94 (14.60)	45.44 (43.40)	0.70	0.30
0.30	0.9797	17.28 (1.12)	32.57 (14.27)	49.32 (42.43)	0.68	0.32
0.40	0.9859	15.73 (0.90)	31.51 (11.45)	50.88 (34.04)	0.67	0.33
0.50	0.9718	14.28 (1.11)	27.28 (14.14)	41.91 (42.02)	0.68	0.32
0.60	0.9824	13.04 (0.81)	25.29 (10.29)	39.53 (30.58)	0.67	0.33
0.70	0.9747	11.98 (0.82)	21.23 (10.42)	29.85 (30.96)	0.70	0.30
0.08	0.9775	11.13 (0.70)	19.44 (8.98)	26.8 (26.68)	0.70	0.30
0.09	0.9902	10.46 (0.44)	18.33 (5.56)	25.38 (16.53)	0.70	0.30
1.00	0.9897	9.83 (0.19)	17.70 (2.51)	25.40 (7.49)	0.69	0.31

Table S5. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in 1,3-butanediol aqueous solutions ($p = 101.3$ kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.05	0.9950	21.05 (0.57)	33.35 (7.21)	39.68 (21.44)	0.73	0.27
0.10	0.9951	20.00 (0.59)	35.46 (7.58)	49.88 (22.53)	0.70	0.30
0.20	0.9979	18.17 (0.39)	36.38 (5.03)	58.75 (14.95)	0.67	0.33
0.30	0.9953	16.44 (0.57)	34.79 (7.24)	59.19 (21.51)	0.65	0.35
0.40	0.9889	14.75 (0.87)	34.36 (11.05)	63.28 (32.85)	0.64	0.36
0.50	0.9944	13.42 (0.54)	30.46 (6.93)	54.97 (20.59)	0.64	0.36
0.60	0.9830	12.17 (0.80)	25.54 (10.22)	43.13 (30.37)	0.66	0.34
0.70	0.9811	11.26 (0.74)	22.28 (9.40)	35.54 (27.93)	0.67	0.33
0.08	0.9973	10.59 (0.26)	20.78 (3.29)	32.86 (9.77)	0.67	0.33
0.09	0.9986	10.08 (0.18)	20.36 (2.30)	33.14 (6.83)	0.66	0.34
1.00	0.9953	9.51 (0.14)	19.18 (1.83)	31.22 (5.45)	0.66	0.34

Table S6. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in 1,2-pentamediol aqueous solutions ($p = 101.3$ kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.05	0.9973	20.67 (0.46)	37.12 (5.85)	53.08 (17.40)	0.69	0.31
0.10	0.9909	18.65 (0.73)	31.83 (9.30)	42.50 (27.66)	0.71	0.29
0.20	0.9960	15.73 (0.43)	28.19 (5.45)	40.18 (16.19)	0.69	0.31
0.30	0.9803	13.66 (0.89)	26.36 (11.36)	40.97 (33.78)	0.67	0.33
0.40	0.9742	12.35 (0.92)	23.79 (11.77)	36.90 (35.00)	0.68	0.32
0.50	0.9858	11.48 (0.59)	20.48 (7.47)	29.06 (22.21)	0.69	0.31
0.60	0.9886	11.02 (0.41)	16.07 (5.24)	16.29 (15.57)	0.76	0.24
0.70	0.9546	10.43 (0.67)	12.94 (8.59)	8.12 (25.53)	0.84	0.16
0.08	0.9646	10.02 (0.58)	12.72 (7.41)	8.72 (22.04)	0.82	0.18
0.09	0.9805	9.71 (0.44)	13.13 (5.64)	11.01 (16.76)	0.79	0.21
1.00	0.9848	9.71 (0.24)	18.13 (3.13)	27.19 (9.34)	0.68	0.32

Table S7. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in [1.0.0] aqueous solutions (p = 101.3 kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.05	0.9637	20.41 (1.18)	25.56 (15.09)	16.62 (44.85)	0.83	0.17
0.10	0.9856	19.55 (0.77)	26.85 (9.87)	23.57 (29.34)	0.79	0.21
0.20	0.9689	18.23 (1.27)	29.60 (16.14)	36.69 (47.98)	0.72	0.28
0.30	0.9775	16.65 (1.01)	28.04 (12.93)	36.72 (38.45)	0.71	0.29
0.40	0.9953	14.89 (0.52)	31.63 (6.64)	54.01 (19.74)	0.65	0.35
0.50	0.9887	13.56 (0.76)	29.65 (9.63)	51.91 (28.62)	0.65	0.35
0.60	0.9468	12.41 (1.34)	23.65 (17.06)	36.26 (50.70)	0.68	0.32
0.70	0.9479	11.18 (1.08)	19.36 (13.80)	26.39 (41.03)	0.70	0.30
0.08	0.9625	10.08 (0.78)	16.58 (9.95)	20.95 (29.59)	0.72	0.28
0.09	0.9736	9.20 (0.64)	16.32 (8.18)	22.97 (24.31)	0.70	0.30
1.00	0.9948	8.21 (0.11)	13.81 (1.39)	18.07 (4.14)	0.71	0.29

