Universidade de Aveiro Departamento de Química
2014

Fábio Miguel Mendes Ferreira

Valorização de resíduos farmacêuticos usando líquidos iónicos

Valorization of pharmaceutical wastes using ionic liquids

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Dissertação apresentada à Universidade de Aveiro para cumprimento dos requisitos necessários à obtenção do grau de Mestre em Biotecnologia, ramo Biotecnologia Industrial e Ambiental, realizada sob a orientação científica do Professor Doutor João Manuel da Costa e Araújo Pereira Coutinho, Professor Catedrático do Departamento de Química da Universidade de Aveiro e coorientação da Doutora Sónia Patrícia Marques Ventura, Estagiária de Pós – Doutoramento da Universidade de Aveiro.

Aos meus pais…

o júri

presidente

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palavras-chave

Extração líquido-líquido, sistemas aquosos bifásicos, líquidos iónicos, paracetamol, cafeína, resíduos farmacêuticos.

resumo

Este trabalho tem como objetivo estudar a aplicação de sistemas aquosos bifásicos (SAB) com líquidos iónicos (LIs) na extração de compostos ativos de resíduos farmacêuticos.

Atualmente os medicamentos fora do mercado são recolhidos e incinerados, sendo por isso a totalidade dos compostos ativos de interesse perdidos por combustão completa. Apesar da valorização energética destes resíduos há grandes perdas ao nível dos vários compostos presentes. Deste modo o desenvolvimento de novos processos simples de extração com vista a' valorização dos compostos ativos presentes nos mesmos é de grande importância, minimizando os impactos ambientais e permitindo a utilização destes resíduos como fonte de matérias primas.

Foi possível desenvolver novos SAB com LIs e proceder ao estudo e otimização dos mesmos na extração de cafeína e paracetamol. Por fim o processo otimizado foi aplicado a um resíduo medicamentoso (ALGIK). O resultado obtido demonstra uma completa extração dos compostos ativos com os SAB utilizados.

keywords

Liquid-liquid extraction, aqueous biphasic systems, ionic liquids, paracetamol, caffeine, pharmaceutical wastes.

abstract

The main objective of the present work is the application of aqueous biphasic systems (ABS) with ionic liquids (ILs) in the extraction of pharmaceutical wastes.

Nowadays, after their expiration date time recovered medicine waste are incinerated, destroying most of the valuable compounds present, with only a minor thermic valorisation. Therefore the development of new processes that allow this wastes valorization is crucial to create a more environmentally friendly process and the use of wastes as a source of raw materials.

The development of new ABS with ILs and their study allowed the optimization of the extraction of paracetamol and caffeine. Finally the application of this simple and fast process to a medical waste (ALGIK) resulted in a complete extraction of both paracetamol and caffeine.

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List of abbreviations

ABS	Aqueous Biphasic Systems
CMC	Critical micelle concentration
DDD	Defined Daily Dose
HPLC	High pressure liquid chromatography
IL	Ionic Liquid
IL-ABS	Ionic Liquid–based ABS
ILs	Ionic Liquids
SIGREM	Sistema Integrado de Gestão de Resíduos de Embalagens Medicamentos
TL	Tie-line
TLLs	Tie-line lengths
TLs	Tie-lines
$[C_2mim]Cl$	1-ethyl-3-methylimidazolium chloride
$[C_4mim]Cl$	1-butyl-3-methylimidazolium chloride
$[C_6mim]Cl$	1-hexyl-3-methylimidazolium chloride
$[C_8mim]Cl$	1-methyl-3-octylimidazolium chloride
$[C_4mpip]Cl$	1-butyl-1-methylpiperidinium chloride
$[C_4mpy]Cl$	1-butyl-3-methylpyridinium chloride
$[C_4mpyr]Cl$	1-butyl-1-methylpyrrolidinium chloride
$[N_{4444}]Cl$	tetrabutylammonium chloride
$[P_{4444}]Cl$	tetrabutylphosphonium chloride

[BzChol]Cl	benzyldimethyl(2-hydroxyethyl)ammonium chloride
$[C_4mim][N(CN)_2]$	1-butyl-3-methylimidazolium dicyanamide
$[C_4mim][CH_3SO_3]$	1-butyl-3-methylimidazolium methanesulfonate
$[C_4mim][CF_3SO_3]$	1-butyl-3-methylimidazolium triflate
$[C_4mim][SCN]$	1-butyl-3-methylimidazolium thiocyanate

List of symbols

wt %	weight percentage (%)
λ	wavelength (nm)
std	standart deviation
k_{ow}	octanol-water partition coefficient
R^2	correlation coefficient
α	hydrogen bond acidity
α’	ratio between the top weight and the total weight of the mixture
β	hydrogen bond basicity
EE %	Percentage extraction efficiencies (%)
m_x^{Top}	amount of paracetamol or caffeine (replacing x) in the top phase
$m_x^{Mixture}$	amount of paracetamol or caffeine (replacing x) in the mixture

1. General Introduction

1.1. Pharmaceutical Industry

The pharmaceutical industry in Europe has been growing and with it, the consumption of drugs and medicines. This increasing consumption is nowadays responsible for a big problem in the society due to the high level of waste produced, and its environmental impact [1]. This demand can only be solved by a better control and processing of the pharmaceutical wastes. In this context, some recent studies report the indexes of pharmaceutical waste collected, which vary from 0.19 tonnes/million *capita* in Croatia and 237 tonnes/million *capita* in Switzerland, being estimated that, about 50% of these wastes produced were not properly collected [1]. Recent studies show that in Germany, in average, the medicines returned have 65% of the original content [2] while in England, 77.7% of the returned medicine have at least half of the original content [3]. It is also estimated that 10% of the prescribed medication in England is wasted [4] and 20% of the population have wasted medicine in their houses [3].

There are many reasons for the disposal of the drugs. According to a recent Portuguese study it was determined that 21.7% of the defined daily dose (DDD) prescript was not used, 9.7% due to the inadequate package size (excessive dosage) and 10.2%, due to non-adhesion or necessity to change the therapeutic [2]. Since 2006, Portugal has its own pharmaceutical waste collection system maintained by Valormed company and designated by "Sistema Integrado de Gestão de Resíduos de Embalagens e Medicamentos", abbreviated by SIGREM [5]. According to recent databases provided by the SIGREM project, in the year of 2012, 849 tonnes of pharmaceutical wastes were collected in Portugal (Figure 1) and 882.367 tonnes were treated, from which around 500 tonnes were recycled through energetic valorisation [6]. This valorisation consists in the drugs and packages incineration at high temperatures (1500ºC-2000ºC) to promote the disintegration of organic molecules avoiding the environmental contamination [7, 8].

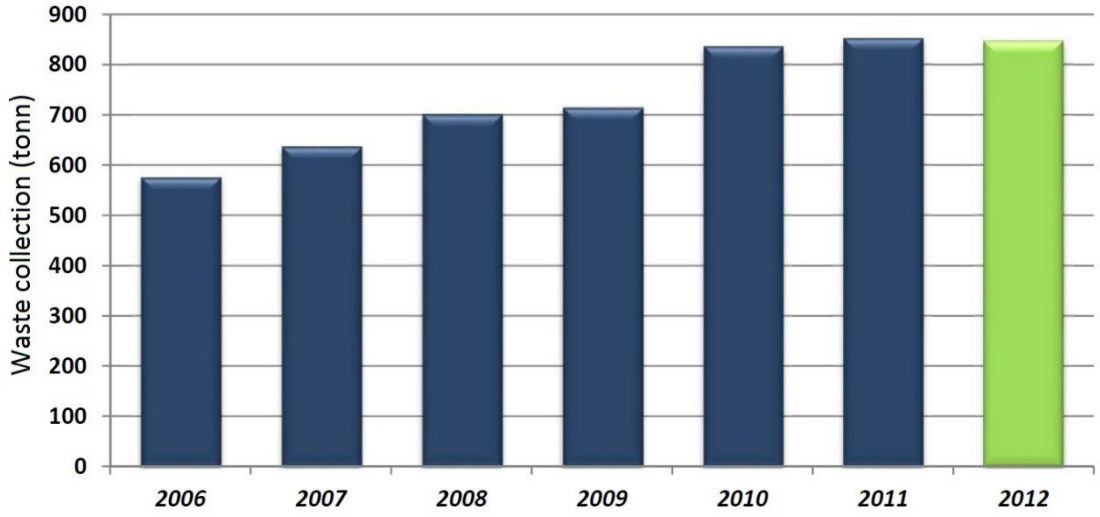


Figure 1 - Quantity of pharmaceutical wastes collected (tonnes) by year in Portugal. Adapted from Valormed [6].

However, and despite the known advantages such as the complete waste destruction and the large amounts of waste being simultaneously processed (drugs, packages and blisters) there are some pronounced disadvantages which are essentially related with the high costs of the equipment and process of incineration, and from our point of view, the large amounts of active compounds and chemical products with high added-value that are being consistently incinerated and lost, and which can be re-used as for example, laboratory standards or raw materials for production of chemical specialities or other drugs. Most European countries have drug waste recovery systems (regulated by European Union Directive 2004/27/EC) and people have a crescent tendency to use these systems to dispose the medicine wastes [9, 10], as depicted in Figure 2.

However and until now, these systems were only aiming at the complete destruction of the drugs, and were not considering the sustainability of the incineration process or even the recovery of some chemical compounds. This work is actually trying to implement a more sustainable treatment of some of these drugs by the recovery of some important active chemical compounds which are presently incinerated. As described by the authorities, a medicine is out of date when 10% of its principal active compound is no longer active [11]. This means that around 90% of the active chemical compound is still active and with enough stability to be recovered and reused in an industry of interest. Thus, several extraction techniques, like solid-liquid extraction and liquid-liquid extraction processes, among which aqueous biphasic systems (ABS), could be applied to the extraction, recovery and

purification of chemical active compounds aiming to add commercial value to these residues (Table 1).

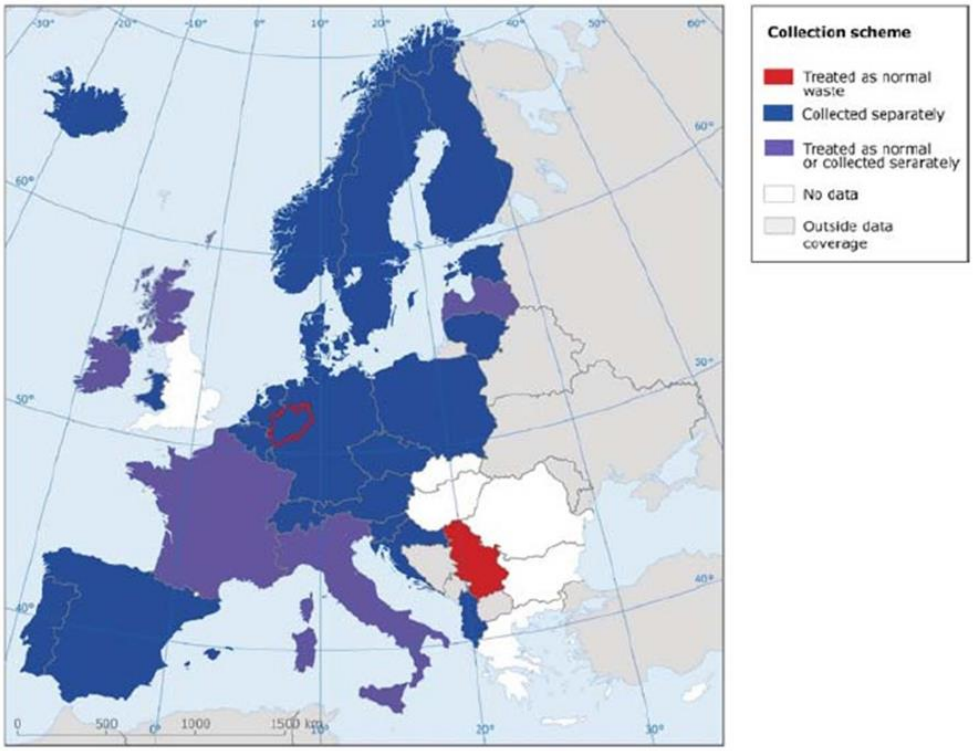


Figure 2 - European Union map and the adopted systems used for the drug waste collection [10].

As we consider extraction processes we need to consider medicine composition, because they are composed of both the active substance and excipients [12–14]. This excipients are essential on the medicine composition, giving the medicine control of the active substance bioavailability, stability and protection from degradation, ensuring a robust and reproducible physical product, and facilitating the administration to the target group [12, 14]. Despite their essential nature in the pharmaceuticals [12] they cannot be considered merely as inert or inactive ingredients [13], as they play an important role in the medicine formulation.

Table 1 - Price of some active substances used in the preparation of some anti-depressant drugs [15].

Active Substance	Price *per* g of active ingredient	Drugs
Fluoxetine	7320.00€	Prozac; Selectus; Psipax; Fluoxetina Tuneluz
Citalopram	12020.00€	Zitolex; Citalopram Actavis
Escitalopram oxalate	4420.00€	Cipralex
Paroxetine	25300.00€	Zanoxina; Voltak; Paxetil; Paroxetina Cinfa
Sertraline	45178.57€	Zoloft; Serlin; Serpax; Sertralina

Taken this into account, it is important to optimise the ABS focusing the extraction and isolation of the active substances apart from all the excipients present on the medicine. However, in order to perform these experiments, it is necessary to optimize the practical details of the extraction and thus, the use of simple systems is required. Simple systems are here defined as medicines in which their active substances are cheaper, structurally less complex and present in high concentrations. Following the optimization step, it will be possible to extrapolate the results for more complex systems and pharmaceutical matrices, in which the active substances are in lower concentrations (meaning higher concentrations of excipients), but with higher commercial value, namely anti-depressant active compounds (Table 1). As our model we will use an anti-flu medication called ALGIK (Table 2) which is easily obtained.

Table 2 – Quantity of active substance present in some medicines *per* pill [16].

Name	Class	Active substance	Quantity (mg)
Ben-U-Ron	Anti-flu	Paracetamol	1000
Melhoral	Anti-flu	Acetyl salicylic acid	500
Brufen Retard	Anti-flu	Ibuprofen	800
ALGIK	Anti-flu	Paracetamol	500
Prozac	Anti-depressant	Fluoxetine	20
Cipralex	Anti-depressant	Escitalopram	20
Zitolex	Anti-depressant	Citalopram	40

1.2. Aqueous Biphasic Systems (ABS)

Aqueous biphasic systems (ABS) are formed by mixing two aqueous solutions of mutual incompatible solutes that formed two immiscible phases above a critical concentration. [17, 18] Due to their aqueous nature they are the ideal media for the extraction, recovery and purification of biomolecules [19]. The most commonly used systems are polymer-polymer, polymer-salt or salt-salt combinations [18, 20]. The systems based on polymers have been exploited because they have a set of significant advantages, such as their versatility (with a large range of applications reported), high extraction efficiencies and selectivity, and also, these systems are capable of to extract compounds with a high purity level [21, 22]. Furthermore, they are considered biocompatible due to their high water content [18], they present a simple scale-up [17, 23, 24] and they ally two important processes in just one step, recovery and purification, these systems can be used as a continuous separation process [22]. Their extractive capacity is normally dependent of several factors, mainly the biomolecule affinity for the two phases (described by parameters like its solubility in the phases, its molecular size and the competition of interactions between the biomolecule and the components of the system [25]), being a good separation process for enzymes [19, 25, 26], proteins [19, 25–27], alkaloids [25] and antibiotics [25, 26]. However, the common ABS have some disadvantages, such as the high viscosity of the polymer [18, 22], the slow separation of phases [18] and the turbidity often found in the phases when polymers are used [28]. Despite some studies addressing these problems through the choice of the polymer (type, concentration and molecular weight), and the possible change of the salt (type and concentration) [29], some new alternatives are required to avoid the problems of opacity of the phases and more important, their poor polarity difference [18, 30]. In this context, other liquid-liquid extraction systems are appearing, namely alcohol-salt systems [22] and, more recently, ionic liquids-salt ABS [19].

Ionic Liquids (ILs) are salts with a melting temperature below 100 ºC [31, 32]. They are characterized by some interesting properties such as their non-flammability, negligible vapour pressure, high chemical and thermal stability and a large liquid temperature range [33, 34]. Moreover, and because they have a highly tunable nature, the possibility of different anion/cation/alkyl chain combinations is huge [21, 25] (Figure 3), which is in part related with the fact that, in recent years, ILs have received a great attention as “designer solvents” [35].

