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SEPARATION OF *p*-AMINOBENZOIC ACID BY REACTIVE EXTRACTION

1. MECHANISM AND INFLUENCING FACTORS

The comparative study on the reactive extraction of p-aminobenzoic acid with Amberlite LA-2 and D2EHPA in two solvents with different polarity (n-heptane and dichloromethane) indicated that the extractant type and solvent polarity control the extraction mechanism. Thus, the reactive extraction with Amberlite LA-2 occurs by means of the interfacial formation of an aminic adduct with three extractant molecules in low-polar solvent, or of a salt with one extractant molecule in higher polar solvent. Similarly, the extraction with D2EHPA is based on the formation of an acidic adduct with two extractant molecules in n-heptane, or of a salt with one extractant molecule in dichloromethane. The most efficient extraction has been reached for the combination Amberlite LA-2-dichloromethane.

Key words: p-aminobenzoic acid; reactive extraction; Amberlite LA-2; di-(2-ethylhexyl) phosphoric acid; extraction constant; distribution coefficient.

p-Aminobenzoic acid (PABA), also called vitamin B₁₀ or factor R, was found to be the part of the folic acids, its chemical structure being given in Figure 1. Being component of the pteroylglutamate, it is considered to act as provitamin for some bacteria and a growth factor for some superior animals, in the human body possessing the capacity to synthesize folates [1].

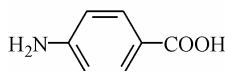


Figure 1. The chemical structure of *p*-aminobenzoic acid.

p-Aminobenzoic acid is used in cosmetics as additive in sunscreen lotions. The medical applications are for skin protection against vitiligo, sclerodermy and male infertility treatment. It is also used in diagnostic tests for the state of a gastrointestinal tract in medicine. It was established that PABA is an inducer of endogenous interferon and immunomodulator, displays a virucidal, synergistic antiviral effect when combined with chemical drugs and possesses the pro-

erties of a direct anticoagulant [2]. Although this compound is not considered to be an essential nutrient, it is included in vitamin B or multivitamin(s) supplements [1].

The most recent methods for the PABA production are the chemical synthesis using methyl-4-formylbenzoate as a starting material [3] or biosynthesis by mutant strains of *Escherichia coli* [4]. In both cases, the separation stages are complex and require a large energy and material consumption.

For many technologies, the liquid-liquid extraction constitutes a viable solution, due to its technical accessibility and high efficiency. But, its application is limited for the ionizable compounds, namely the carboxylic acids, due to their low solubility in usual organic solvents. In these cases, the performances of the extraction process can be enhanced by a reactive extraction with an extractant added into the organic phase. Thus, the reactive extraction using extractants of organophosphoric or height molecular amines types have been successfully applied to the separation of some carboxylic acids, namely as: acetic acid, lactic acid, citric acid, succinic acid, malic acid, ascorbic acid or beta-lactamic antibiotics [5-15].

Due to the insolubility of PABA in organic solvents immiscible with water, its separation by physical extraction is impossible. But its extraction could become possible by adding into the solvent of an extractant

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which could react with this acid, leading to the formation of a hydrophobic compound, as in the case of the above mentioned acids extraction. Because the chemical structure of PABA contains an acidic group, -COOH, and a basic one, -NH₂, the reactive extraction could become possible by using extractants of aminic or organophosphoric acid types. For this purpose, the comparative study on the reactive extraction of PABA with Amberlite LA-2 (lauryl-trialkyl-methylamine) and di-(2-ethylhexyl) phosphoric acid (D2EHPA) is presented in the paper. The extraction mechanisms and the influencing factors are discussed in both cases. Because the solvent polarity is an important factor that controls the extraction efficiency, the extraction mechanisms and optimum conditions have been analyzed in direct correlation with the polarity of the two used solvents (*n*-heptane and dichloromethane).