Table S8. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in 2-ethoxyethanol aqueous solutions (p = 101.3 kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.05	0.9495	20.21 (1.48)	26.91 (18.88)	21.62 (56.11)	0.80	0.20
0.10	0.9657	18.99 (1.29)	28.77 (16.49)	31.56 (49.02)	0.75	0.25
0.20	0.9891	16.53 (0.75)	29.97 (9.57)	43.33 (28.45)	0.69	0.31
0.30	0.9821	14.25 (0.92)	28.47 (11.69)	45.87 (34.74)	0.67	0.33
0.40	0.9923	12.27 (0.50)	23.58 (6.32)	36.47 (18.78)	0.68	0.32
0.50	0.9497	10.63 (0.94)	17.17 (12.02)	21.11 (35.73)	0.72	0.28
0.60	0.7702	9.29 (1.42)	10.89 (18.10)	5.16 (53.80)	0.87	0.13
0.70	0.7733	8.50 (1.34)	10.36 (17.06)	6.00 (50.72)	0.85	0.15
0.08	0.8002	7.87 (1.20)	10.06 (15.29)	7.05 (45.44)	0.82	0.18
0.09	0.8649	7.48 (0.94)	9.98 (12.00)	8.08 (35.69)	0.80	0.20
1.00	0.9816	7.64 (0.14)	9.96 (1.89)	7.50 (5.64)	0.81	0.19

Table S9. Determination coefficient (R^2), apparent thermodynamic properties of solutions (ΔG_{dis}^0), (ΔH_{dis}^0) and (ΔS_{dis}^0) and relative contribution of enthalpy ζ_H and entropy ζ_{TS} for syringic acid in cyrene aqueous solutions (p = 101.3 kPa).

w_H	R^2	ΔG_{dis}^0 (kJ.mol ⁻¹)	ΔH_{dis}^0 (kJ.mol ⁻¹)	ΔS_{dis}^0 (J.mol ⁻¹)	ζ_H	ζ_{TS}
0.05	0.867	20.60 (2.16)	23.15 (27.59)	8.21 (82.00)	0.90	0.10
0.10	0.9259	19.28 (1.61)	23.83 (20.51)	14.68 (60.96)	0.84	0.16
0.20	0.9252	17.41 (1.90)	27.94 (24.18)	33.95 (71.87)	0.73	0.27
0.30	0.9445	15.45 (1.80)	31.20 (23.01)	50.80 (68.39)	0.66	0.34
0.40	0.9730	13.74 (1.26)	31.74 (16.07)	58.05 (47.78)	0.64	0.36
0.50	0.9633	12.22 (1.36)	29.16 (17.30)	54.65 (51.44)	0.63	0.37
0.60	0.9815	10.95 (0.77)	23.37 (9.76)	40.08 (29.01)	0.65	0.35
0.70	0.9786	10.05 (0.69)	19.53 (8.79)	30.61 (26.11)	0.67	0.33
0.08	0.9943	9.58 (0.32)	17.96 (4.13)	27.02 (12.27)	0.68	0.32
0.09	0.9388	9.41 (0.80)	13.15 (10.21)	12.05 (30.36)	0.78	0.22
1.00	0.9816	10.16 (0.24)	16.57 (3.15)	20.69 (9.39)	0.72	0.28