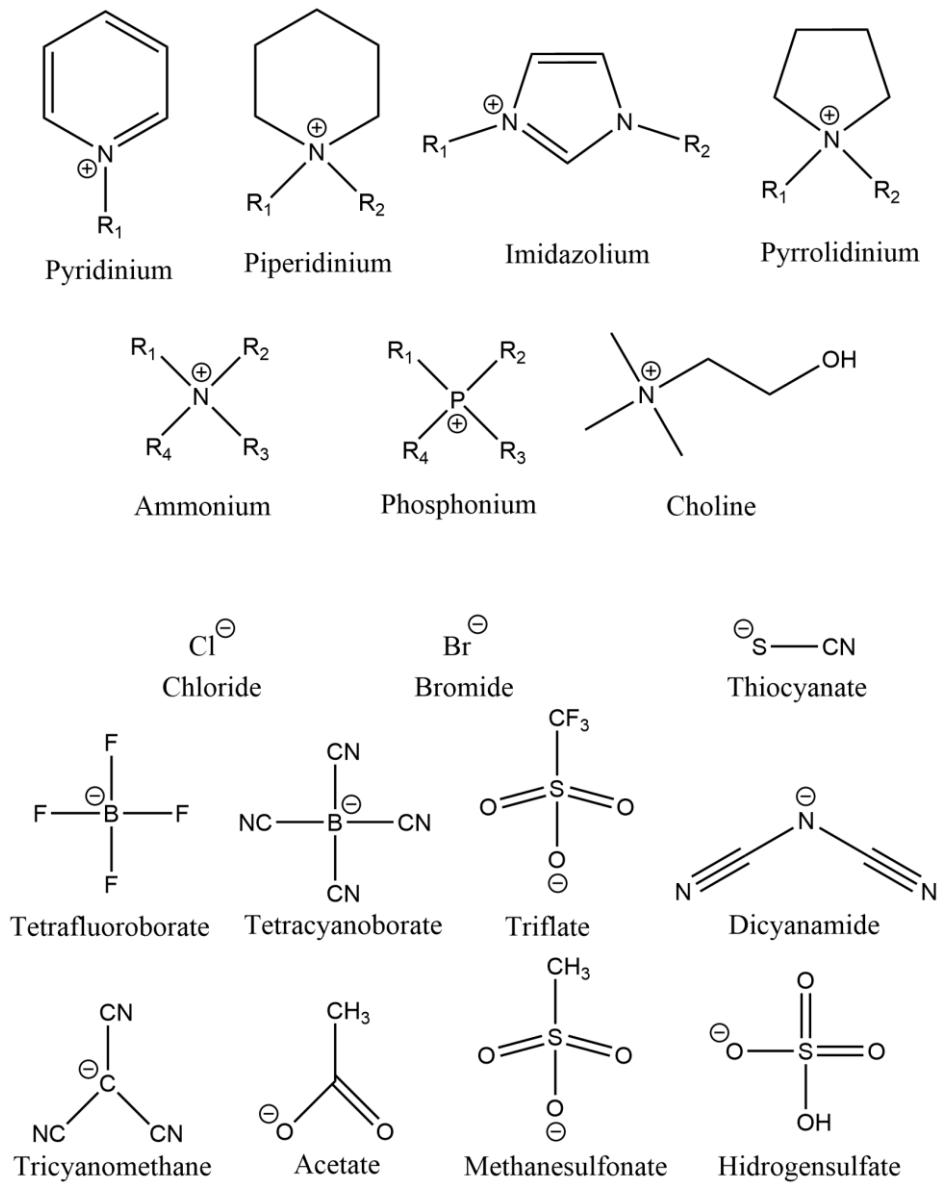


Figure 3 - Commonly used cations and anions on the ILs synthesis.

Ionic Liquid-based ABS (IL-ABS) were reported for the first time in 2003, by Rogers and co-workers [36], the number of works using these systems increasing since then [18]. This crescent interest in the IL-ABS is related with an easier mass transfer between the two phases when compared with the polymer-polymer ABS, since they have lower viscosities [17–19, 21] and due to the high solvency power of ILs, these ABS can be applicable to the extraction of a large range of compounds, including hydrophobic molecules [25]. Despite the high versatility in the ILs design, most IL-ABS reported are based on the imidazolium family [28, 33, 37–45], whereas the information concerning phosphonium [19, 41], quaternary ammonium [41,

[46, 47], pyridinium [28, 33, 38, 41, 45], cholinium [47], piperidinium [28, 33, 38] and pyrrolidinium [28, 33, 38] families in the ABS formation is still limited. The effects of the ionic liquid (IL) anion [28, 38, 42, 44, 45, 48] and the influence of diverse organic and inorganic salts, specifically phosphates [19, 41, 49–52], hydrogenophosphates [37, 41, 42, 50, 51, 53], citrates [54–57] and carbonates [37, 41, 42, 49, 51, 58] were analysed. These ABS were extensively studied in what concerns their potential for the separation of diverse biomolecules, such as amino-acids [19, 40, 59], antibiotics [60, 61], steroids [62], alkaloids [63], proteins [34], among others [19]. Moreover, there are also some studies regarding the biomolecules extraction from more complex matrices, namely the single-step extraction of Bisphenol A, using IL-based ABS prepared from biological samples [20], the extraction of enzymes from the fermentation broth [64], the selective recovery of dyes from the fermentation media [46] and the 100% extraction of paracetamol (an analgesic also known by acetaminophen) from a solid state pharmaceutical drug (Ben-U-Ron) [65]. Despite the differences between these and common ABS, some aspects are maintained in these systems such as the effect of pH and temperature being of great importance, especially when we are working with proteins or enzymes because of their denaturation or possible inactivation [34, 52, 66]. Regarding the temperature effect, in the extraction of vanillin with IL-based ABS the temperature greatly influences their extraction efficiency [21] and it is also reported a significant influence in the efficiency extraction of proteins [34]. The pH can be also used to control the direction of the biomolecule partitioning, as reported by Cláudio *et al.* [17] and if we look for the specific case of proteins and their acidic points or the amino-acids and their surface charges, the salt and pH choice is of utmost importance, due to the influence of this parameter in the 'molecule-water' and 'molecule-ILs' interactions [23].

From all these results, we believe that it will be possible to extract and purify the proposed active compounds from drug matrices with significant extraction yields and high purities (Table 3).

Table 3 - Pharmaceutical molecules recovered using polymeric ABS [29].

Pharmaceuticals	Recovery (%)	Purification factors
Human insulin-like growth factor 1 (IGF-1)	70	
Human recombinant interferon α1 (rhIFN-α1)	76	25
Tumor necrosis factor (TNF)	75	6
α_1-Antitrypsin (AAT)	91	
Apolipoprotein A-1	80	4.5
Milano variant of apolipoprotein A-1	85	7.2
Protein A	80	2.6
IgG		5.9
Penincillin	76	

This work is thus concentrated in the study of extraction of distinct active pharmaceutical ingredients from a drug matrix using ABS based in ILs.

In this work, paracetamol [67] and caffeine were considered as model compounds, present in the anti-flu pharmaceutical named ALGIK, and available as a powder solution. The aim is the extraction of these two chemicals from the medicine and, consequently, from the excipients bulk, and selectively isolate them one from each other, using liquid-liquid extraction technologies.

1.3. Main objectives and Scopes

The main objective of this work is to explore alternative extraction processes and purification techniques to separate a set of active substances from the correspondent pharmaceutical wastes. The criteria for the choice of the active chemical compounds are related with their concentration in the anti-flu medicine, the availability of the medicine in the market, and the availability and chemical simplicity of the pure molecules. In this context, this work will be divided in tree major parts as described below:

i) Design and characterization of new ABS composed of the McIlvaine buffer, different ILs and water.

ii) Application of the different ABS based in ILs and previously characterized as separation methodologies for the extraction of paracetamol and caffeine, here used as model compounds. In the final of this step, the determination of the best extraction systems for recovering the target compounds from the pharmaceutical waste is expected.

iii) Application of the extraction processes selected in *Step ii*, to perform the selective extraction of both paracetamol and caffeine (used as model molecules) from real pharmaceutical matrices.

The scope of this work is to design a process for the extraction and purification of these active chemical compounds from an anti-flu medicine, ALGIK (here used as model) and then to isolate both molecules from the excipients presence, since our main idea in the future is to control the extraction process to recover and purify chemicals with high value.

2. Optimization Study

Design of ternary phase diagrams and their application to the extraction of paracetamol and caffeine

2.1. Experimental section

2.1.1. Materials

The ABS studied in this work were prepared by using the McIlvaine buffer and different aqueous solutions of ILs. The McIlvaine buffer was prepared using potassium phosphate dibasic, K_2HPO_4 (98 wt% from JMVP) and citric acid monohydrate, $C_6H_8O_7 \cdot H_2O$ (100 wt% from Fisher Scientific). The ILs studied were: 1-ethyl-3-methylimidazolium chloride, [C_2mim]Cl (98 wt%); 1-butyl-3-methylimidazolium chloride, [C_4mim]Cl (99 wt%); 1-hexyl-3-methylimidazolium chloride, [C_6mim]Cl (98 wt%); 1-methyl-3-octylimidazolium chloride, [C_8mim]Cl (99 wt%); 1-butyl-1-methylpiperidinium chloride, [C_4mpip]Cl (99 wt%); 1-butyl-3-methylpyridinium chloride, [C_4mpy]Cl (98 wt%); 1-butyl-1-methylpyrrolidinium chloride, [C_4mpyr]Cl (99 wt%); tetrabutylammonium chloride, [N_{4444}]Cl (97 wt%); tetrabutylphosphonium chloride, [P_{4444}]Cl (98 wt%); benzyldimethyl(2-hydroxyethyl)ammonium chloride, [BzChol]Cl (97 wt%); 1-butyl-3-methylimidazolium dicyanamide, [C_4mim][$N(CN)_2$] (98 wt%); 1-butyl-3-methylimidazolium methanesulfonate, [C_4mim][CH_3SO_3] (99 wt%); 1-butyl-3-methylimidazolium triflate, [C_4mim][CF_3SO_3] (99 wt%); 1-butyl-3-methylimidazolium thiocyanate, [C_4mim][SCN] (98 wt%). The ILs structures are shown in Figure 5. All ILs were purchased from Iolitec, with the exception of [P_{4444}]Cl and [N_{4444}]Cl that were acquired at Cytec and Sigma-Aldrich®, respectively. Before use, all ILs were dried under constant stirring at vacuum and moderate temperature (≈ 353K) for a minimum 24h. The water used was double distilled, passed by a reverse osmosis system and further treated with a Milli-plus 185 water purification apparatus.

Paracetamol, N-(4-hydroxyphenyl)acetamide, or acetaminophen (≥99 wt%), was acquired at Sigma-Aldrich® and caffeine (≥99 wt%) was obtained from Fluka. The water used was double distilled, passed by a reverse osmosis system and further treated with a Milli-plus 185 water purification apparatus.

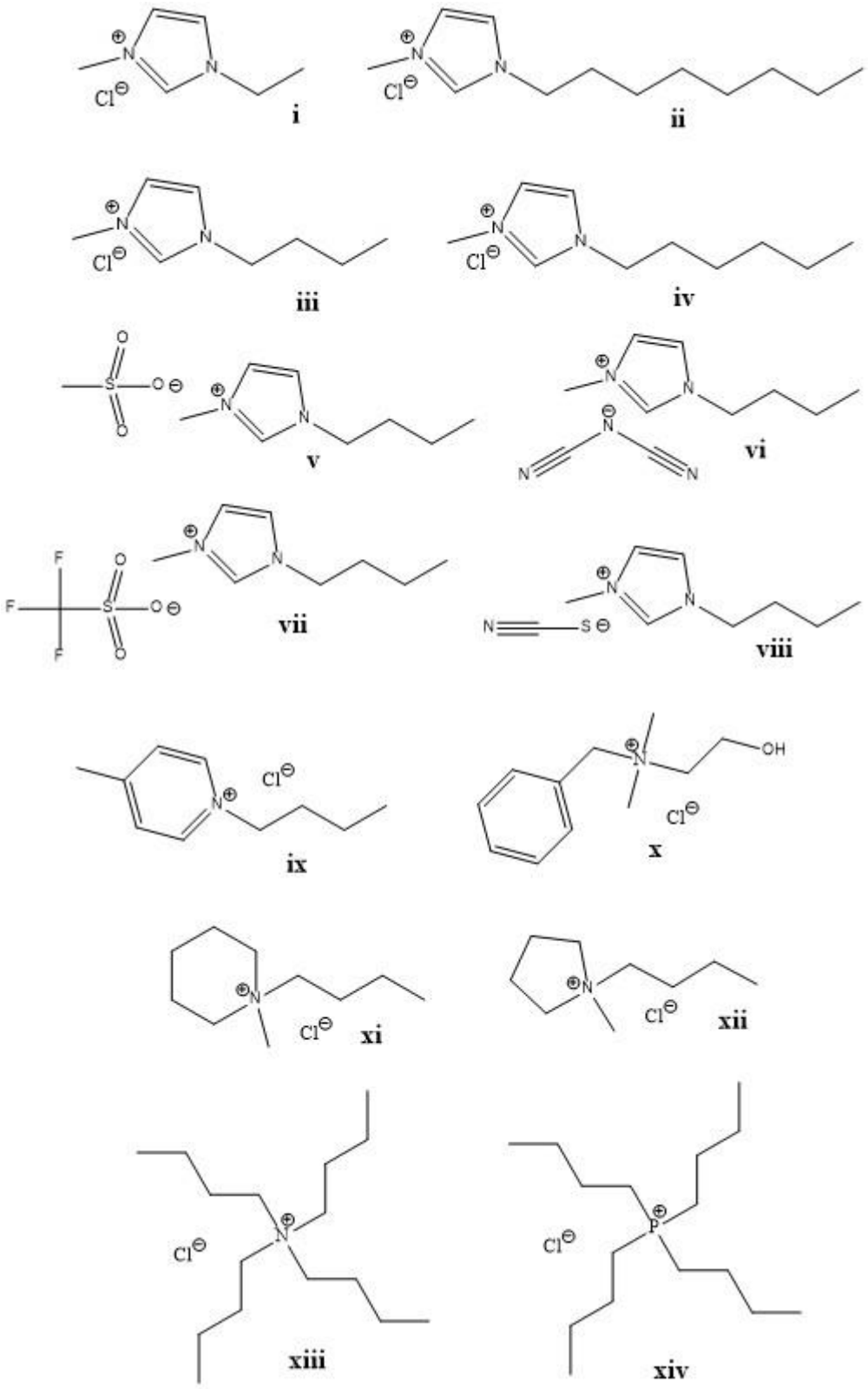


Figure 4 - Chemical structure of the ILs studied: (i) [C_2mim]Cl; (ii) [C_8mim]Cl; (iii) [C_4mim]Cl; (iv) [C_6mim]Cl; (v) [C_4mim][CH_3SO_3]; (vi) [C_4mim][$N(CN)_2$]; (vii) [C_4mim][CF_3SO_3]; (viii) [C_4mim][SCN]; (ix) [C_4mpy]Cl; (x) [BzChol]Cl; (xi) [C_4mpip]Cl; (xii) [C_4mpyr]Cl; (xiii) [N_{4444}]Cl; (xiv) [P_{4444}]Cl.

2.1.2. Methods

2.1.2.1 Phase diagrams determination

The binodal curves were determined using the cloud point titration method at atmospheric pressure and 298 (± 1) K [59]. Aqueous solutions of McIlvaine buffer at 50 wt%, pH 7 and aqueous solutions of different ILs at variable concentrations were prepared and used for the determination of the binodal curves. Repetitive drop-wise addition of the aqueous buffer solution to the IL aqueous solution was carried out up to the establishment of a cloudy solution, the biphasic region, followed by the drop-wise addition of ultrapure water up to the establishment of a limpid solution, the monophasic region. The procedure was carried out with under constant stirring. The ternary system compositions were determined by the weight quantification of all components added within an uncertainty of ± 10^{-4} g.