EXPERIMENTAL

The experiments have been carried out using an extraction column with vibratory mixing, which offers high interfacial area and the possibility to reach the equilibrium state rapidly. The laboratory equipment has been described in detail in previous papers [9,15]. The phase mixing was made by means of a perforated disk with 45 mm diameter and 20 % free section. The vibrations had a frequency of 50 s⁻¹ and 5 mm amplitude. The perforated disk position was maintained at the initial contact interface between the aqueous and organic phases. The extraction time was 1 min at a constant temperature of 25 °C. The resulted emulsion was broken in a centrifugal separator at 8000 rpm.

The initial concentration of PABA in aqueous solution was of 5 g/l (3.65×10⁻² M). The reactive extraction was made with Amberlite LA-2 and D2EHPA respectively, both solved in two solvents with different dielectric constants (*n*-heptane with the dielectric constant of 1.90, and dichloromethane with dielectric constant of 9.08, respectively, both at 20 °C [16]). The extractant concentration in the organic phase was varied between 20 and 120 g/l (5.3×10⁻²-0.32 M for Amberlite LA-2, 6.1×10⁻²-0.36 M for D2EHPA). The volumetric ratio of aqueous and organic phase was of 1 (30 ml of each phase).

The pH value of the initial aqueous solution was varied between 1 and 8. The pH adjustment was made with a solution of 3 mass % sulfuric acid or 3 mass % sodium hydroxide, function on the prescribed pH value. The pH values were determined using a digital pH-meter of Consort C836 type and have been

recorded throughout each experiment. Any pH change was noted during the extraction experiments.

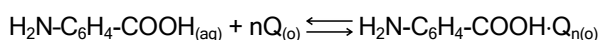
The extraction process was analyzed by means of the extraction degree and a distribution coefficient. The extraction degree has been defined as the ratio between the solute concentration in the extract phase and its initial concentration in the aqueous solution. For calculating these parameters, the PABA concentrations in the initial solution and raffinate were determined using the spectrophotometry, by measuring these phases absorption at 267 nm [17]. For calculating the PABA concentration in the organic phase the mass balance has been used.

Each experiment has been carried out for three or four times, under the identical conditions, the average value of the considered parameters being used. The maximum experimental error was of ±3.93 %.

RESULTS AND DISCUSSION

Indifferent of the extractant type, the reactive extraction of PABA occurs by means of the formation of a strong hydrophobic compound at the interface between the aqueous and organic phases. The chemical structure of this compound and the extraction parameters depend on the extractant type.

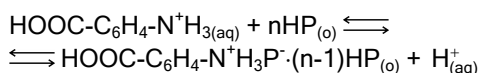
The carboxylic group of the PABA is involved in the reactive extraction process with Amberlite LA-2 (Q). The interfacial interactions between the acid and the extractant could be of hydrogen bonding type with the undissociated carboxylic groups, or of ionic type, if the acid dissociates in the aqueous solution:



Furthermore, in function of the structures of system components and solvent polarity, the acidic or aminic adducts could be formed at the interface [5,18]. However, as it was observed for the reactive extraction with Amberlite LA-2 of other compounds having voluminous molecules and due to the initial concentration of PABA, which is lower than that of Amberlite LA-2, it could be assumed that the formation of acidic adducts is steric hindered. Therefore, the interfacial compounds could be of ammonium salt type, formed by neutralization of the carboxylic group with one extractant molecule, or by aminic adducts type, where $n \geq 2$ [5,7,18]. In the second case, the formation of these molecular associations is more pronounced in low-polar solvents and increases the hydrophobicity of the interfacial compound [5].

Since the PABA contains the N aminic, it could react with acidic extractants, such as D2EHPA (HP). In this case, the reactive extraction could occur by

means of an interfacial reaction of ion-exchanging type, which requires the acid to be on protonated form into the aqueous solution:



The pH value of the aqueous phase exhibits a significant influence on a reactive extraction degree in both cases. As it can be observed from Figure 2, for both used extractants a certain value of pH value corresponding to the maximum of the reactive extraction degree was recorded.