S3. REFERENCES

- BAIRD, Z. S. *et al.* Vapor Pressures, Densities, and PC-SAFT Parameters for 11 Bio-compounds. [s.l.] **Springer** US, 2019. v. 40
- GEORGE, J.; SASTRY, N. V. Densities, Dynamic Viscosities, Speeds of Sound, and Relative Permittivities for Water + Alkanediols (Propane-1,2- and -1,3-diol and Butane-1,2-, -1,3-, -1,4-, and -2,3-Diol) at Different Temperatures. **Journal of Chemical and Engineering Data**, v. 48, n. 6, p. 1529–1539, 2003.
- LEAL-DUASO, A. *et al.* Glycerol as a source of designer solvents: physicochemical properties of low melting mixtures containing glycerol ethers and ammonium salts. **Phys. Chem. Chem. Phys.**, v. 19, n. 41, p. 28302–28312, 2017.
- LI, Q. S.; SU, M. G.; WANG, S. Densities and excess molar volumes for binary glycerol + 1-propanol, + 2-propanol, + 1,2-Propanediol, and + 1,3-propanediol mixtures at different temperatures. **Journal of Chemical and Engineering Data**, v. 52, n. 3, p. 1141–1145, 2007.
- NAIN, A. K. Densities and volumetric properties of binary mixtures of formamide with 1-butanol, 2-butanol, 1,3-butanediol and 1,4-butanediol at temperatures between 293.15 and 318.15 K. **Journal of Solution Chemistry**, v. 36, n. 4, p. 497–516, 2007.
- PARSA, J. B.; HAGHRO, M. F. Excess molar volume and viscosity deviation for binary mixtures of polyethylene glycol dimethyl ether 250 with 1,2-alkanediols (C3-C6) at T = (293.15 to 323.15) K. **Journal of Chemical Thermodynamics**, v. 40, n. 5, p. 782–788, 2008.
- SHI, X. *et al.* Density, Viscosity, and Excess Properties of Binary Mixtures of 2-(Methylamino)ethanol with 2-Methoxyethanol, 2-Ethoxyethanol, and 2-Butoxyethanol from 293.15 to 353.15 K. **Journal of Chemical and Engineering Data**, v. 64, n. 9, p. 3960–3970, 2019.

APPENDIX H

S1. COMPLEMENTARY FIGURES

Figure S1: Chemical structure of the alkanediols and glycerol ethers used in this work as extractive solvents.