Tie-lines (TLs) were determined by a gravimetric method described by Merchuk *et al.* [68]. In order to determine the TLs, a mixture of the biphasic region was prepared, vigorously stirred and allowed to reach the equilibrium by the separation of both phases for 12h at 298 (± 1) K. Both top and bottom phases were weighted after the separation. To conclude, each individual TL was determined by application of the lever-arm rule [68].

All experimental binodal curves were correlated using Equation 1 [68],

$$Y = A\exp[(\mathrm{B}X^{0.5}) - (CX^3)] \quad (1)$$

where Y and X are the IL and salt weight percentages, respectively, and A, B, C are constants obtained by the regression.

In order to determine the TLs the following system of four equations (Equations 2 to 5) and four unknown values (Y_T, Y_B, X_T and X_B) was solved:

$$\begin{cases} Y_T = A\,exp[\left(BX_T{}^{0.5}\right) - \left(CX_T{}^3\right)] & (2) \\ Y_B = A\,exp[\left(BX_B{}^{0.5}\right) - \left(CX_B{}^3\right)] & (3) \\ Y_T = \dfrac{Y_M}{\propto'} - \left(\dfrac{1-\propto'}{\propto'}\right) Y_B & (4) \\ X_T = \dfrac{X_M}{\propto'} - \left(\dfrac{1-\propto'}{\propto'}\right) X_B & (5) \end{cases}$$

The subscript letters T, B and M represent the top, bottom and mixture phases, respectively. The parameter α' is the ratio between the top and total mass of the mixture. For the calculation of the tie-line lengths (TLLs) Equation 6 was applied:

$$TLL = \sqrt{(X_T - X_B)^2 + (Y_T - Y_B)^2} \quad (6)$$

2.1.2.2 Partitioning of paracetamol and caffeine

A mixture point was selected in the biphasic region to evaluate the paracetamol and caffeine partitions based on the phase diagrams previously determined. The mixtures compositions were: 25 wt% McIlvaine buffer at pH 7 + 25 wt% IL + H_2O+ 10 wt% paracetamol or caffeine solutions with a concentration of 10 $g.dm^{-3}$ and 2.5 $g.dm^{-3}$, respectively.

Each mixture was vigorously stirred and allowed to reach the equilibrium for 12h at 298 (± 1) K to allow the phase separation and also the complete partition of both compounds to extract. The phases were carefully separated, and their weight and pH values were measured. The quantification of paracetamol and caffeine were done in triplicate and, at least, three different assays for each system were performed (average values accompanied by the respective standard deviations are reported).Their concentration was determined by UV-Vis spectrometry, using a SHIMADZU UV-1700, Pharma-Spec Spectrometer, at a wavelength of 243 nm for paracetamol and 273 nm for caffeine, using calibration curves previously established (Appendix E). The wavelength of the absorption peak of paracetamol and caffeine was confirmed to remain unaffected, within the range of conditions tested. Possible interferences of the ILs and salt components with the paracetamol and caffeine quantification method were eliminated by the regular application of blank controls, where the paracetamol/caffeine aqueous solution was substituted by distilled water.

The extraction efficiencies of paracetamol and caffeine - EE (%) - are defined as the percentage ratio between the amount of chemical in the top phase (IL-rich phase) and in the total mixture, according to Equation 7,

$$EE\% = \frac{m_x^{Top}}{m_x^{Mixture}} \quad (7)$$

where m_x^{Top} and $m_x^{Mixture}$ are the amount of paracetamol or caffeine (replacing x) in the top (IL-rich) phase and in the total mixture, respectively. The top phase was chosen because it evidences the preferential migration of both molecules.

The pH values of the systems were measured using a METTER TOLEDO SevenMulti™ pH meter within an uncertainty of ± 0.02 and at 298 (± 1) K.

2.2. Results and discussion

2.2.1. Phase diagrams and tie-lines

Some new ternary phase diagrams were determined for several systems composed of ILs + water + McIlvaine buffer (pH 7) at 298 (± 1) K and atmospheric pressure. The binodal curves are illustrated in Figure 5 and the respective experimental data is given at Appendix A, from Table A.1 to Table A.9. All binodal curves illustrated in Figure 5 are presented in molality units to avoid differences resulting from the different molecular weights of the ABS constituents, allowing a better understanding of the IL structure impact on the phase diagrams. All systems are described by top phase representing the IL-rich phase and bottom phase representing the salt-rich phase. The results shown in Figure 5 show that the higher the IL' capacity to form ABS, the larger the biphasic region area presented. To support an easier and more detailed evaluation of the influence of the IL structure on the phase diagrams, separated Figures are used being thus, the alkyl side chain effect (Figure 5a), cation (Figure 5b) and anion effect (Figure 5c) included.

Considering the alkyl side chain effect (Figure 5a), the results obtained show that the IL capacity to form ABS follows the order $[C_6mim]Cl > [C_8mim]Cl \geq [C_4mim]Cl > [C_2mim]Cl$. There is a trend from $[C_2mim]Cl$ to $[C_6mim]Cl$, where the elongation of the alkyl side chain on the imidazolium cation facilitates de formation of ABS, due to the increase in the IL hydrophobic nature, reducing their affinity for water, consequently promoting the phase split. This effect is well documented for other salts [33, 69, 70]. On the other hand, $[C_8mim]Cl$ appears with an outsider, its behavior not following the trend of hydrophobicity. This behavior is not new [33, 44, 71] and it is justified by the presence of aggregates, formed by the self-aggregation of this specific IL. In ILs with longer alkyl chains than hexyl, their self-aggregation occurs promoted by the presence of a salting-out-inducing salt that lowers the critical micelle concentration (CMC), increasing the IL solubility in water and decreasing its ability to form ABS [33, 44, 71].

The effect of the cation core on the ABS formation shows that, at 1.0 $mol \cdot kg^{-1}$, the following decreasing order of aptitude to form ABS is (Figure 5b): $[P_{4444}]^+ > [N_{4444}]^+ > [C_4mpip]^+ > [C_4mpyr]^+ \geq [C_4mpy]^+ \geq [BzChol]^+ > [C_4mim]^+$. Since the side alkyl chain and the anion are the same, Figure 5b intends to demonstrate the impact of distinct cations/family on the phase formation. The results from Figure 5b clearly suggest the presence of two

distinct groups, the first one constituted by cyclic nitrogen-based ILs (aromatic: pyridinium, imidazolium and benzylcholine; non-aromatic: pyrrolidinium and piperidinium) and another constituted by acyclic cation structures, namely phosphonium and quaternary ammonium. This behavior is justified essentially by the higher hydrophobic character of the acyclic ILs (representing their lower affinity for water), in particular phosphonium- and ammonium. On the other hand we observe that the cyclic nitrogen-based ILs have lower ability to form ABS when compared with pyrrolidinium- and piperidinium-based ILs. Moreover, and comparing all cyclic nitrogen-based ILs, it is possible to conclude that the 5-atom ring cations (imidazolium and pyrrolidinium) have lower capacity to form ABS when compared with the 6-atom rings (pyridinium and piperidinium), being this behavior closely correlated with the molar volume of these ILs [33, 41]. The behavior of the series of IL + McIlvaine buffer (pH 7) + water ABS here assessed is in good agreement with the results of previous studies where other "*salting-out*" agents, as potassium phosphate buffer [33], potassium citrate salt [70] and potassium citrate buffer [72, 73] were applied. The smaller and hydroxyl containing chains of the cholinium cation are enhancing its affinity/solvation for water, consequently reducing its ability to induce ABS formation, result also in agreement with literature [72].

The anion impact on the ABS design is shown in Figure 5c, being the ability of the different anions to form ABS following the decreasing order: $[C_4mim][CF_3SO_3]$ > $[C_4mim][SCN]$ > $[C_4mim][N(CN)_2]$ > $[C_4mim][CH_3SO_3]$ ≥ $[C_4mim]Cl$. The anion rank observed for the $[C_4mim]$-based series studied here is in close agreement with the results previously obtained for $[C_2mim]$- and $[C_4mim]$-based ILs with K_3PO_4, Na_2SO_4, potassium phosphate buffer as the main salting-out species [38, 48, 58], among others [18]. As a general trend, it was already established that, for the same salting-out salt species, the hydrophobic/hydrophilic nature of the ILs is a crucial key in the ABS formation.

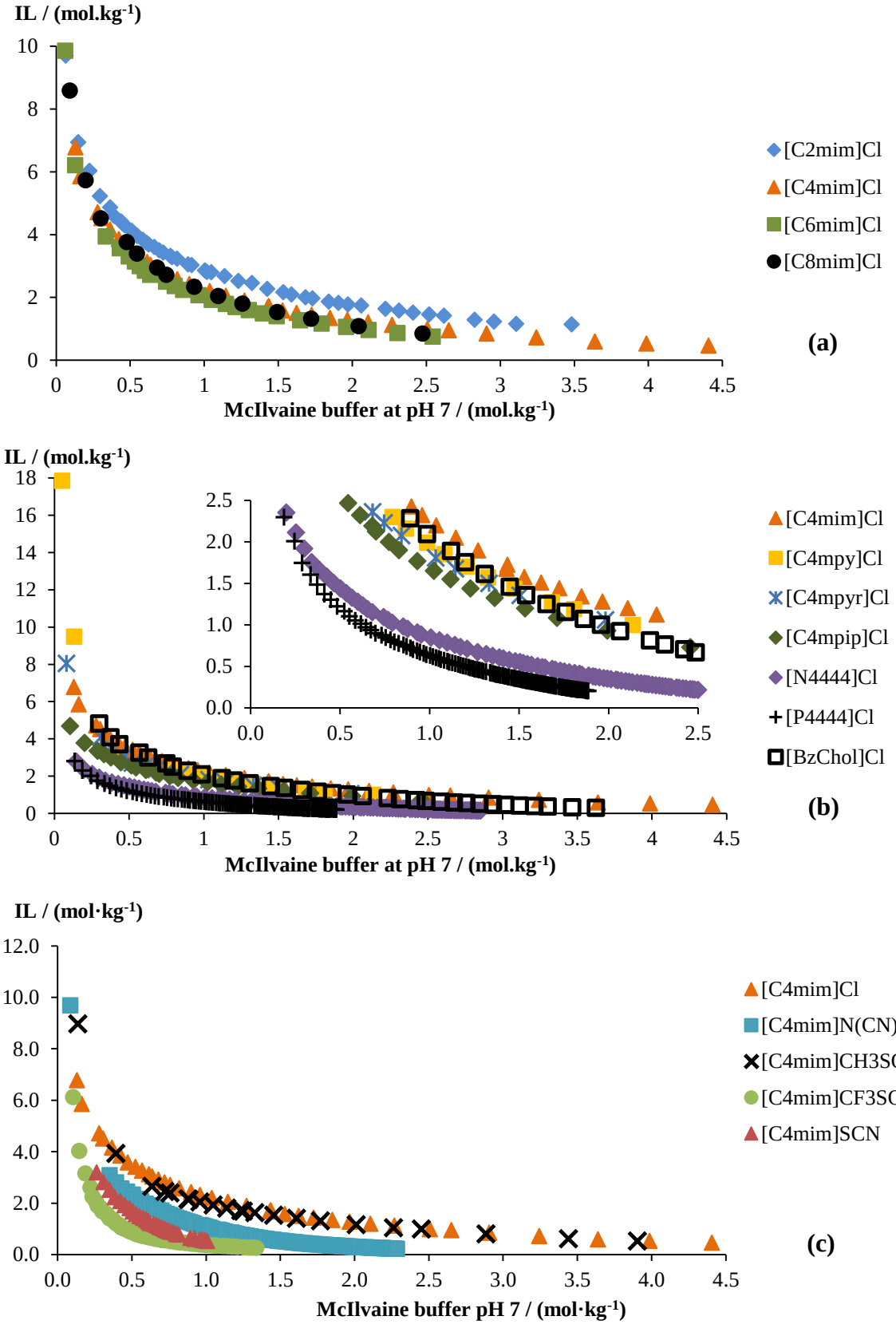
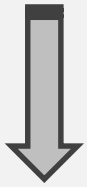


Figure 5 – Evaluation of the effects of the IL alkyl side chain length (a), cation (b) and anion (c) in the formation of ABS.

It is well-known that hydrophobic ILs are more easily salted-out due to their poorer affinity for water. In this sense, the ability of the IL anions to promote the phase split closely correlates with their decrease on the hydrogen bond basicity (β) [74] shown in Table 4. Therefore, IL anions that tend to preferentially interact with the protons of water (and thus, to create hydration complexes) require more quantities of inorganic salt to undergo liquid−liquid demixing.

Table 4 - Hydrogen Bond Acidity (α) and Hydrogen Bond Basicity (β) of [C_4mim]-based ILs with the solvatochromic probe $[Fe(phen)_2(CN)_2]ClO_4$ [74].

IL anion	α	β	ABS formation ability
Cl	0.32	0.95	
$[CH_3SO_3]$	0.36	0.85	
[SCN]	0.43	0.71	
$[N(CN)_2]$	0.44	0.64	
$[CF_3SO_3]$	0.50	0.57	

In general, all anions investigated in this work and whose results are depicted in Figure 5c follow the trend showed in Table 4, between β and the ABS formation ability.

The correlation parameters obtained from the experimental data of the binodal curves (Appendix A) by application of Equation 1, firstly proposed by Merchuk and co-workers [68] are provided in Table 5. The TLs and TLLs obtained for each system are described in Table 6 and Figures (Figure A 1 to Figure A 5 in Appendix A). As an example, the ABS composed of [BzChol]Cl + McIlvaine buffer at pH 7 and the respective TL is illustrated in Figure 6.

Table 5 - Parameters obtained from Equation 1 and respective standard deviations (std) plus the correlation coefficients (R^2) for the different ILs + McIlvaine buffer at pH 7 + H_2O ternary systems studied.

IL	A ± std	B ± std	10^5 C ± std	R^2
[C_2mim]Cl	74.15 ± 0.37	-0.2347 ± 0.0018	0.4585 ± 0.0250	0.9992
[C_4mim]Cl	79.77 ± 0.49	-0.2611 ± 0.0022	0.8117 ± 0.0278	0.9995
[C_6mim]Cl	86.80 ± 0.89	-0.2750 ± 0.0040	1.190 ± 0.1028	0.9983
[C_8mim]Cl	89.33 ± 1.05	-0.2392 ± 0.0048	1.419 ± 0.1189	0.9989
[C_4mpip]Cl	66.77 ± 0.66	-0.2453 ± 0.0038	1.365 ± 0.0978	0.9988
[C_4mpy]Cl	109.3 ± 0.81	-0.3627 ± 0.0040	0.1155 ± 0.1437	0.9996
[C_4mpyr]Cl	86.39 ± 1.65	-0.3273 ± 0.0079	3.251×10^{-10} ± 0.2777	0.9967
[N_{4444}]Cl	70.43 ± 0.26	-0.3114 ± 0.0013	2.712 ± 0.0195	0.9998
[P_{4444}]Cl	80.20 ± 0.36	-0.3823 ± 0.0017	4.526 ± 0.0443	0.9996
[BzChol]Cl	91.08 ± 0.74	-0.2614 ± 0.0026	1.864 ± 0.0281	0.9998
[C_4mim][$N(CN)_2$]	114.3 ± 0.88	-0.4386 ± 0.0028	4.325 ± 0.0707	0.9987
[C_4mim][CH_3SO_3]	107.4 ± 2.16	-0.3114 ± 0.0065	2.786 ± 0.0823	0.9960
[C_4mim][CF_3SO_3]	179.8 ± 2.53	-0.7579 ± 0.0067	1.796×10^{-8} ± 0.5189	0.9980
[C_4mim][SCN]	115.1 ± 3.91	-0.5050 ± 0.0142	0.2000 ± 0.9359	0.9984

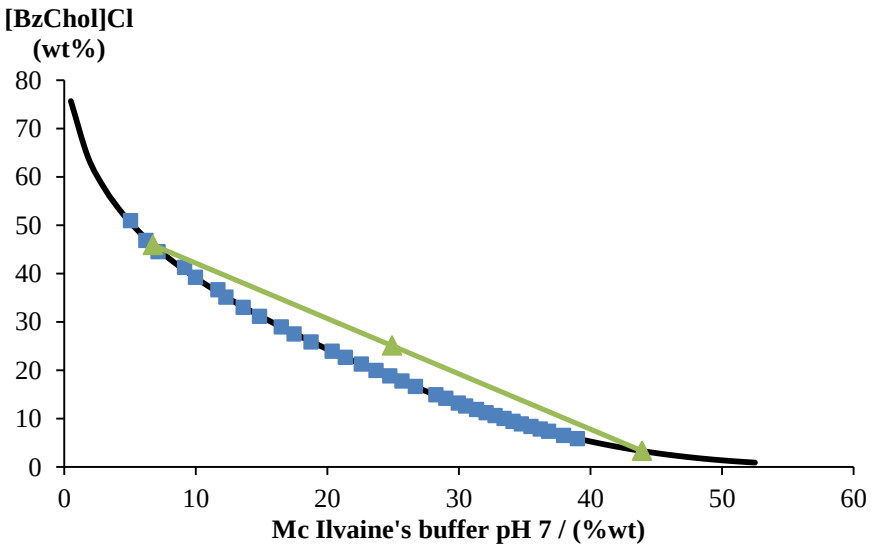


Figure 6 - Phase diagram of the system [BzChol]Cl + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (▲); fitting of the experimental data through Equation 1 (▬).