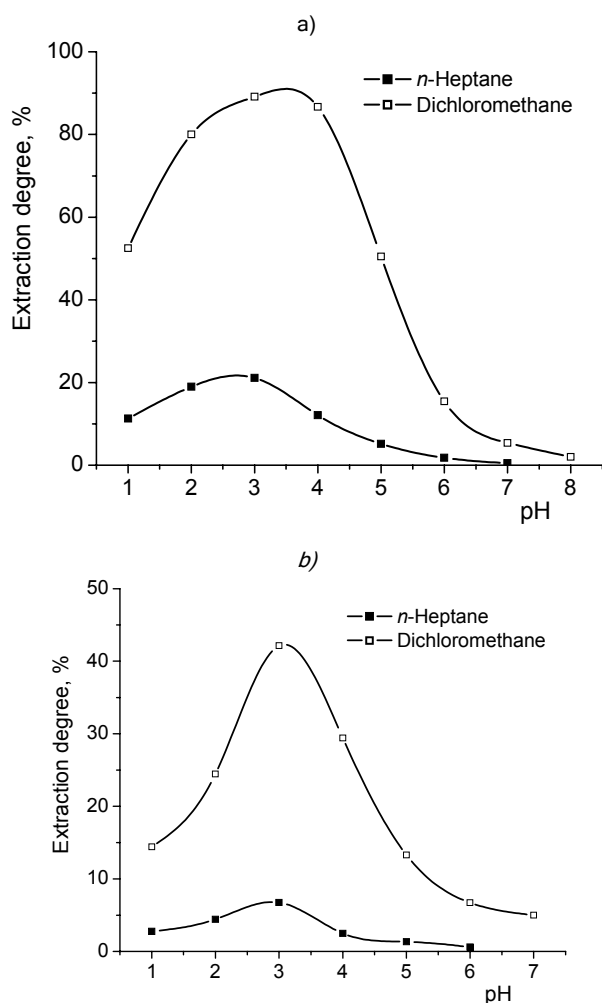


Figure 2. The influence of pH value of the aqueous phase on the efficiency of the reactive extraction of PABA with Amberlite LA-2 (a) and D2EHPA (b) (extractant concentration: 20 g/l).

For the extraction with Amberlite LA-2, the maximum is the result of the aminic group ionization at strong acidic pH domain ($pK_{a1} = 2.50$ [16]), this phenomena limiting the extraction efficiency. But, the increase of pH value induces the dissociation of -COOH group ($pK_{a2} = 4.87$ [16]) and, implicitly, the reduction

of the extraction yield. Thus, as the result of the two contrary effects generated by the increase of pH, the optimum value of the aqueous phase is pH 3 for the reactive extraction with Amberlite LA-2, indifferent of the used solvent.

The reactive extraction with D2EHPA is possible if the N aminic is in ammonium form at low pH value. But, at highly acidic pH, the extractant is protonated too and thus unable to react with the acid [19]. Due to these two opposite phenomena, the optimum pH value for the extraction with D2EHPA is also 3, but for other reasons than in the case of the extraction with Amberlite LA-2.

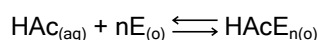
The efficiency of the physical extraction of PABA with the two solvents does not allow the possibility to apply this technique practically (at pH 3, the extraction degrees were of 11.2 % in dichloromethane and 3.1 % in *n*-heptane).

By comparing the reactive extraction degrees for a given extractant concentration of 20 g/l, at the optimum pH value of 3, it can be concluded that Amberlite LA-2 is the most efficient extractant (the difference between the corresponding extraction yields recorded for the two extractants is 47 % for dichloromethane and 14.5 % for *n*-heptane).

For analyzing the extraction mechanism of PABA (HAc) with the two extractants, it was assumed that *n* extractant (E) molecules and one acid molecule participate in the formation of the interfacial compound, through hydrogen bonds as it was discussed above. Thus, the mechanisms of the reactive extraction with the two extractants will be separately analyzed.