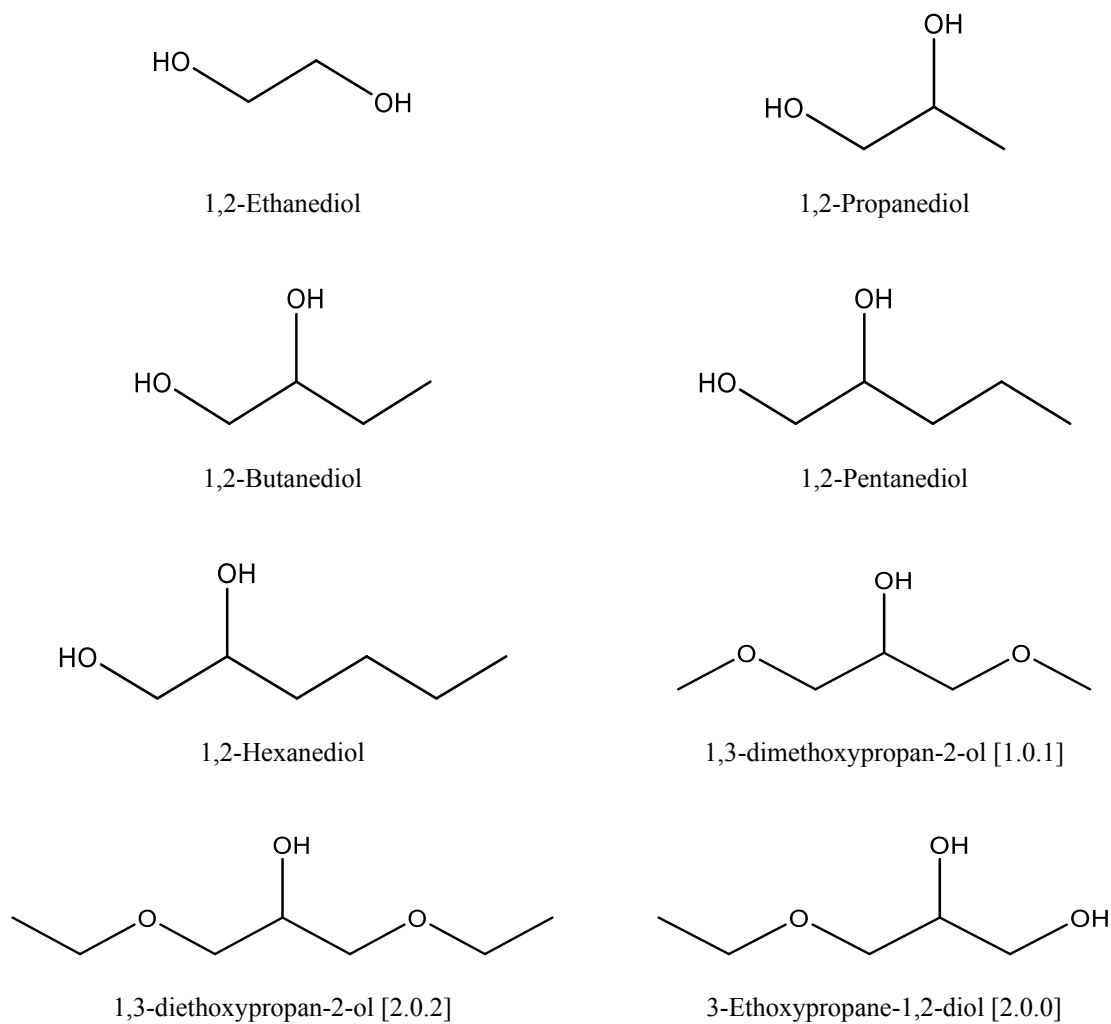


Figure S2: Kinetics assays of juçara pulps at 10 MPa, 353.2 K and 1 mL.min⁻¹ for PLE and the identified mass-transfer mechanisms: CER - constant extraction rate period, FER - falling extraction rate period, DC - diffusion-controlled period.

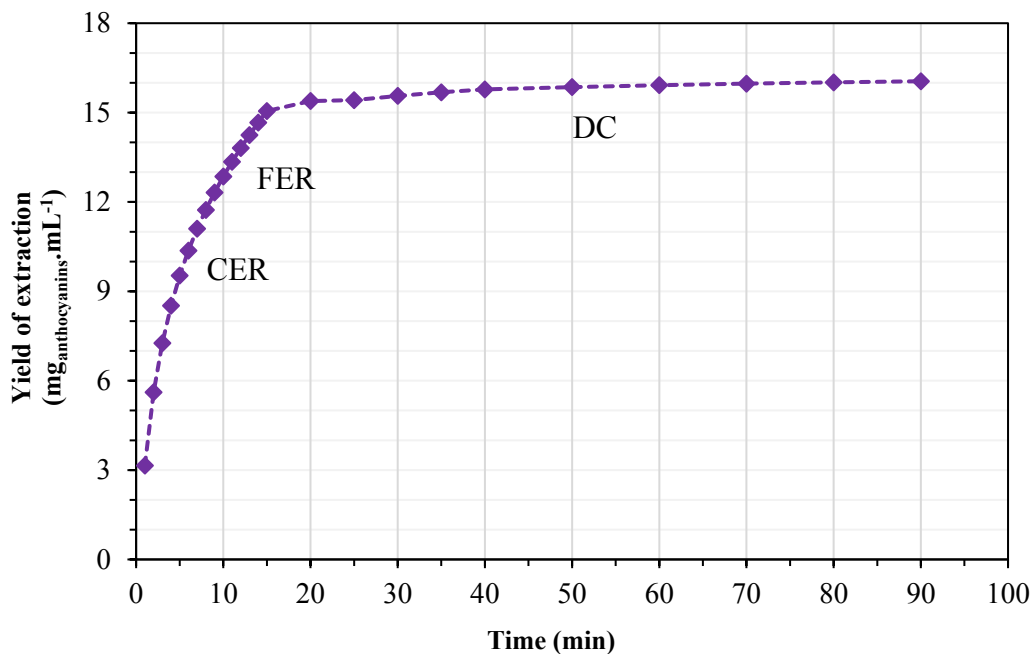


Figure S3: Pareto Chart of the CCRD (2³) regarding the yield of anthocyanins by PLE method.