Table 6 – Mass fraction compositions (in percentage) and tie-line length (TLL) data for all ABS studied.

IL	Weight fraction composition (wt%)								TLL
	$[IL]_T$	$[Salt]_T$	$[H_2O]_T$	$[IL]_M$	$[Salt]_M$	$[IL]_B$	$[Salt]_B$	$[H_2O]_B$	
$[C_2mim]Cl$	32.39	12.20	55.41	24.92	24.91	4.649	59.39	35.96	54.74
$[C_4mim]Cl$	35.80	9.264	54.94	25.03	25.06	2.093	58.69	39.22	59.83
$[C_6mim]Cl$	38.56	8.546	52.89	25.03	24.80	2.168	52.28	45.55	56.90
$[C_8mim]Cl$	43.75	8.676	47.57	24.76	25.03	8.814	38.77	52.42	46.11
$[C_4mpip]Cl$	44.56	2.715	52.73	24.92	25.02	1.838	51.25	46.91	64.65
$[C_4mpy]Cl$	34.76	9.957	55.28	24.99	24.93	6.505	53.26	40.24	51.71
$[C_4mpyr]Cl$	34.49	7.870	57.64	25.02	25.07	7.246	57.34	35.41	56.48
$[N_{4444}]Cl$	56.23	0.5228	43.25	25.30	29.72	0.1135	53.50	46.39	77.17
$[P_{4444}]Cl$	62.51	0.4249	37.07	24.88	25.19	0.2772	41.39	58.33	74.50
[BzChol]Cl	45.93	6.747	47.32	25.11	24.91	3.322	43.92	52.76	56.54
$[C_4mim][N(CN)_2]$	58.57	2.323	39.11	25.28	29.81	1.82×10^{-2}	50.67	49.31	75.93
$[C_4mim][CH_3SO_3]$	30.88	15.75	53.37	24.71	24.99	8.835	48.77	42.40	39.71
$[C_4mim][CF_3SO_3]$	74.16	1.377	24.46	25.10	29.74	0.9251	43.72	55.35	84.59
$[C_4mim][SCN]$	69.08	1.021	29.90	24.82	25.03	5.590×10^{-5}	38.49	61.51	78.59

2.2.2. Optimization study of paracetamol and caffeine partitioning

The applicability of the studied ABS (last chapter) to promote the partitioning of paracetamol and caffeine is investigated using model systems for the optimization of the various operating conditions, namely different alkyl chain lengths, cation cores and anion moieties, while maintaining constant the pH of the aqueous medium at 7. The system composed of [C_4mpy]Cl + McIlvaine buffer at pH 7 + H_2O was not applied in the partitioning of caffeine and paracetamol due to some practical limitations. All ABS were prepared with the composition 25 wt% of IL + 25 wt% of McIlvaine buffer solution at pH 7 + 40 wt% of H_2O + 10 wt% of an aqueous solution with paracetamol (10 g·dm^{-3}) or caffeine (2.5 g·dm^{-3}). At pH 7, value adopted during all the partitioning experiments, both paracetamol and caffeine are listed as non-charged species (for more details see the speciation curves of both molecules in Appendix B). In order to confirm the constant pH value, this parameter was checked after the partitioning process (Table 7).

Table 7 - Experimental pH values of both ABS phases.

IL	Paracetamol		Caffeine	
	pH_{IL} ± sd	**pH_{Salt} ± sd**	**pH_{IL} ± sd**	**pH_{Salt} ± sd**
[C_2mim]Cl	7.499 ± 0.002	7.506 ± 0.011	7.651 ± 0.005	7.682 ± 0.029
[C_4mim]Cl	7.347 ± 0.007	7.115 ± 0.009	7.446 ± 0.061	7.429 ± 0.117
[C_6mim]Cl	7.180 ± 0.010	7.067 ± 0.004	7.500 ± 0.004	7.498 ± 0.005
[C_8mim]Cl	7.491 ± 0.057	7.333 ± 0.004	7.019 ± 0.019	7.012 ± 0.052
[C_4mpip]Cl	7.990 ± 0.033	7.653 ± 0.196	7.989 ± 0.028	7.499 ± 0.002
[C_4mpyr]Cl	7.820 ± 0.080	7.967 ± 0.067	7.333 ± 0.125	7.152 ± 0.040
[N_{4444}]Cl	7.665 ± 0.014	7.856 ± 0.012	7.227 ± 0.072	7.122 ± 0.099
[P_{4444}]Cl	7.903 ± 0.046	7.258 ± 0.002	8.112 ± 0.027	7.329 ± 0.011
[BzChol]Cl	7.512 ± 0.023	7.470 ± 0.032	7.487 ± 0.054	7.500 ± 0.209
[C_4mim][$N(CN)_2$]	7.425 ± 0.006	6.918 ± 0.001	7.368 ± 0.033	7.214 ± 0.186
[C_4mim][CH_3SO_3]	7.763 ± 0.008	7.440 ± 0.022	7.771 ± 0.007	7.453 ± 0.130
[C_4mim][CF_3SO_3]	7.163 ± 0.070	6.833 ± 0.005	7.240 ± 0.093	7.171 ± 0.108
[C_4mim][SCN]	7.856 ± 0.071	7.432 ± 0.172	7.993 ± 0.008	7.276 ± 0.008

There are other reports of extraction of paracetamol and caffeine with ILs ABS. Partitioning of paracetamol was study by e Silva *et al* using ABS of quaternary ammonium

halides, among which [N_{4444}]Cl, and different salts. In all systems paracetamol migrates preferentially to the ammonium-rich phase, accomplishing EE% between 80% and 100% [65]. Freire *et al* reported high-performance extraction of caffeine with ABS of ILs and K_3PO_4 salt. The also caffeine migrate preferentially to the IL-rich phase and the EE% obtained were between 80% and 100% [75].

The general results considering all the ABS tested allow the identification of a preferential migration of both molecules for the top (IL-rich) phase, which translated in high extraction efficiencies for both molecules, meaning a high affinity for the IL (top)-rich phase. This behavior can be explained by the affinity of both molecules with the ILs. This behavior can be firstly identified in the results of the alkyl chain length effect (Figure 7), where the extraction efficiencies are superior to 80% for the [$C_n$mim]Cl series. Looking more carefully at the individual partitioning behavior of paracetamol and caffeine with the alkyl chain length, it is possible to identify the crescent tendency of the molecules re-concentration in the IL-rich phase:

paracetamol = [C_2mim]Cl < [C_8mim]Cl ≈ [C_6mim]Cl < [C_4mim]Cl;

caffeine = [C_8mim]Cl < [C_2mim]Cl ≈ [C_4mim]Cl ≈ [C_6mim]Cl.

Concerning the lower alkyl chain lengths it seems that, again, the hydrophobic nature of the IL is playing a key role in the migration of both molecules, being the results of [C_8mim]Cl, one more time justified by the formation of IL' aggregates, which are unfavorable to the extraction process [73]. Regarding alkyl chain length results obtain by Freire *et al* [75]with caffeine, a similar trend is observed with a decrease on EE% with long side chain ILs, only beginning in the [C_{10}mim]Cl. This changes could be justified by the stronger salting-out salt used by Freire *et al*.

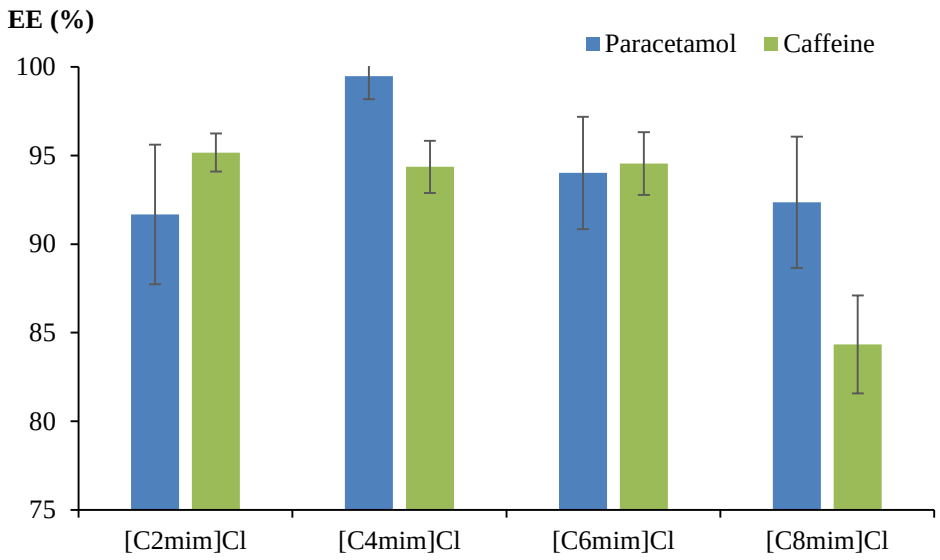


Figure 7 - Extraction efficiencies, EE (%) of paracetamol and caffeine by applying different ABS based in the series [$C_n$mim]Cl, n = 2, 4, 6 and 8.

These results seems to indicate that, not only the hydrophobic/hydrophilic nature of the ILs is important, but also that there a higher capability of both molecules to establish interactions between the imidazolium cation and the aromatic rings of caffeine and paracetamol, the so called π···π interactions. When the octanol-water partition coefficients (K_{ow}) of the active ingredients are considered, paracetamol has more affinity to organic bulks, namely IL-rich phases, K_{ow} = 3.02 [76], and this behavior is somewhat different from the caffeine profile, K_{ow} = 0.85 [76], value closer to the unit which means that caffeine has affinity for both aqueous and organic bulks). Having into consideration these data it seems that the partition of the molecules between the two phases is directly related with the 'water-molecules' and 'ILs-molecules' interactions and the hydrophobic/hydrophilic balance between the aqueous phases.

Figure 8 shows the extraction efficiency results described for different families of ILs, being the crescent order of the paracetamol and caffeine migration towards the top phase as follows:

paracetamol = [P_{4444}]Cl < [N_{4444}]Cl ≈ [C_4mim]Cl ≈ [C_4mpip]Cl ≈ [C_4mpyr]Cl ≈ [BzChol]Cl;

caffeine = [N_{4444}]Cl < [P_{4444}]Cl ≈ [C_4mim]Cl ≈ [C_4mpyr]Cl ≈ [BzChol]Cl ≈ [C_4mpip]Cl.

From this tendency, it is possible to determine the lower extraction efficiency of paracetamol for the acyclic ILs, phosphonium and quaternary ammonium when compared with the cyclic species, which can be justified by the elimination of some important interactions between the IL and paracetamol, principally the π···π interactions occurring for the aromatic ILs, namely [C_4mim]Cl. Considering the caffeine case, the extraction efficiency values are quite similar and higher than 90%, with a unclear tendency between the aromatic/non-aromatic and cyclic/non-cyclic nature.

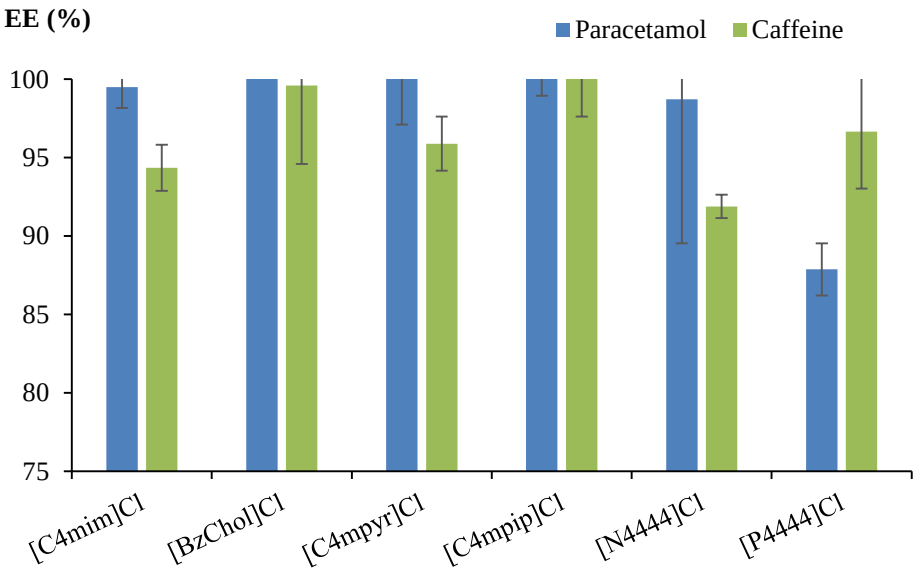


Figure 8 - Extraction efficiencies, EE (%) of paracetamol and caffeine by applying ABS with distinct [cation]Cl-based ILs.

Figure 9 shows the extraction efficiency values of paracetamol and caffeine in different ABS in order to evaluate the influence of distinct anions conjugated with the imidazolium cation. As previously analyzed, again both compounds are preferentially migrating to the top phase, following the crescent order:

paracetamol = [C_4mim][$N(CN)_2$] < [C_4mim][CH_3SO_3] < [C_4mim][CF_3SO_3] < [C_4mim]Cl ≈ [C_4mim][SCN];

caffeine = [C_4mim][CF_3SO_3] < [C_4mim][$N(CN)_2$] < [C_4mim][CH_3SO_3] ≈ [C_4mim]Cl < [C_4mim][SCN].

In fact, and despite the higher extraction efficiencies for the IL-rich phase, mainly when we analyze the paracetamol case (92.12% < EE < 100 %), when the data of caffeine were evaluated [C_4mim][CF_3SO_3] appears as the IL promoting the lower migration of caffeine for the top phase which can be probably justified by their higher salting-in nature and also by the lower water content present in the [CF_3SO_3]-rich phase, and that for some reason this behavior is only perceptible in the caffeine case. It should also be highlighted that, a similar behavior was already oberved for caffeine in presence of [C4mim][CF3SO3] and [C2mim][CF3SO3] plus the strong salting-out inorganic salt, tripotassium phosphate [75].

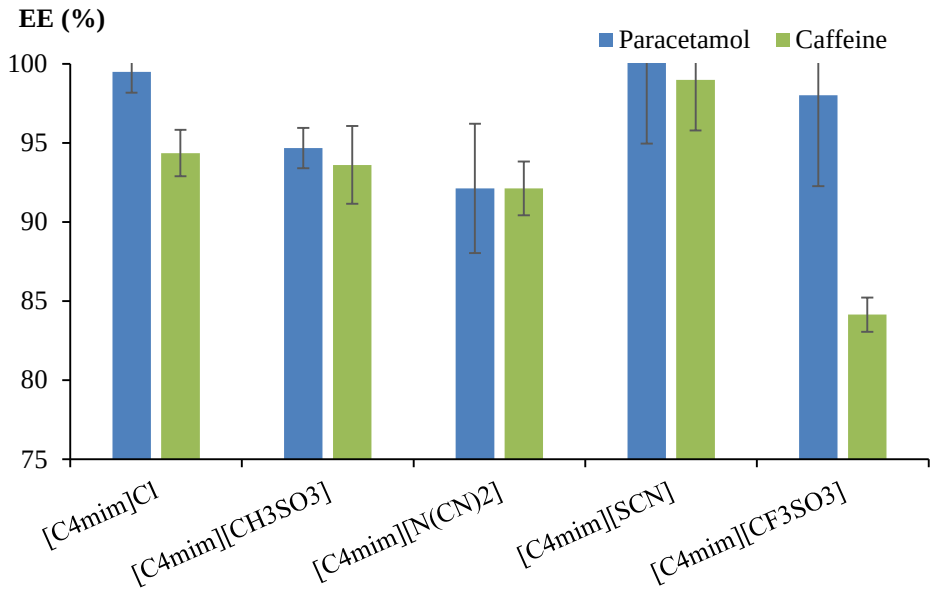


Figure 9 - Extraction efficiencies, EE(%) of paracetamol and caffeine by application of all ABS based in [C4mim]X ILs.