Reactive extraction with Amberlite LA-2

The reactive extraction of PABA with Amberlite LA-2 can be described by the following interfacial equilibrium:



Therefore, the distribution coefficient, *D*, is calculated with the relationship:

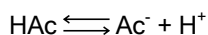
$$D = \frac{[\text{HAcE}_{n(o)}]}{[\text{HAc}_{(\text{aq})}]} \quad (1)$$

where $[\text{HAc}_{(\text{aq})}]$ and $[\text{HAcE}_{n(o)}]$ symbolize the overall concentrations of PABA and the extracted compound at the equilibrium state.

According to the interfacial equilibrium, the extraction constant, K_E , can be calculated with the following expression:

$$K_E = \frac{[\text{HAcE}_{n(o)}]}{[\text{HAc}_{(\text{aq})}][\text{E}_{(o)}]^n} \quad (2)$$

The study on the mechanism of the reactive extraction was carried out at the optimum pH value of 3, therefore the concentration of undissociated acid from the aqueous phase, $[\text{HAc}_{(\text{aq})}]$, is calculated by means of its overall concentration in the aqueous phase, $[\overline{\text{HAc}_{(\text{aq})}}]$, and the dissociated acid concentration, $[\text{Ac}^-_{(\text{aq})}]$. The dissociation constant, K_{a2} , corresponds to the following dissociation equilibrium:



and is determined with the relationship:

$$K_{a2} = \frac{[\text{Ac}^-_{(\text{aq})}][\text{H}^+]}{[\text{HAc}_{(\text{aq})}]} \quad (3)$$

Thus, the concentration of undissociated PABA is:

$$[\text{HAc}_{(\text{aq})}] = \frac{[\overline{\text{HAc}_{(\text{aq})}}]}{1 + \frac{K_{a2}}{[\text{H}^+]}} \quad (4)$$

Therefore, by combining the Eqs. (1), (2) and (4), the following expression for the distribution coefficient, D , is obtained:

$$D = K_E [E_{(o)}]^n \left(1 + \frac{K_{a2}}{[\text{H}^+]}\right) \quad (5)$$

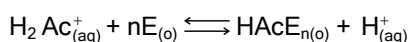
The correlation (5) represents the equation of a straight line in a logarithmic form:

$$\ln D - \ln \left(1 + \frac{K_{a2}}{[\text{H}^+]}\right) = \ln K_E + n \ln [E_{(o)}] \quad (6)$$

Because the initial concentration of the extractant is higher than the initial concentration of PABA, $[E_{(o)}]$ could be assumed to be the initial concentration of Amberlite LA-2 in the organic phase. Consequently, from the slope of the straight line given by Eq. (6) it is possible to determine the number of extractant molecules, n , which participate in the formation of the interfacial compound, and from its intercept the value of the extraction constant, K_E .

Reactive extraction with D2EHPA

The study on the mechanism of the reactive extraction with D2EHPA was also carried out at pH 3. In this case, the N of the aminic group is the active center of the solute, the extraction efficiency being controlled by its ionization capacity. The interfacial reaction could be described by the following equilibrium:



According to this interfacial equilibrium, the distribution coefficient, D , is:

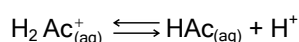
$$D = \frac{[\text{HAcE}_{n(o)}]}{[\text{H}_2\text{Ac}^+_{(\text{aq})}]} \quad (7)$$

where $[\text{H}_2\text{Ac}^+_{(\text{aq})}]$ and $[\text{HAcE}_{n(o)}]$ represent the overall concentrations of the considered components at the equilibrium state.

The extraction constant, K_E , can be calculated with the following relationship:

$$[\text{HAcE}_{n(o)}]^2 = K_E [\text{H}_2\text{Ac}^+_{(\text{aq})}] [E_{(o)}]^n \quad (8)$$

The ionization constant of N aminic, K_{a1} , is calculated by means of the following equilibrium:



$$K_{a1} = \frac{[\text{HAc}_{(\text{aq})}][\text{H}^+]}{[\text{H}_2\text{Ac}^+_{(\text{aq})}]} \quad (9)$$