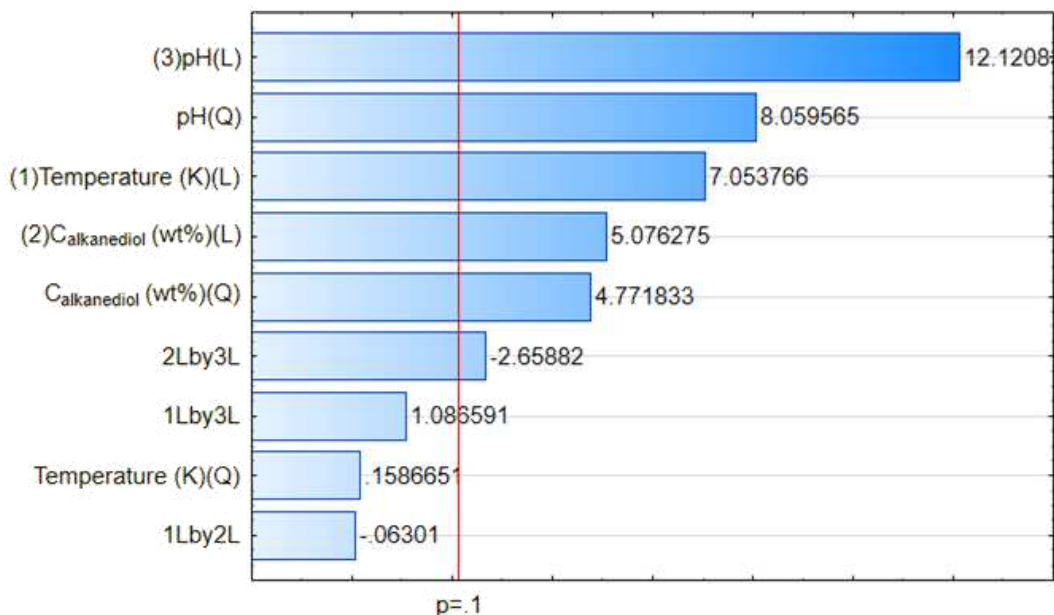


Figure S4: Kinetics assays of juçara pulps at 30% of amplitude for UAE.

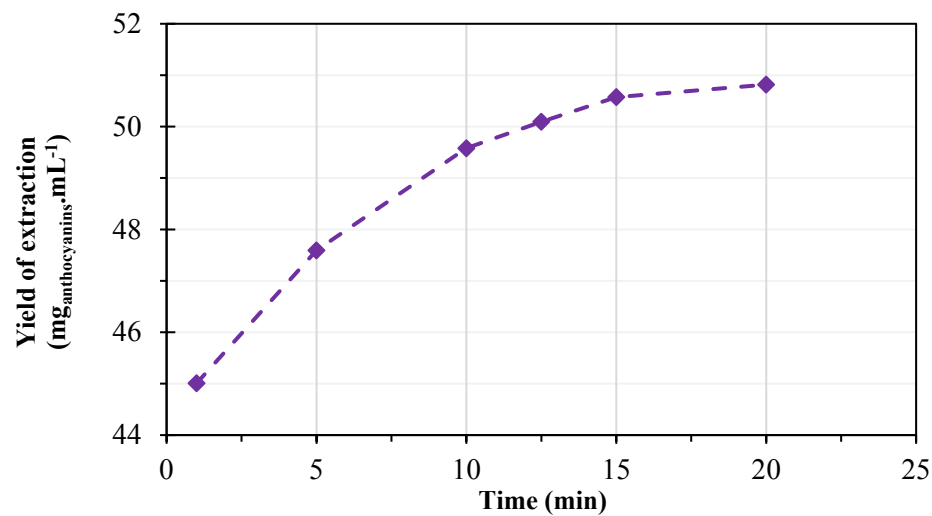
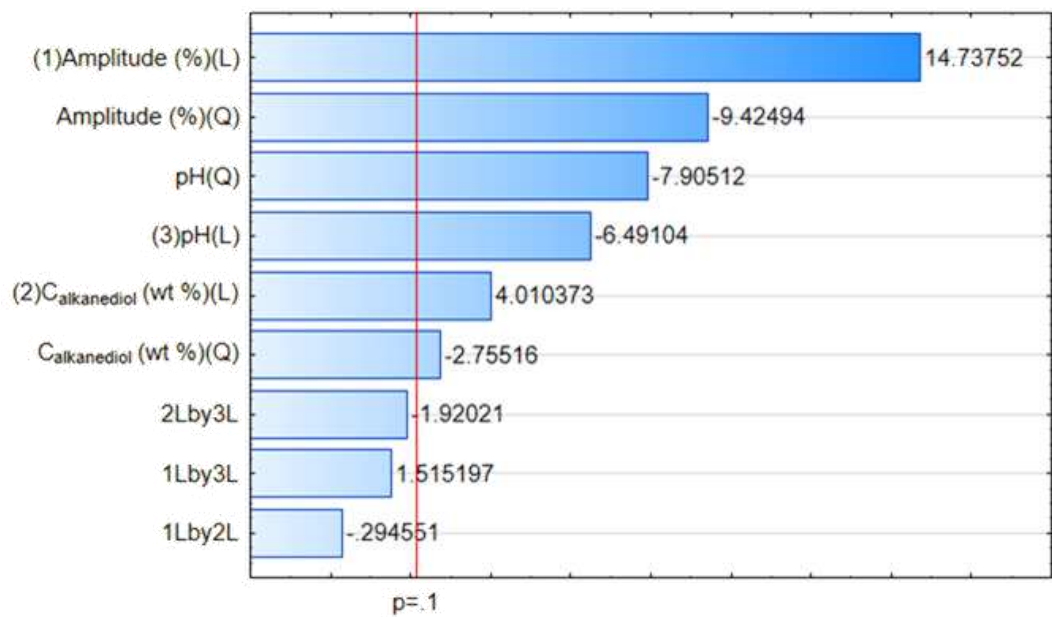