2.3. Conclusions

In this chapter, new phase diagrams for systems composed of a large range of ILs and the McIlvaine buffer are evaluated. With this large set of systems it was possible to identify the impact of different ILs structural features in terms of their impact in the two phase formation, namely the alkyl side chain length, the cation core and the anion moiety. In general, it was concluded the significant influence of all the structural features

investigated, being explicit the crucial role of the hydrophobic/hydrophilic nature of the ILs. It was concluded the higher aptitude of long alkyl chains to promote the ABS formation, namely ILs with hexyl side chains and acyclic cation cores, represented by phosphonium and ammonium-based ILs. The anion has also a significant impact in the phase split, which is related with its hydrophobic nature (represented by the hydrogen bond basicity parameter - β).

These ABS were applied in the partition of paracetamol and caffeine (here used as model molecules present in medicines) and several conditions were optimized, namely different alkyl chain lengths, cation cores and anion moieties, while maintaining constant the pH of the aqueous medium at 7. In general, all ABS were preferentially concentrating both structures in the IL-rich phases, which is described by extraction efficiency values from 80 to 100%.The main results have evidenced that some trends should be considered in the extraction of these compounds, like the possible formation of micelles by the application of ILs with longer alkyl chains, namely [C_8mim]Cl, or the presence of acyclic cations or even, the application of ILs based in the anion [CF_3SO_3] which is decreasing the extraction efficiency, value.

3. Extraction of paracetamol and caffeine from ALGIK

3.1. Experimental section

3.1.1. Materials

The ILs studied in this part of the experimental work were the 1-butyl-3-methylimidazolium chloride, [C_4mim]Cl (99 wt%) and tetrabutylammonium chloride, [N_{4444}]Cl (97 wt%). The ILs [C_4mim]Cl and [N_{4444}]Cl were acquired at Iolitec and Sigma-Aldrich®, respectively. The paracetamol and caffeine compounds used in this section are the same used in the optimization study. The medicine ALGIK was purchased in a local pharmacy at Aveiro (more information on Appendix C).

In the preparation of the mobile phase, it was used methanol (HPLC grade) and acetic acid (≥ 99.99 wt%), acquired from Fisher Scientific and Sigma-Aldrich®, respectively. Membrane filters (0.22 μm of pore size) were acquired on Sartorius Stedim Biotech and syringe filters (0.45 μm of pore size) were purchased at Specanalitica. The water used was double distilled, passed through a reverse osmosis system and further treated with a Milli-plus 185 water purification apparatus.

The Merck Hitachi HPLC equipment consisted of a L-6200A intelligent pump, a L-4250 UV-Vis detector and a D2500 Chromato-integrator. The analytical column used was a Merck LiChrospher 100 RP-18 (5μm).

3.1.2. Methods

A mixture point and ILs were selected to evaluate the paracetamol and caffeine extraction from the medicine based on the partitioning studies previously analyzed. The mixture composition used was: 25 wt% of McIlvaine buffer at pH 7, 25 wt% of each IL ([C_4mim]Cl or [N_{4444}]Cl) and 10 wt% of an ALGIK solution, with at *circa* 5.0 $g \cdot dm^{-3}$ of paracetamol and 0.5 $g \cdot dm^{-3}$ of caffeine (content of just one bag of ALGIK), previously filtered with a syringe filter of 0.45 μm of pore size.

Each mixture was vigorously stirred and allowed to reach the equilibrium for 12h at 298 (± 1) K to allow the separation of both phases and the complete partition of both molecules. The phases were carefully separated, and their weight and pH values measured. The paracetamol and caffeine concentrations were, in this part of the work, determined by high pressure liquid chromatography (HPLC), at 273 nm, using calibration curves previously established (Appendix E).

The HPLC mobile phase was composed of an aqueous phase (A) containing 1 M of acetic acid (filtered using a membrane filter with 0.22 µm of pore size) and an organic phase (B) containing methanol (degassed by ultrasonication). The elution was performed with 70% of A and 30% of B, using a flow rate of 0.8 $mL \cdot min^{-1}$, for about 12 minutes. The injection volume of each sample was fixed at 20 µL.

The experimental procedure for the extraction experiments was the same described in section 2.1.2.2. The equation used for the calculation of the extraction efficiencies of paracetamol and caffeine EE (%) is the same exposed before, Equation 7.

Again, the pH values of the phases were measured using a METTER TOLEDO SevenMulti pH meter within an uncertainty of ± 0.02 at 298 (± 1) K.

3.2. Results and discussion

The systems used for this work, $[C_4mim]Cl$ and $[N_{4444}]Cl$, were choosen regarding the previous studies. Both systems have high EE% values for the two molecules, allowing to evaluate the behavior with a real matrix. It was also taken into account their reported low toxicity, promising biocompatibility[77] and for being cost effective ILs[47].

The application of the previously optimized ABS with a real matrix using the medicine ALGIK was successful. The high complexity of the pharmaceutical system, due to the use of the medicine ALGIK (Appendix C), composed by paracetamol and caffeine (lower amount, not only when compared with the paracetamol content, but also when compared with the concentration previously used in the optimization study) and full of excipients, demanded the use of a more sensitive analytical technique for the proper quantification of the target molecule, namely HPLC (Appendix D). The ALGIK powder was dissolved in excess water, and left under continuous stirring overnight, to guarantee the complete dissolution of paracetamol and caffeine, while some of the insoluble excipients remained in suspension. Having into account the experimental procedure normally required and also followed in this work, for the use of an HPLC methodology, for example in what concerns the necessity of a first filtration (with the syringe filter of 0.45 µm of pore size) after the ALGIK bag content solubilisation in water, it was possible to observe the first purification step, since the filtration allowed the exclusion of some insoluble excipients. At the end, of this task a clear aqueous solutions rich in paracetamol and cafeine was obtained, and then

used for the ABS preparation. Unlike what happens with the stock solutions used in the optimization step, where the amounts of caffeine and paracetamol where accurately known, the paracetamol and caffeine concentrations in the medicine solution were not known. In order to calculate the extraction efficiency parameter, the paracetamol and caffeine concentrations present in the aqueous solutions obtained from the filtration were measured by HPLC (caffeine around 44 mg and paracetamol around 481 mg *per* bag of ALGIK).

Table 7 exhibits the extraction efficiencies for the two molecules when their extraction is carried directly from the pharmaceutical waste. From these results, it can be concluded that both molecules maintain their preference for the IL-rich phase (EE between 90 and 100%).

The application of the proposed ABS based in the McIlvaine buffer has revealed a great performance in the extraction of paracetamol, but principally caffeine (complete extraction) from the complex matrix of ALGIK as shown in Table 7. As observed in other study [65], the real systems present higher EE_{PC} than the model systems. It seems that, despite the low concentrations of the six excipients, their presence affects positively the caffeine and paracetamol migration to the IL-rich phase, being [C_4mim]Cl the only exception (the extraction efficiency from the real matrix is decreased in 10%, which can be explained by some specific interactions acting between IL-excipients' or by the exclusion of paracetamol by the stronger presence of excipients in the too phase of the [C_4mim]Cl system).

Table 8 - Extraction efficiencies, EE (%) of paracetamol and caffeine obtained in the optimization study and determined for the extraction of paracetamol from ALGIK (real matrix) by applying both ABS selected.

	Paracetamol (EE% ± sd)		**Caffeine (EE% ± sd)**	
	Pure compound	**Real matrix**	**Pure compound**	**Real matrix**
[C_4mim]Cl	99.49 ± 1.31	88.39 ± 1.47	94.36 ± 1.47	100.0
[N_{4444}]Cl	98.73 ± 9.19	99.28 ± 1.39	91.89 ± 0.75	100.0

Finally with the development of the extraction process of caffeine and paracetamol from the real matrix (ALGIK) it was possible to propose a process for the extraction of both the chemicals from anti-flu medication (Figure 10).

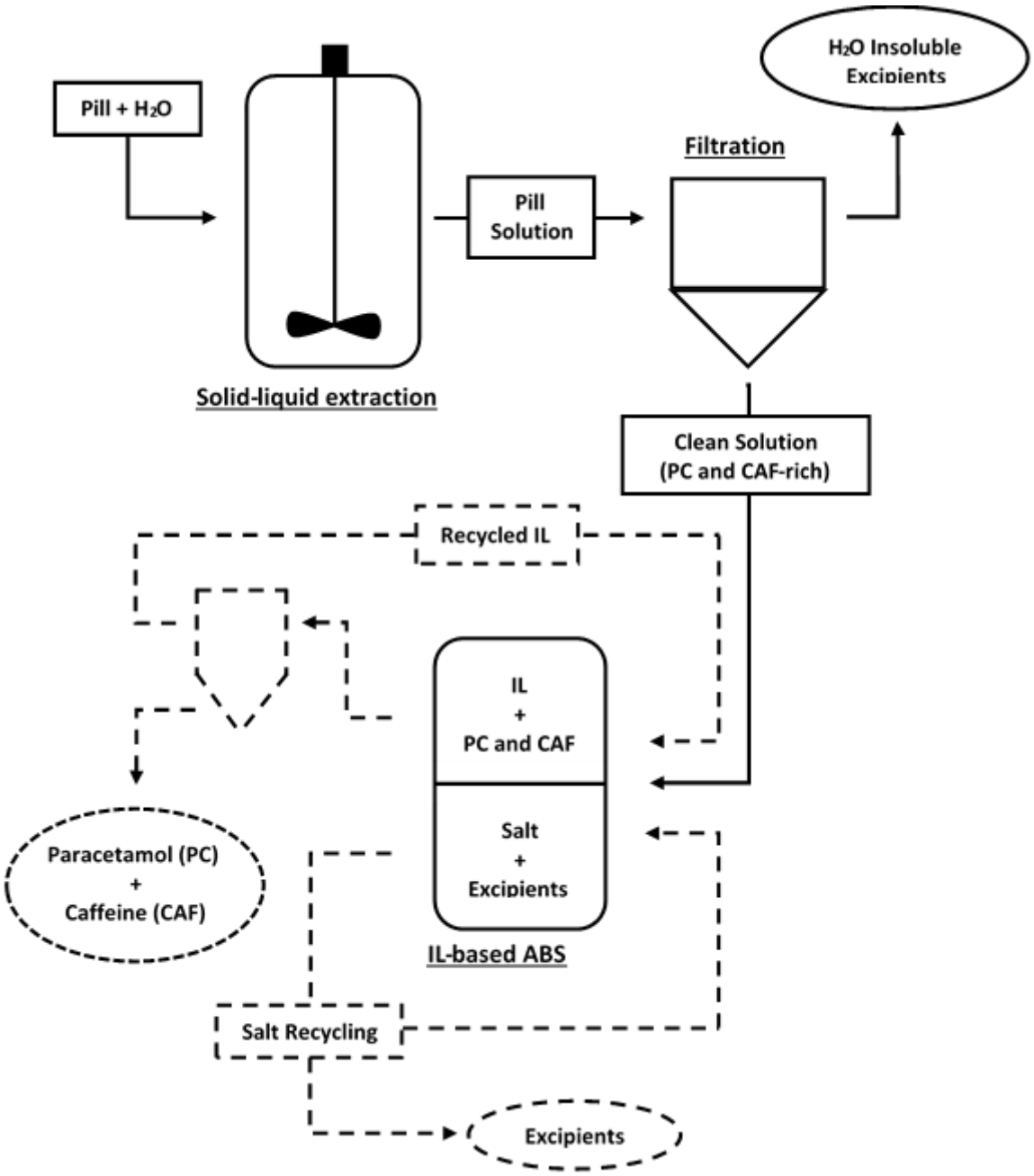


Figure 10 - Process line for the extraction of paracetamol and caffeine from anti-flu medicine.

3.3. Conclusions

The complete recovery of the paracetamol and caffeine present in the pharmaceutical sample to the top phase was accomplished, with extraction efficiencies between 90 and 100%, for both systems applied. It seems that, for most cases, the presence of excipients composing the medicine is positively influencing the extraction of both compounds for the IL-rich phase. The suitable ABS for this application is the $[N_{4444}]Cl$ + McIlvaine pH 7 + H_2O regarding the high EE% values and is low-cost when compared with ABS containing other ILs.

4. Final remarks

4.1. General conclusions

In this thesis new phase diagrams for systems composed of a large range of ILs and the McIlvaine buffer were studied. It was possible to evidence the impact of different ILs structural features, namely the alkyl side chain length, the cation core and the anion moiety. The higher aptitude of ILs to promote the ABS formation is described for Ils with long alkyl chains, acyclic cations, and more hydrophobic anions.

These ABS were applied in the partition of paracetamol and caffeine (here used as model molecules) and several conditions were optimized (alkyl chain length, IL family and the anion), while maintaining constant the pH at 7. In general, all ABS were preferentially concentrating both molecules in the IL-rich phase, being extraction efficiency values from 80 to 100% achieved. Then, two different ILs structures were selected, namely one aromatic and one acyclic IL to evaluate the extraction performance of these novel IL-based ABS based in the McIlvaine buffer extracting paracetamol and caffeine from ALGIK (anti-flu medicine), being the results also describing high extraction efficiencies (from 90 to 100%). Finally a process was proposed for extraction of this two chemicals from anti-flu medication.

4.2. Future work

In the future, it would be interesting to continue this study aiming the selective isolation of paracetamol from caffeine, by the manipulation of the pH of the media. In this context, the final step of the process diagram (Figure 10) involving the recovery of paracetamol and caffeine from the main components of the ABS, namely IL and salt will be conducted and optimized.

The process efficacy has been proved and further tests should be conducted with other active compounds with higher economical interest, namely some antidepressants.

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Appendix

A. Experimental data of the binodal curves

Table A.1 - Experimental mass fraction compositions for the system composed of [C_2mim]Cl (1) + McIlvaine buffer at pH 7 (2) + H_2O (3), at 298 (± 1) K and atmospheric pressure.