The concentration of protonated PABA from the aqueous phase, $[\text{H}_2\text{Ac}^+_{(\text{aq})}]$, is calculated by means of its overall concentration in the aqueous phase, $[\overline{\text{HAc}_{(\text{aq})}}]$, and its non-ionized molecules concentration, $[\text{HAc}_{(\text{aq})}]$:

$$[\text{H}_2\text{Ac}^+_{(\text{aq})}] = [\overline{\text{HAc}_{(\text{aq})}}] - [\text{HAc}_{(\text{aq})}] \quad (10)$$

$$[\text{HAc}_{(\text{aq})}] = K_{a1} \frac{[\text{H}_2\text{Ac}^+_{(\text{aq})}]}{[\text{H}^+]} \quad (11)$$

$$[\text{H}_2\text{Ac}^+_{(\text{aq})}] = \frac{[\overline{\text{HAc}_{(\text{aq})}}]}{1 + \frac{K_{a1}}{[\text{H}^+]}} \quad (12)$$

Thus, the expression for the distribution coefficient becomes:

$$D^2 = \frac{[\text{HAcE}_{n(o)}]^2}{[\text{H}_2\text{Ac}^+_{(\text{aq})}]^2} = \frac{K_E [E_{(o)}]^n}{[\text{H}_2\text{Ac}^+_{(\text{aq})}]} = \frac{K_E [E_{(o)}]^n \left(1 + \frac{K_{a1}}{[\text{H}^+]}\right)}{[\overline{\text{HAc}_{(\text{aq})}}]} \quad (13)$$

The logarithmic form of Eq. (13) gives a straight line equation:

$$2 \ln D^2 - \ln \frac{\left(1 + \frac{K_{a1}}{[\text{H}^+]}\right)}{[\overline{\text{HAc}_{(\text{aq})}}]} = \ln K_E + n \ln [E_{(o)}] \quad (14)$$

which is used for the n and K_E graphical determination, similar to the calculations for the reactive extraction with Amberlite LA-2 ($[E_{(o)}]$ could be also assumed to be equal with the initial concentration of D2EHPA in the organic solvent).

For determining the number of extractant molecules which reacts with PABA, in correlation with the solvent polarity, the influence of the extractant concentration of the extraction efficiency has been ana-

lyzed for the two extractants solved in *n*-heptane and dichloromethane, respectively.

The experimental results, plotted in Figure 3, indicated that the extraction efficiency increases with the solvent polarity for used extractants (for 40 g/l extractant in an organic solvent, the extraction degree in dichloromethane was for about 2 times greater than that in *n*-heptane if Amberlite LA-2 is used, for about 3.3 times greater in the case of D2EHPA utilization).