Figure S5: Pareto Chart of the CCRD (2³) regarding the yield of anthocyanins by UAE method.



S2. COMPLEMENTARY TABLES

Table S1: Regression coefficients and statistical parameters of the multiple regression of PLE responses in the Central Composite Rotatable Design (CCRD).

Response variables	Factors	Regression coefficient	Standard error	t-value	p-value	Lower limit	Upper limit
Yield of extraction of anthocyanins ($\text{mg}_{\text{anthocyanins}} \cdot \text{g}_{\text{dry biomass}}^{-1}$)	Constant	-3.67840	3.314466	-1.10980	0.329307	-10.7443	3.38753
	x_1	0.05881	0.008330	7.05959	0.002124	0.0410	0.07656
	x_2^2	0.00320	0.000523	6.10938	0.003633	0.0021	0.00431
	x_3	-3.14282	0.689633	-4.55723	0.010361	-4.6130	-1.67263
	x_3^2	0.59853	0.073937	8.09514	0.001266	0.4409	0.75615
	$x_2 \cdot x_3$	-0.02983	0.006971	-4.27947	0.012853	-0.0447	-0.01497
R^2	0.81						
adjusted R^2	0.74						
$F_{\text{calculated}}$	26.99						
p-value	$1.89 \cdot 10^{-6}$						
p_{model}	< 0.1						

Note – x_1 : temperature (K); x_2 : $C_{\text{alkanediol}}$ (wt%); x_3 : pH. Confidence level 90%.

Table S2: Regression coefficients and statistical parameters of multi regression adjustment of UAE responses in the Central Composite Rotatable Design (CCRD).

Response variables	Factors	Regression coefficient	Standard error	t-value	p-value	Lower limit	Upper limit
Yield of extraction of anthocyanins (mg _{anthocyanins} ·g _{dry biomass} ⁻¹)	Constant	27.25774	1.570059	17.36097	0.000065	23.91062	30.60487
	x ₁	0.82742	0.068345	12.10644	0.000267	0.68172	0.97312
	x ₁ ²	-0.01053	0.001117	-9.42494	0.000707	-0.01291	-0.00815
	x ₂	0.15912	0.043657	3.64482	0.021871	0.06605	0.25219
	x ₂ ²	-0.00194	0.000706	-2.75516	0.051103	-0.00345	-0.00044
	x ₃	1.60348	0.250899	6.39095	0.003077	1.06860	2.13836
	x ₃ ²	-0.13904	0.017589	-7.90512	0.001385	-0.17654	-0.10155
R ²	0.95						
adjusted R ²	0.93						
F _{calculated}	30.26						
p-value	9.36*10 ⁻⁷						
p _{model}	< 0.1						

Note – x₁: amplitude (%); x₂: C_{alkanediol} (wt %); x₃: pH. Confidence level 90%.

Table S3: Temperature average for each different amplitude and the corresponding standard deviation (s) of the assays of the Central Composite Rotatable Design (CCRD) by UAE.

Amplitude (%)	Final temperature average (K) ± s
10	341
18	365.2 ± 4
30	366.2 ± 2
42	370.2 ± 3
50	372.3

Table S4: Initial pH (42.6 wt.% of solvent) and final extract pH for different solvents by UAE.

Solvent	Initial solvent/water mixture pH (before adjusting)	Final extract pH
Water	5.65	4.12
1,2-ethanediol	6.44	4.35
1,2-propanediol	6.88	4.49
1,2-butanediol	6.63	4.47
1,2-pentanediol	6.30	4.45
1,2-hexanediol	6.22	4.31
Ethanol	7.28	4.53
[1.0.1]	3.75	4.82
[2.0.2]	3.60	4.72
[2.0.0]	7.67	4.56