[C_2mim]Cl					
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
58.711	1.1157	33.931	10.942	22.724	22.895
50.450	2.5634	33.457	11.314	22.382	23.386
46.913	3.8062	32.744	11.978	21.504	24.519
43.366	4.9515	32.501	12.124	21.101	25.175
41.651	6.0565	32.169	12.596	20.641	25.801
40.231	6.4816	30.928	13.569	20.376	26.655
39.333	7.1765	30.772	13.892	19.321	28.182
38.898	7.3916	29.501	15.063	18.782	28.995
38.657	7.5623	29.166	15.236	18.167	29.834
37.649	8.2676	29.092	15.609	17.598	30.761
36.780	8.7899	28.206	16.707	17.185	31.602
36.690	8.8120	27.039	17.838	15.880	33.266
36.008	9.3634	26.471	18.914	15.239	34.283
35.401	9.7338	25.013	20.089	14.511	35.404
35.092	9.9374	24.068	21.300	14.309	38.049
34.590	10.480	23.491	21.901		

Table A.2 - Experimental mass fraction composition for the systems composed of [C_4mim]Cl or [C_6mim]Cl (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[C_4mim]Cl				[C_6mim]Cl	
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
54.227	2.2542	17.391	27.096	66.635	1.0747
50.591	2.7842	16.457	28.578	55.727	2.2289
45.165	4.7013	14.752	30.660	44.389	5.5894
44.133	5.1212	14.309	31.862	41.940	7.0677
42.087	6.0525	12.950	33.895	40.118	7.9764
40.249	6.9499	11.260	36.388	38.933	8.5714
38.530	7.7004	9.4031	39.094	37.758	9.0555
37.377	8.4545	8.5376	41.290	36.619	9.5689
36.349	9.1089	7.5301	43.731	35.522	10.089
35.311	9.7847			33.628	11.582
34.679	10.111			32.385	12.372
33.807	10.699			31.220	13.146
32.927	11.272			29.544	14.511
32.101	11.794			28.017	15.650
31.053	12.616			26.675	16.808
29.797	13.683			25.607	17.598
28.861	14.465			24.408	18.674
27.773	15.474			23.240	19.760
26.383	16.826			22.133	20.799
24.914	18.319			20.474	22.527
23.172	20.203			19.099	24.022
22.656	20.135			17.642	25.680
21.638	21.249			16.352	27.131
20.877	22.263			14.853	28.905
20.131	23.334			13.180	30.971
19.036	24.600				
18.308	25.755				

Table A 3 - Experimental mass fraction composition for the systems composed of [C_8mim]Cl, [C_4mpyr]Cl or [C_4mpy]Cl (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[C_8mim]Cl		**[C_4mpyr]Cl**		**[C_4mpy]Cl**	
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
66.461	1.5993	58.863	1.3978	76.820	0.9200
56.944	3.4075	42.098	5.2002	63.785	2.2878
51.031	5.0323	37.098	6.0998	29.951	12.280
46.462	7.7701	35.155	7.5914	28.641	13.271
43.912	8.7766	32.461	8.4364	26.994	14.825
40.510	10.749	30.936	9.6319	25.599	16.030
38.534	11.598	29.574	10.731	24.032	17.568
35.017	14.130	28.395	11.621	22.604	18.968
32.012	16.188	26.972	12.939	21.070	20.640
29.344	18.160	24.325	15.434	19.152	22.895
26.140	20.838	22.974	16.785	18.109	24.182
23.381	23.302	21.056	19.031	15.661	27.363
20.061	26.487	19.429	20.997		
16.302	30.377	15.872	25.921		

Table A 4 - Experimental mass fraction composition for the systems composed of [BzChol]Cl or [C_4mpip]Cl (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[BzChol]Cl				[C_4mpip]Cl	
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
50.971	5.0303	12.627	30.499	47.317	1.8293
46.822	6.2063	11.882	31.345	42.028	3.4424
44.494	7.1328	11.216	32.061	39.143	4.8445
41.285	9.1521	10.610	32.738	37.892	5.5354
39.253	9.9727	10.007	33.426	36.589	6.2483
36.627	11.680	9.4254	34.119	34.459	7.2885
35.110	12.275	8.9155	34.743	33.033	8.3215
33.012	13.600	8.3489	35.463	32.139	8.7734
31.137	14.827	7.8206	36.181	30.838	9.7475
28.966	16.505	7.3517	36.814	29.624	10.721
27.480	17.459	6.5266	37.948	28.968	11.004
25.821	18.752	5.8726	39.005	27.731	11.995
23.954	20.360			26.713	12.753
22.668	21.359			25.340	14.106
21.276	22.582			24.083	15.302
19.981	23.705			22.940	16.462
18.801	24.734			21.637	17.814
17.772	25.673			20.278	19.391
16.639	26.699			18.718	21.287
14.955	28.245			17.229	23.189
14.180	28.989			15.189	26.015
13.218	29.946			12.311	30.238

Table A 5 - Experimental mass fraction composition for the systems composed of $[N_{4444}]Cl$ (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[N_{4444}]Cl							
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
43.630	2.5213	13.564	20.699	7.3720	28.173	4.5037	32.660
39.570	3.3980	13.325	20.977	7.2480	28.357	4.4451	32.766
37.011	4.2896	13.081	21.179	7.1234	28.539	4.3888	32.894
34.805	5.0086	12.848	21.430	6.9993	28.720	4.3390	32.979
32.838	5.6637	12.620	21.681	6.8935	28.876	4.2896	33.030
31.620	6.3698	12.323	22.054	6.7859	29.021	4.2356	33.136
30.543	6.9910	12.082	22.319	6.6724	29.192	4.1851	33.237
29.501	7.5906	11.893	22.489	6.5907	29.303	4.1261	33.350
28.611	8.1419	11.580	22.911	6.4188	29.562	4.0701	33.473
27.796	8.6320	11.390	23.093	6.3280	29.692		
26.984	9.0875	11.143	23.394	6.2144	29.879		
26.272	9.5558	10.929	23.628	6.1251	30.026		
25.598	9.9407	10.717	23.906	5.9895	30.224		
24.915	10.323	10.486	24.206	5.9063	30.347		
24.270	10.707	10.279	24.478	5.8413	30.446		
23.362	11.593	10.185	24.494	5.7582	30.595		
22.816	11.929	10.039	24.626	5.6968	30.690		
22.253	12.255	9.7841	24.998	5.5794	30.864		
21.452	13.070	9.5934	25.235	5.5089	30.964		
20.955	13.395	9.4711	25.370	5.4515	31.056		
20.236	14.117	9.3178	25.588	5.3947	31.146		
19.782	14.386	9.1438	25.816	5.3622	31.206		
19.123	15.077	8.9891	26.010	5.2998	31.311		
18.497	15.703	8.8314	26.209	5.2329	31.398		
17.948	16.237	8.6522	26.482	5.1523	31.551		
17.432	16.757	8.5549	26.569	5.0985	31.646		
17.000	17.145	8.4134	26.755	5.0326	31.750		
16.540	17.593	8.2429	26.992	4.9646	31.861		
15.792	18.281	8.1032	27.171	4.9183	31.927		
15.300	18.849	7.9673	27.341	4.8459	32.059		
14.891	19.267	7.8072	27.586	4.7856	32.176		
14.498	19.688	7.6796	27.780	4.7232	32.281		
14.224	19.937	7.5506	27.954	4.6650	32.398		
13.878	20.315	7.4664	28.059	4.5824	32.511		

Table A 6 - Experimental mass fraction composition for the systems composed of $[P_{4444}]Cl$ (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[P4444]Cl							
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
45.289	2.3280	14.996	15.644	9.7805	20.439	7.3458	23.011
40.406	3.1979	14.694	15.786	9.6930	20.580	7.2752	23.029
37.217	4.1369	14.488	16.028	9.5622	20.622	7.2200	23.079
34.037	4.8323	14.272	16.252	9.4681	20.731	7.1635	23.145
32.140	5.5897	14.010	16.379	9.3850	20.834	7.0672	23.315
30.485	6.1553	13.820	16.578	9.3038	20.933	6.9995	23.347
28.800	6.7857	13.645	16.797	9.2047	21.006	6.9532	23.438
27.808	7.3182	13.458	16.994	9.1245	21.098	6.9024	23.500
26.521	7.8617	13.213	17.135	9.0430	21.203	6.8377	23.512
25.604	8.4529	13.049	17.317	8.9600	21.289	6.7941	23.599
24.578	8.8699	12.892	17.497	8.8570	21.335	6.7463	23.650
23.762	9.3511	12.719	17.682	8.7800	21.420	6.6987	23.723
23.029	9.8144	12.503	17.788	8.7077	21.509	6.6406	23.757
22.318	10.213	12.373	17.959	8.6297	21.597	6.5963	23.822
21.671	10.597	12.170	18.096	8.5592	21.693	6.5443	23.868
21.012	10.963	12.037	18.242	8.4707	21.738	6.5030	23.948
20.581	11.435	11.897	18.387	8.4051	21.859	6.4600	24.009
20.032	11.753	11.769	18.544	8.3342	21.966	6.4182	24.066
19.511	12.129	11.600	18.662	8.2439	21.995	6.3683	24.095
19.041	12.405	11.465	18.834	8.1795	22.104	6.3249	24.157
18.674	12.728	11.193	19.098	8.1141	22.208	6.2482	24.278
18.324	13.097	11.032	19.149	8.0293	22.247	6.1965	24.296
17.942	13.330	10.927	19.295	7.9691	22.343	6.1566	24.354
17.560	13.605	10.806	19.415	7.9019	22.424	6.1204	24.412
17.214	13.793	10.659	19.485	7.8252	22.466	6.0840	24.481
16.935	14.096	10.552	19.616	7.7664	22.523	6.0483	24.545
16.588	14.306	10.437	19.756	7.7135	22.612	5.9707	24.663
16.304	14.575	10.343	19.905	7.6512	22.695	5.8889	24.804
15.983	14.768	10.235	20.018	7.5785	22.728	5.8451	24.833
15.739	15.036	10.099	20.069	7.5256	22.824	5.8032	24.838
15.417	15.227	10.002	20.225	7.4517	22.844	5.7722	24.897
15.202	15.442	9.8728	20.283	7.3989	22.934	5.7377	24.959

Table A 7 - Experimental mass fraction composition for the systems composed of [C_4mim][$N(CN)_2$] (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[C_4mim][$N(CN)_2$]							
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
66.562	1.4924	16.144	16.230	9.8459	21.090	6.4101	25.272
38.813	5.8147	15.877	16.362	9.7368	21.102	6.3324	25.369
36.594	6.4935	15.642	16.532	9.5930	21.315	6.2324	25.572
34.582	7.0402	15.400	16.667	9.5006	21.350	6.1557	25.689
33.478	7.6164	15.050	17.050	9.3679	21.516	6.0805	25.802
32.342	8.2033	14.834	17.158	9.2404	21.679	6.0059	25.927
30.782	8.5791	14.606	17.321	9.1162	21.822	5.9360	26.021
29.881	9.0319	14.387	17.436	8.9996	21.951	5.8639	26.125
29.002	9.4789	14.179	17.520	8.8845	22.071	5.7968	26.226
28.220	9.8619	13.984	17.619	8.7723	22.195	5.7286	26.330
27.476	10.234	13.691	17.930	8.6973	22.217	5.6731	26.397
26.751	10.600	13.516	18.020	8.5738	22.391	5.5692	26.665
26.076	10.988	13.261	18.291	8.4511	22.528	5.4915	26.835
25.152	11.209	13.119	18.322	8.3391	22.674	5.4182	26.983
24.519	11.519	12.956	18.388	8.2596	22.692	5.3593	27.072
23.948	11.799	12.718	18.643	8.1387	22.865	5.3022	27.155
23.402	12.065	12.554	18.708	8.0219	23.039	5.2464	27.238
22.867	12.357	12.335	18.961	7.9507	23.064	5.1646	27.432
22.359	12.626	12.192	19.029	7.8368	23.239	5.0954	27.587
21.857	12.909	12.061	19.087	7.7330	23.385	5.0472	27.661
21.385	13.148	11.847	19.345	7.6333	23.520	4.9799	27.817
20.952	13.372	11.710	19.410	7.5379	23.651	4.9359	27.874
20.529	13.599	11.584	19.449	7.4376	23.792	4.8920	27.952
20.111	13.797	11.405	19.656	7.3340	23.959	4.8462	28.026
19.724	13.960	11.237	19.816	7.2374	24.100	4.7826	28.151
19.134	14.543	11.104	19.895	7.1512	24.215	4.7174	28.298
18.764	14.723	10.909	20.129	7.0558	24.351	4.6587	28.413
18.421	14.886	10.793	20.161	6.9759	24.463	4.5893	28.601
18.064	15.070	10.622	20.389	6.8924	24.574	4.5353	28.732
17.733	15.233	10.520	20.425	6.8107	24.696		
17.428	15.378	10.417	20.457	6.7331	24.804		
17.122	15.547	10.252	20.687	6.6450	24.946		
16.687	15.997	10.146	20.716	6.5596	25.070		
16.409	16.108	9.9980	20.893	6.4835	25.169		

Table A 8 - Experimental mass fraction composition for the systems composed of [C_4mim][CF_3SO_3] (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[C_4mim][CF_3SO_3]					
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
63.863	1.8156	18.042	8.8928	9.9674	14.945
53.755	2.5228	17.716	9.0649	9.7745	15.088
47.626	3.1877	17.448	9.1347	9.5909	15.448
42.976	3.7730	17.062	9.4127	9.3529	15.743
39.406	3.9999	16.496	9.6719	9.0897	15.928
37.319	4.3800	16.227	9.8437	8.7961	16.447
35.407	4.5375	15.826	10.075	8.5501	16.831
34.278	4.8825	15.506	10.415	8.3104	17.192
32.675	5.0300	15.142	10.418	8.0831	17.590
31.701	5.4168	14.855	10.667	7.7520	18.144
30.273	5.6527	14.403	10.943	7.5014	18.478
28.946	5.8123	14.203	11.010	7.3819	18.729
28.158	6.1750	13.989	11.188	7.1868	19.097
27.049	6.2989	13.753	11.406		
25.884	6.5083	13.500	11.693		
25.244	6.7641	13.158	11.802		
24.218	6.9459	12.887	12.144		
23.385	7.0075	12.556	12.324		
22.842	7.2839	12.203	12.574		
22.363	7.4425	12.020	12.808		
21.894	7.6744	11.816	13.037		
21.134	7.7793	11.624	13.274		
20.728	7.9425	11.274	13.628		
20.321	8.1028	10.935	13.832		
19.611	8.4291	10.714	14.172		
19.046	8.4595	10.574	14.312		
18.688	8.6382	10.350	14.496		
18.372	8.7678	10.109	14.798		

Table A 9 - Experimental mass fraction composition for the systems composed of [C_4mim][SCN] or [C_4mim][CH_3SO_3] (1) + McIlvaine buffer (2) + H_2O (3) at 298 (± 1) K and atmospheric pressure.

[C_4mim][SCN]				**[C_4mim][CH_3SO_3]**	
100 w_1	**100 w_2**	**100 w_1**	**100 w_2**	**100 w_1**	**100 w_2**
38.664	4.4414	15.140	11.757	67.781	2.3544
35.752	5.1602	14.882	12.021	48.018	6.4771
33.210	5.8149	14.180	12.312	38.402	10.076
30.606	6.3556	13.755	12.367	36.502	11.311
29.145	6.9362	13.436	12.560	35.973	11.789
27.563	7.3855	11.479	13.620	33.552	13.397
26.328	7.8693	11.091	13.974	33.507	13.502
24.905	8.1933	10.830	13.989	32.399	14.462
23.695	8.5199	10.509	14.474	31.099	15.600
22.821	8.8807	10.210	14.690	29.837	16.730
22.027	9.2207	9.9120	14.885	28.677	17.869
21.267	9.5250	9.6342	15.071	28.393	18.068
20.399	9.7080			27.734	18.974
19.571	9.8752			26.381	20.426
19.173	10.225			24.903	22.109
18.575	10.504			23.620	23.795
18.029	10.716			21.521	26.179
17.405	10.854			19.711	28.529
16.931	11.063			18.876	30.173
16.467	11.267			15.913	33.719
15.929	11.388			12.623	37.764
15.525	11.589			11.045	40.780

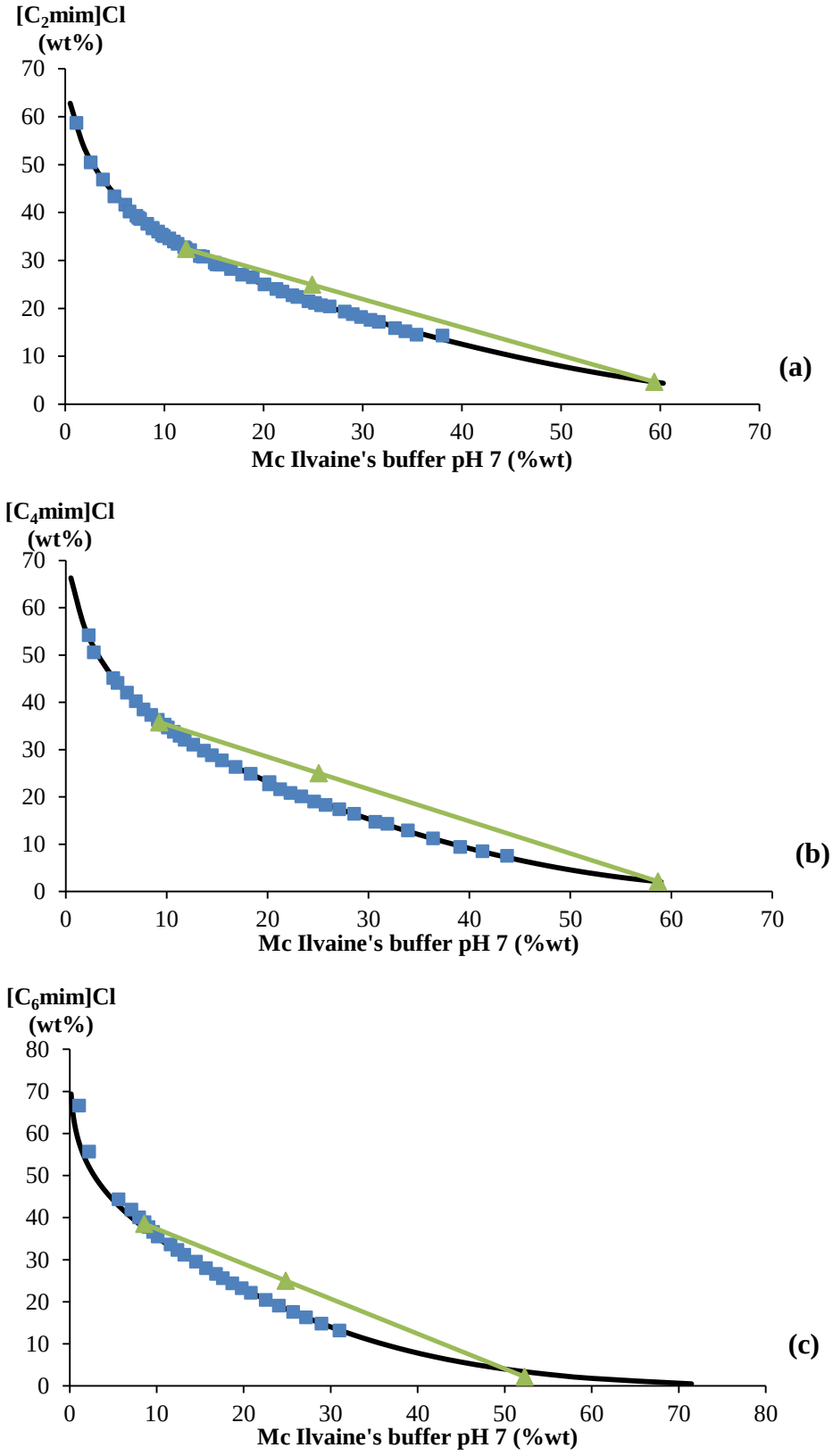


Figure A 1 - Phase diagram of the systems [C_2mim]Cl (a), [C_4mim]Cl (b) and [C_6mim]Cl (c) + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (-▲-); fitting of the experimental data through Equation 1 (▬).