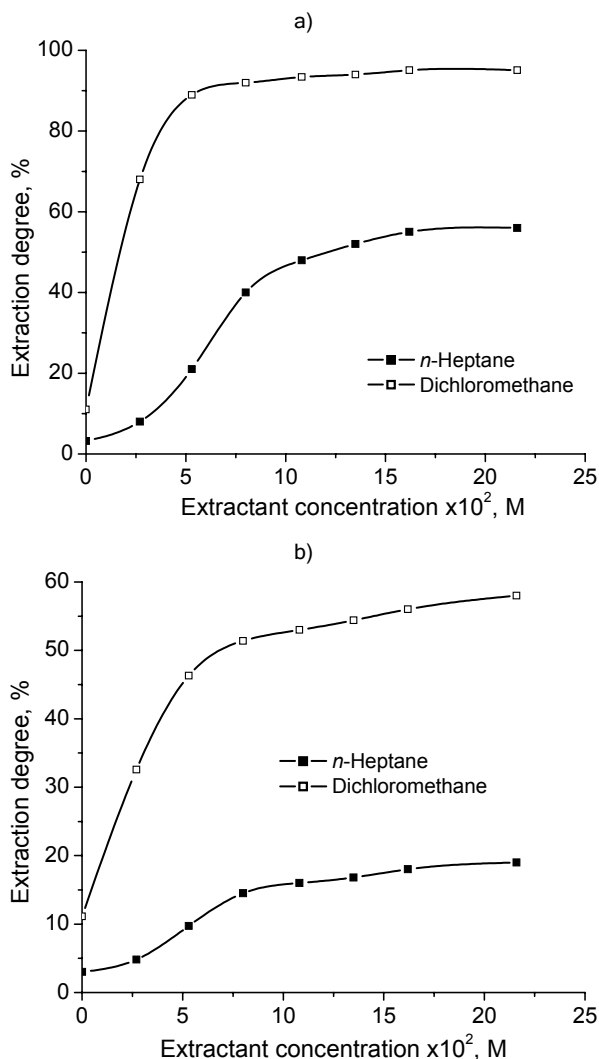


Figure 3. The influence of the concentration of Amberlite LA-2 (a) and D2EHPA (b) on the efficiency of the reactive extraction of PABA in different solvents (pH 3).

By means of the experimental data from Figure 3 and by plotting the Eqs. (6) and (14), the straight lines shown in Figure 4 have been obtained for the two extractants and three solvents considered.

The values of the slopes of straight lines depend on the extractant type and the solvent polarity, varying as follows:

- Reactive extraction with Amberlite LA-2
n-Heptane: $n = 3.19$;
Dichloromethane: $n = 1.35$.
- Reactive extraction with D2EHPA
n-Heptane: $n = 1.89$;
Dichloromethane: $n = 1.01$.

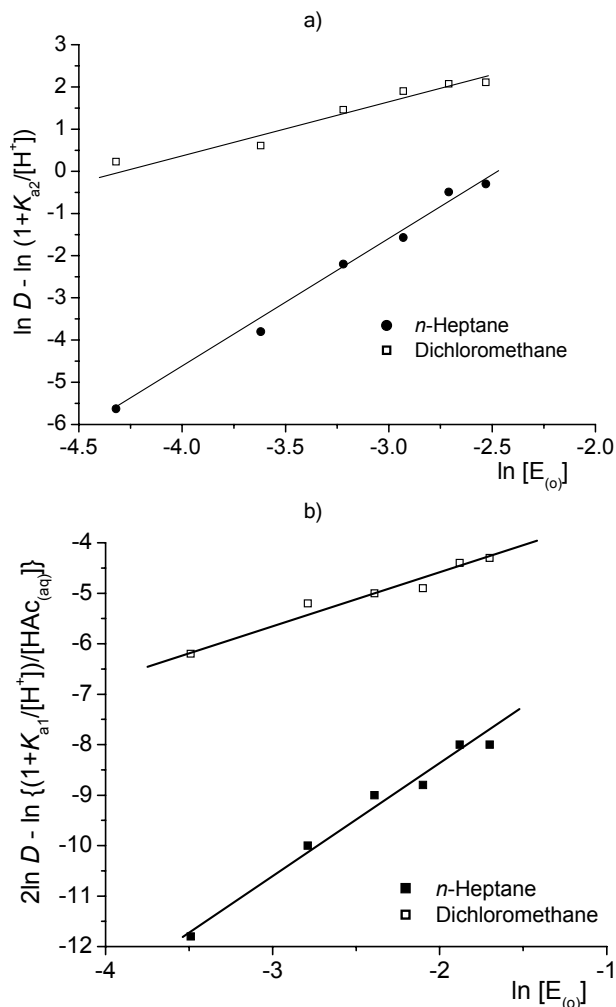


Figure 4. A graphical representation of Eq. (6) for the reactive extraction with Amberlite LA-2 (a) and Eq. (14) for the reactive extraction with D2EHPA (b).

The obtained values indicate the modification of the chemical structure of the interfacial compound in function of the used extractant-solvent combination. Therefore, the reactive extraction with Amberlite LA-2 in the low-polar solvent (*n*-heptane) occurs by means of the interfacial formation of an aminic adduct with 3 extractant molecules. If the solvent with higher polarity is used (dichloromethane), each reactant participates with one molecule to the interfacial reaction.