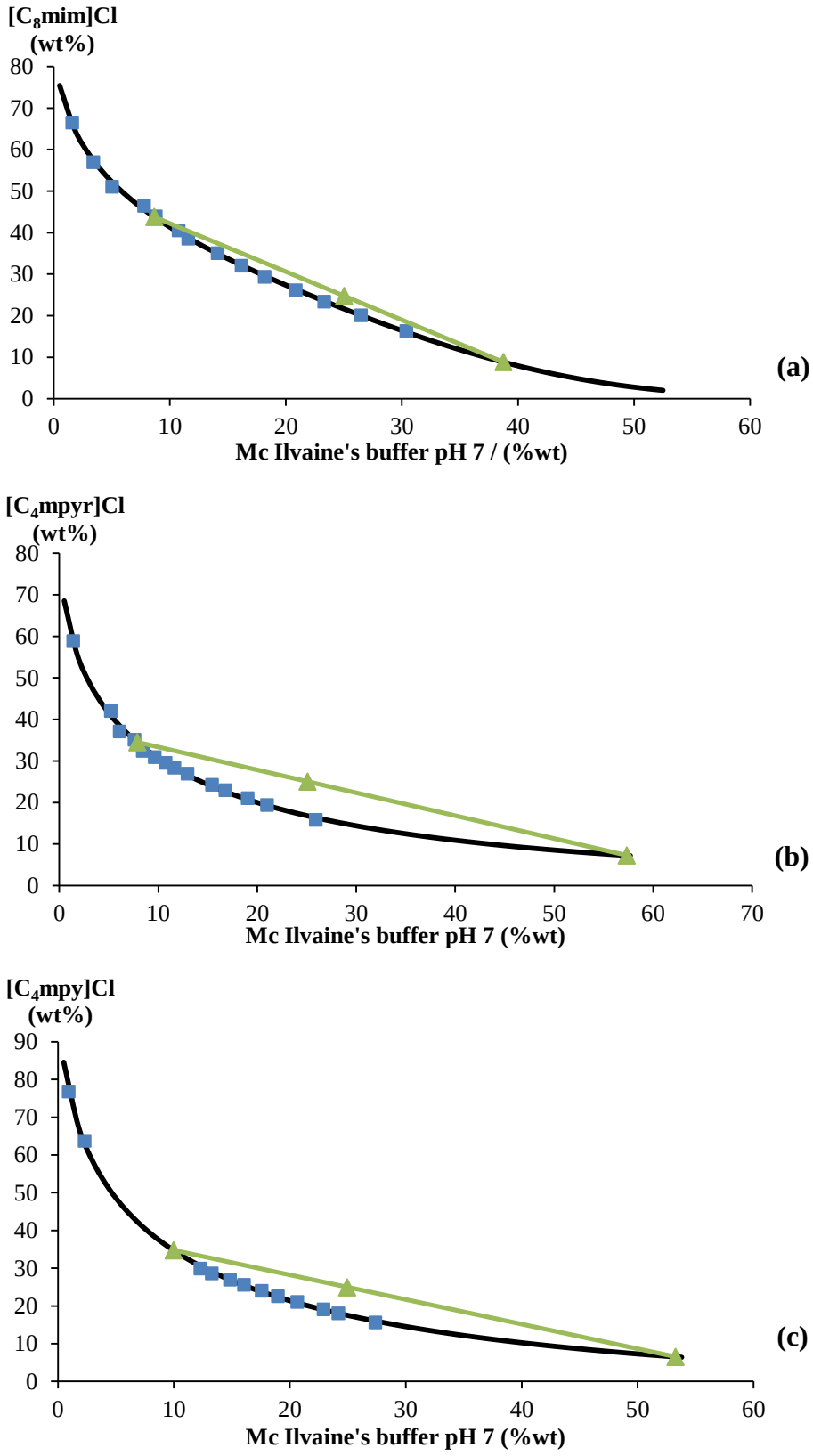


Figure A 2 - Phase diagram of the systems [C_8mim]Cl (a), [C_4mpyr]Cl (b) and [C_4mpy]Cl (c) + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (-▲-); fitting of the experimental data through Equation 1 (▬).

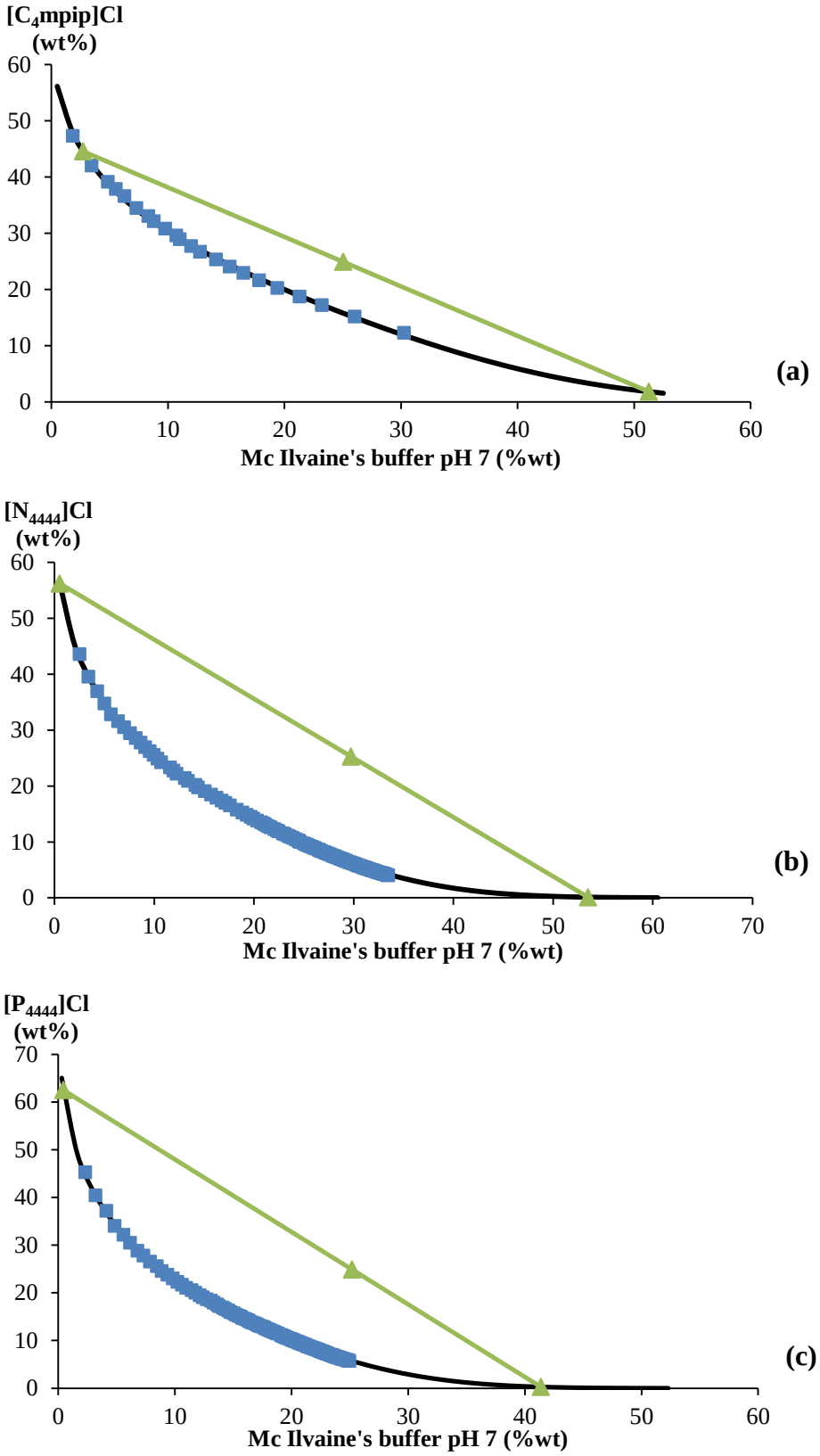


Figure A 3 - Phase diagram of the systems [C_4mpip]Cl (a), [N_{4444}]Cl (b) and [P_{4444}]Cl (c) + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (-▲-); fitting of the experimental data through Equation 1 (▬).

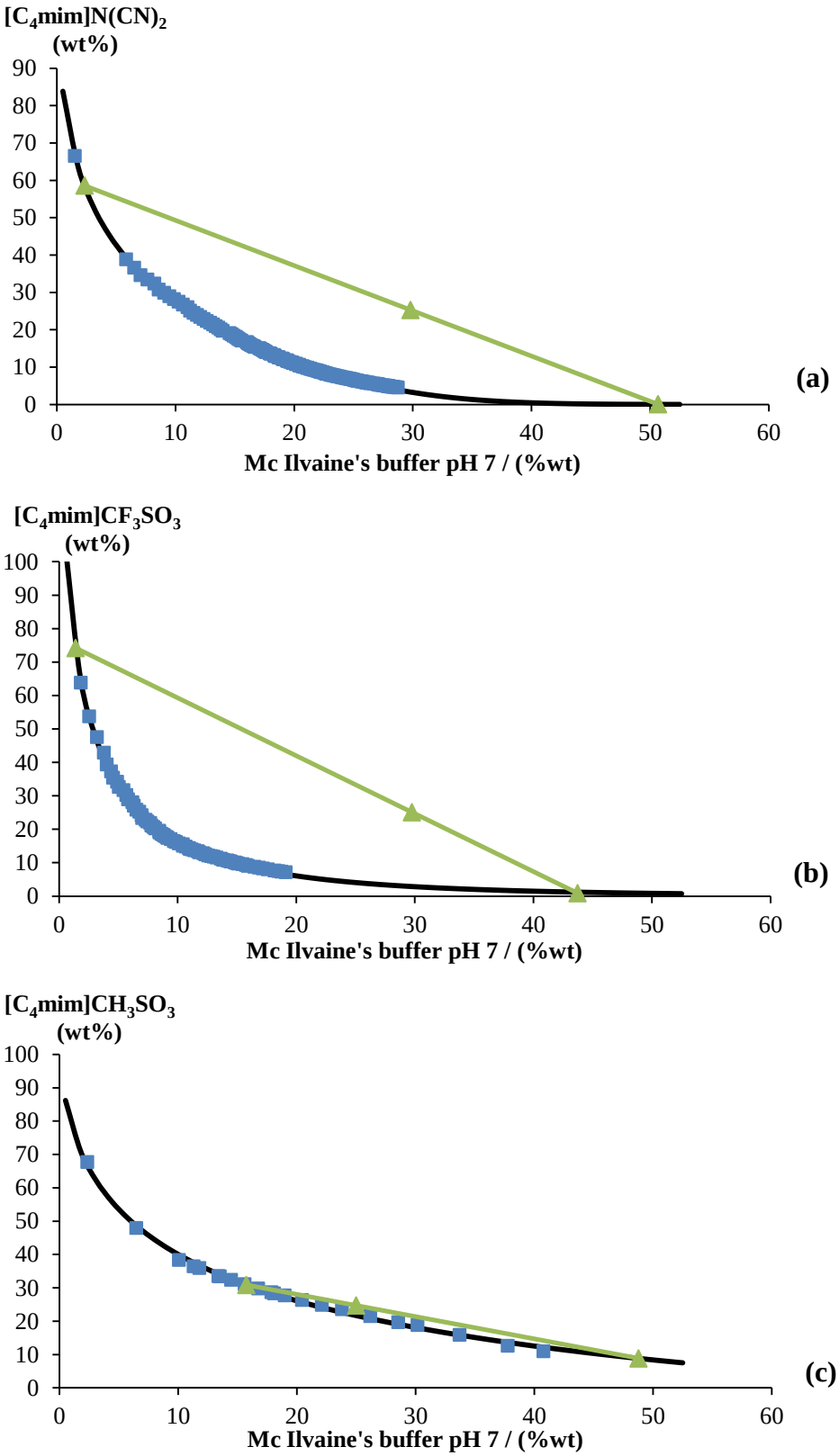


Figure A 4 - Phase diagram of the systems $[C_4mim]N(CN)_2$ (a), $[C_4mim]CF_3SO_3$ (b) and $[C_4mim]CH_3SO_3$ (c) + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (▲); fitting of the experimental data through Equation 1 (▬).

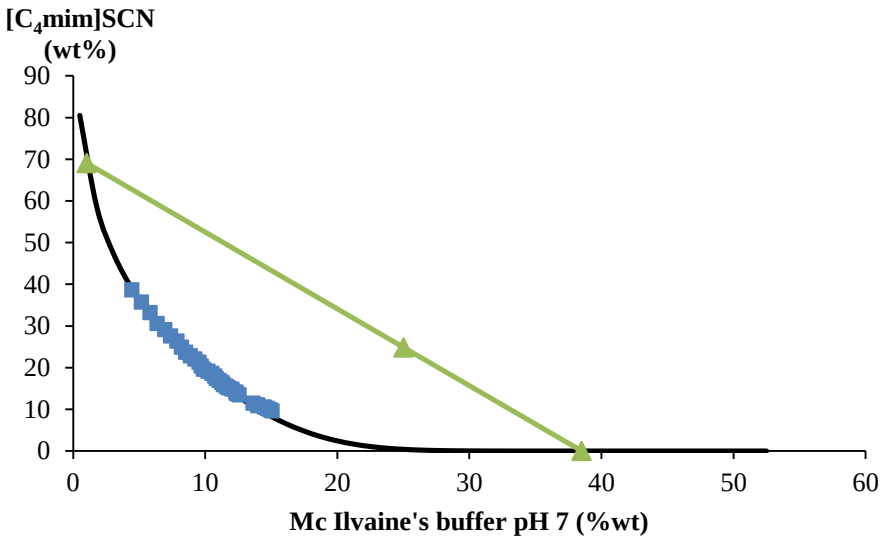


Figure A 5 - Phase diagram of the systems [C_4mim]SCN + McIlvaine buffer at pH 7 + H_2O at 298 (± 1) K: binodal curve data (■); TL data (-▲-); fitting of the experimental data through Equation 1 (▬).

B. Experimental data - Partitioning of paracetamol and caffeine.

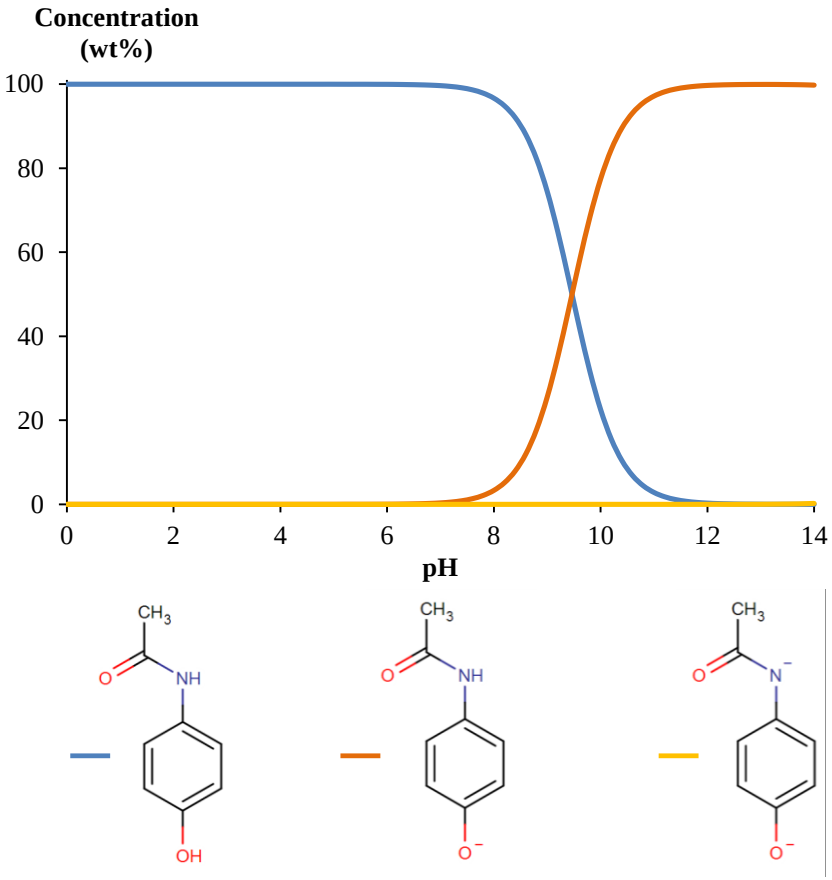


Figure B 2 - Speciation diagram of paracetamol. Adapted from [78].

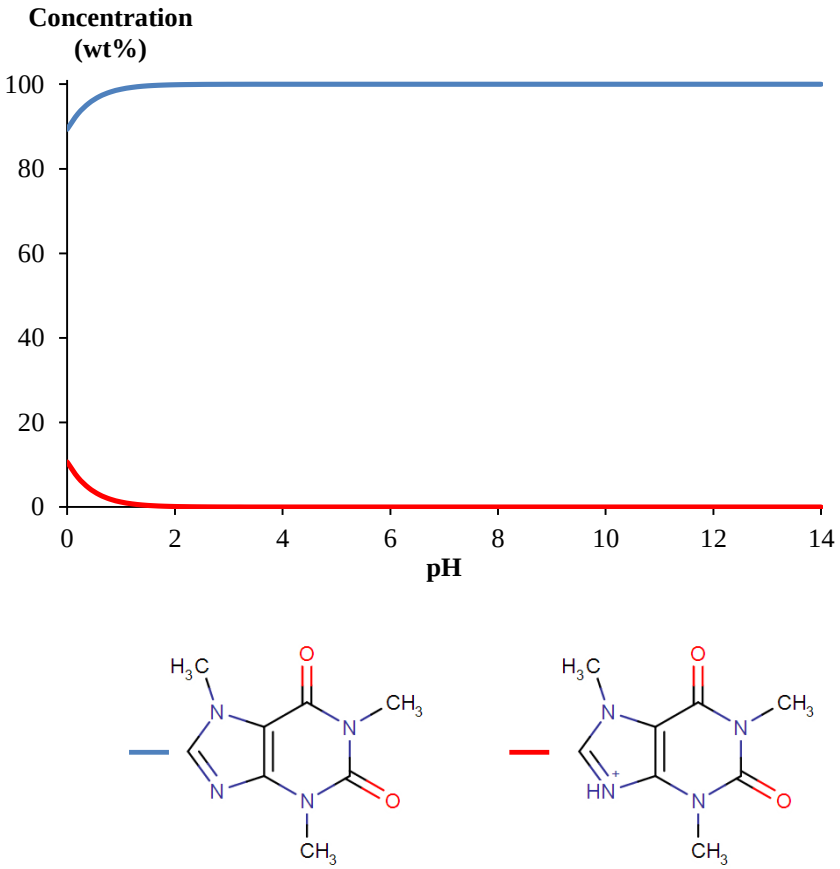


Figure B 1 - Speciation diagram of paracetamol. Adapted from [78].

C. ALGIK ID

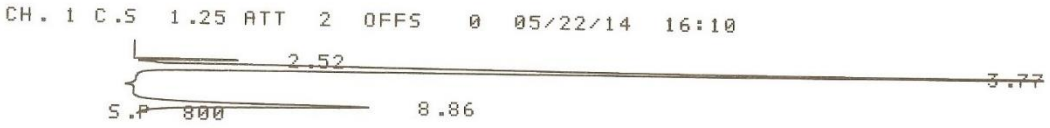
The medicine, ALGIK, is produced by Sofarimex – Indústria Química e Farmacêutica, Lda and it was acquired at a local pharmacy in Aveiro. The package contains 20 Kraft/polyethylene bags with the composition presented in Table D 1.

Table D 1 - ALGIK composition per bag.

Chemical	Quantity (mg)
Paracetamol	500
Caffeine	50
Monohydrated lactose	nd
Sodium stearyl fumarate	nd
Aerosil 200	nd
Aluminium oxide C	nd
Orange tetrarome	nd
Aspartame (E951)	nd

nd – not described.

D. HPLC Chromatograms



D-2500 05/22/14 16:10

METHOD: TAG: 24 CH: 1

FILE: 0 CALC-METHOD: AREA% TABLE: 0 CONC: AREA

NO.	RT	AREA	CONC	BC
1	2.52	3276	2.000	BV
2	3.77	124935	76.260	VB
3	8.86	35617	21.740	BB
TOTAL		163828	100.000	
PEAK REJ :		0		

Figure B 3 – Chromatogram of standard solution.

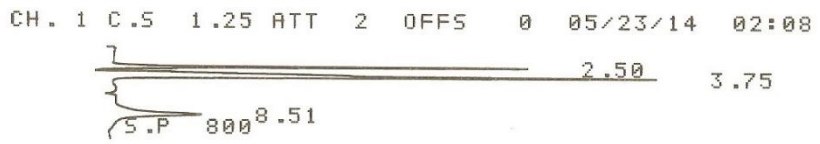
D-2500 05/23/14 02:33

METHOD: TAG: 11 CH: 1

FILE: 0 CALC-METHOD: AREA% TABLE: 0 CONC: AREA

NO.	RT	AREA	CONC	BC
1	2.52	3421	3.964	BB
2	3.74	70260	81.416	BB
3	8.60	12617	14.620	BB
TOTAL		86298	100.000	
PEAK REJ :		0		

Figure B 4 - Chromatogram of the pharmaceutical drug ALGIK.



D-2500 05/23/14 02:08

METHOD: TAG: 9 CH: 1

FILE: 0 CALC-METHOD: AREA% TABLE: 0 CONC: AREA

NO.	RT	AREA	CONC	BC
1	2.50	18450	23.238	BB
2	3.75	50740	63.907	BB
3	8.51	10207	12.856	BB
TOTAL		79397	100.000	
PEAK REJ :		0		

Figure B 5 - Chromatogram of the top phase of the ABS composed of $[N_{4444}]Cl$ + McIlvain buffer at pH 7 + H_2O.

D-2500 05/29/14 15:23

METHOD: TAG: 7 CH: 1

FILE: 0 CALC-METHOD: AREA% TABLE: 0 CONC: AREA

NO.	RT	AREA	CONC	BC
1	2.45	24747	30.827	BB
2	3.47	18304	22.801	BV
3	3.73	28706	35.759	VB
4	8.38	8520	10.613	BB
TOTAL		80277	100.000	
PEAK REJ :		0		

Figure B 6 - Chromatogram of the ABS top phase with $[C_4mim]Cl$ + McIlvain buffer at pH 7 + H_2O.

E. Calibration curves

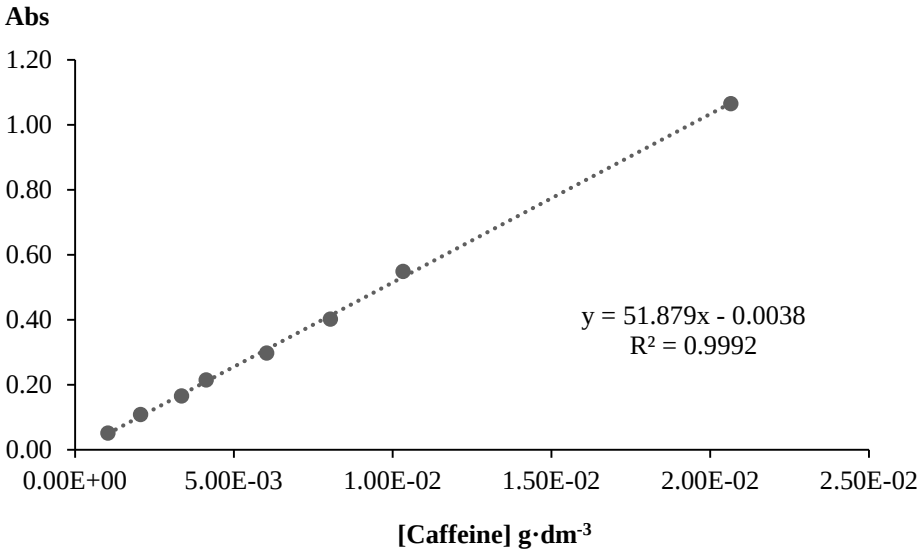


Figure C 1 - Calibration curve for caffeine at λ = 273 nm.

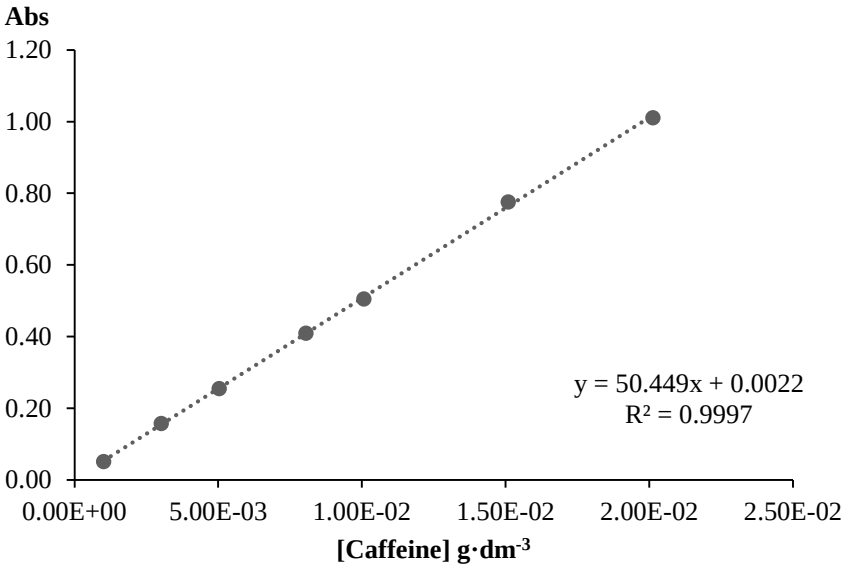


Figure C 2 - Calibration curve for caffeine at λ = 273 nm.

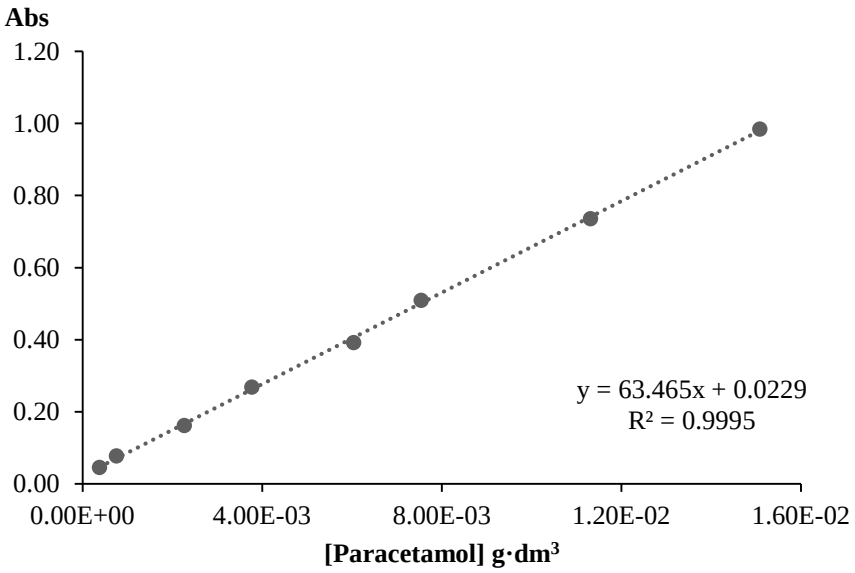


Figure C 3 - Calibration curve for paracetamol at λ = 243 nm.

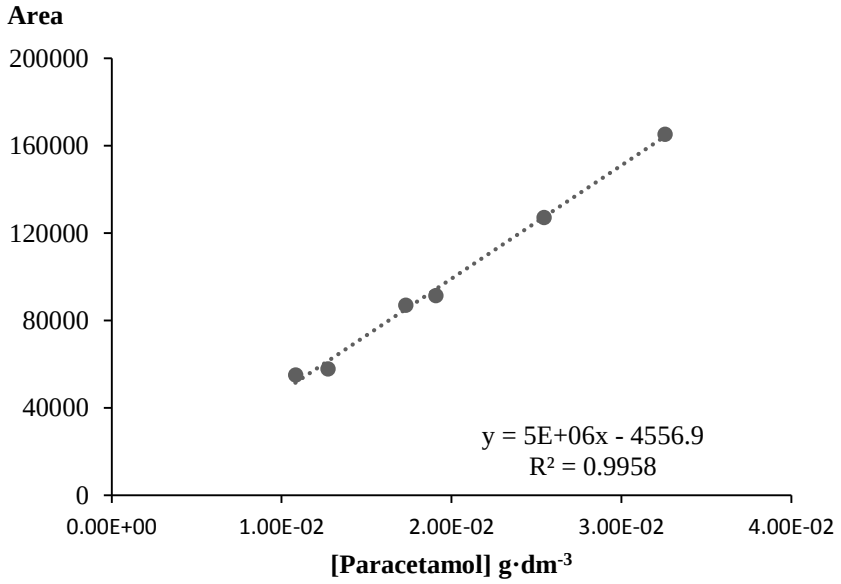


Figure C 4 – HPLC calibration curve for paracetamol.

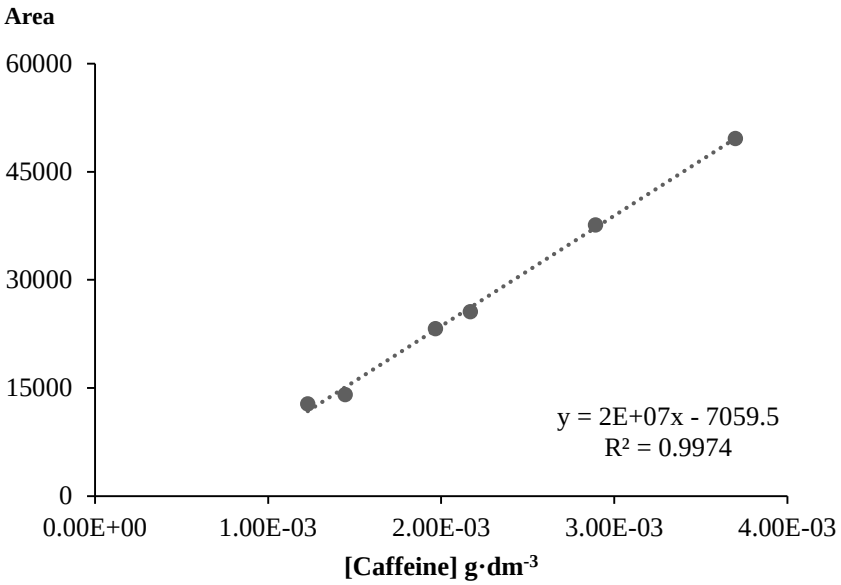


Figure C 5 – HPLC calibration curve for caffeine.