These results have been also suggested by the variation of the reactive extraction yield plotted in Fi-

gure 3a. It can be observed from this figure that for *n*-heptane the extraction degree strongly increases with the increase in Amberlite LA-2 concentration to 40 g/l, the limit that corresponds to a molar ratio between the solute and the extractant of about 1:3. On the other hand, the yield of the extraction in dichloromethane significantly increases with the increase in extractant concentration only for Amberlite LA-2 concentration below 15 g/l, the value that corresponds to a molar ratio of 1:1 between the acid and the extractant. Over these levels of the amine concentration, the extraction degrees increase slowly with the extractant concentration.

For the reactive extraction with D2EHPA, the slopes of the straight lines suggest either the formation of an adduct by the reaction between one molecule of PABA and two molecules of the extractant, if the extraction is performed with *n*-heptane, or the formation of the interfacial product of the salt type by the reaction between one molecule of each reactant if the solvent is dichloromethane. These conclusions have also been indicated by the influence of D2EHPA concentration on the extraction degree plotted in Figure 3b. Thus, for the extractant concentration values over 30 g/l in *n*-heptane, which corresponds to the molar ratio of 1:2 between the reactants, or over 15 g/l in dichloromethane, corresponding to the molar ratio of 1:1, the influence of this parameter is considerably reduced.

As it was presented above, the solvent polarity represents an important parameter that controls the extraction of ionizable solutes. The dielectric constant is considered a characteristic of the solvent-solute local interactions, inducing the limitation of the solute solvation by solvent or extractant, due to the presence of ionizable groups in the solute chemical structure [20]. The modification of dielectric constant has a smaller effect on the solubility and extraction of non-electrolytes or weak electrolytes, but it becomes an important factor for the extraction of dissociable solutes.

Therefore, the solvent polarity controls the extraction constant through its influence on separation efficiency and mechanism. For the reactive extraction of PABA, the values of the extraction constants graphically determined for the considered extractants and solvents are given in Table 2.

By increasing the dielectric constant from 1.90 (*n*-heptane) to 9.08 (dichloromethane), the extraction constant increases 9 times for Amberlite LA-2 and about 3.25 times for D2EHPA.

Table 2. The expressions and values of the extraction constants for the studied systems

Extractant	Solvent	Extraction constant	Value
Amberlite LA-2	<i>n</i> -Heptane	$K_E = \frac{[\text{HAc}_{(\text{aq})}E_{3(\text{o})}]}{[\text{HAc}_{(\text{aq})}][E_{(\text{o})}]^3}$	4.03×10^2 l ³ /mol ³
	Dichloromethane	$K_E = \frac{[\text{HAc}E_{(\text{o})}]}{[\text{HAc}_{(\text{aq})}][E_{(\text{o})}]}$	3.64×10^3 l/mol
D2EHPA	<i>n</i> -Heptane	$K_E = \frac{[\text{HAc}E_{(\text{o})}]^2}{[\text{H}_2\text{Ac}_{(\text{aq})}^+][E_{(\text{o})}]^2}$	2.13×10^{-2} l/mol
	Dichloromethane	$K_E = \frac{[\text{HAc}E_{(\text{o})}]^2}{[\text{H}_2\text{Ac}_{(\text{aq})}^+][E_{(\text{o})}]}$	6.92×10^{-2}

CONCLUSIONS

The studies on the reactive extraction of PABA with Amberlite LA-2 and D2EHPA in two solvents with different polarities (*n*-heptane, dichloromethane), at various extractants concentrations and pH values of aqueous solutions indicated that the reactive extraction occurs by means of the interfacial reaction of the hydrogen bonding or ionic type. The mechanism of the reaction is controlled by the extractant type and solvent polarity.

Compared with D2EHPA, the use of Amberlite LA-2 allows the possibility of reaching the higher extraction efficiency, the extraction degree being supplementary increased by increasing the solvent polarity.

By reducing the dielectric constant of the solvents, the molecular ratio between the solute and the aminic extractant participating at the interfacial compounds formation is changed from 1:1 to 1:3 and from 1:1 to 1:2 if the extractant is D2EHPA.

The highest value of the extraction constant has been obtained for the reactive extraction with Amberlite LA-2 dissolved in dichloromethane.

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