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Distribution Behavior of Amino Acid by Extraction with Di(2-ethylhexyl) Phosphoric Acid

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ABSTRACT

The distribution equilibrium of L-tryptophan (L-Trp) by extraction with di(2-ethylhexyl) phosphoric acid (D2EHPA) dissolved in *n*-hexane was studied. The effects of L-Trp and D2EHPA concentrations, pH, and ionic strength, particularly of L-Trp loading in the organic phase, on extraction equilibrium were examined in detail. When the amino acid loading ratio (the molar concentration ratio of the equilibrium amino acid in the organic phase to the initial dimeric D2EHPA) was less than 3×10^{-3}, one L-Trp molecule was extracted by forming a complex with four monomeric D2EHPA molecules, and the extraction equilibrium constant (K_e) was determined to be 0.045 dm^3/mol. Above this loading ratio the equilibrium formula did not hold, and the apparent equilibrium constant (K_a) increased significantly with increasing loading ratio. The phenomenon was explained by taking into account two parallel reactions in which fewer D2EHPA molecules, two and one respectively, were needed to extract one L-Trp molecule.

INTRODUCTION

Di(2-ethylhexyl) phosphoric acid (D2EHPA) is an important acidic organophosphorus extractant because of its high separation and extraction efficiency (1). In the last few decades, D2EHPA has been extensively applied to the separation and purification processes of uranium, rare earths, and other metals such as copper and zinc (1–4). In general, a metal ion is extracted into the organic phase by forming a complex with D2EHPA (3–5). From 1996 to 1976, Kolarik et al. (5–8) surveyed the

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extraction kinetics and equilibrium behavior of various metals with D2EHPA. By combination with the separation method involving the emulsion liquid membrane (ELM), extraction technology with D2EHPA was further developed in the 1980s (9).

Recently, with developments in biotechnology, the applications of D2EHPA in the extraction of amino acids have greatly attracted the attention of both scientists and engineers. Teramoto et al. (10) studied the extraction of tryptophan by an ELM containing D2EHPA. In their paper they focused on the application of D2EHPA as an ELM carrier, while the precise mechanism of amino acid extraction was not dealt with. More recently, Liao et al. (12) studied the extraction mechanism of isoleucine in a kerosene/water system with D2EHPA as extractant, and an extraction equilibrium constant was proposed. However, the effects of amino acid concentration, amino acid loading in the organic phase, and ionic strength on the extraction equilibrium were not considered in either publication (10, 12). Because amino acid is an amphoteric electrolyte, its extraction mechanism may be different from that of metals. Therefore, for efficient applications of D2EHPA to amino acid extraction, it is very important to determine the extraction mechanism at various conditions.

Thus in this work, using L-tryptophan as a model amino acid, we studied the effects of amino acid and D2EHPA concentrations, pH, and ionic strength, particularly of amino acid loading in the organic phase, on the extraction equilibrium. A general extraction formula is proposed. Moreover, the limit of the general extraction formula is discussed in detail.

MATERIALS AND METHODS

Materials

Di(2-ethylhexyl) phosphoric acid (D2EHPA) was a product of No. 1 Chemical Reagent Factory of Tianjin, People's Republic of China, with a purity of approximately 93%. It was further purified by recrystallization with copper hydroxide, as described by Partridge and Jensen (13). It was determined that the purified D2EHPA had a purity of 99.6%. L-Tryptophan (L-Trp) was of chromatographic grade. Other reagents, such as *n*-hexane, concentrated sulfuric acid (H_2SO_4), sodium sulfate (Na_2SO_4), and sodium hydroxide (NaOH), were all of analytical grade.

Extraction Experiments

All of the extraction experiments were carried out in 25-mL tapered flasks. Organic solutions were prepared by dissolving D2EHPA in *n*-hexane. Aqueous solutions contained 0.38 to 46 mM L-Trp. The ionic strength

of the aqueous phase was 0.2 M $(Na,H)SO_4$. That is, the total concentration of sulfate in the aqueous phase was fixed at 0.2 M except in the experiments for studying the effect of ionic strength background. Organic and aqueous phases of the same volume (5 mL) were vigorously stirred with a magnetic stirrer for 20 minutes. After partition equilibrium had been reached, phase separation was carried out by centrifugation. The experimental temperature was fixed at 298 ± 0.5 K by controlling the room temperature with an air conditioner.

Analysis

The pH of the aqueous phase was measured with a digital pH meter (Model PHS-29A, Leici Instrument Factory, Shanghai, People's Republic of China). The concentration of L-Trp in the aqueous phase was determined using a UV spectrophotometer (Model 752C, No. 3 Analytical Instrument Factory of Shanghai, Shanghai, People's Republic of China) at a wavelength of 264 nm.

RESULTS AND DISCUSSION

Equilibrium Constant of L-Trp Extraction with D2EHPA and the Limit of the Extraction Equilibrium Formulation

Equilibrium Constant of L-Trp Extraction with D2EHPA

Amino acid is a zwitterion with a carboxylic group and an amino group. Two dissociation equilibria exist in aqueous solutions:

$$H_2A^+ \overset{K_1}{\Leftrightarrow} H^+ + HA \tag{1}$$

$$HA \overset{K_2}{\Leftrightarrow} H^+ + A^- \tag{2}$$

where H_2A^+, HA, and A^- are the cationic, neutral, and anionic amino acid, respectively. The dissociation constants for Eqs. (1) and (2) can be described as follows:

$$K_1 = \frac{[H^+][HA]}{[H_2A^+]} \tag{3}$$

$$K_2 = \frac{[H^+][A^-]}{[HA]} \tag{4}$$

In aqueous solutions, pK_1 and pK_2 of L-Trp are 2.38 and 9.38, respectively (14).

In amino acid extraction with D2EHPA, D2EHPA at the interface of the aqueous and organic phases reacts with the cationic amino acid, H_2A^+, forming a complex and releasing H^+. Then the complex diffuses into the organic phase. Due to the polymerization tendency of the complex with D2EHPA in the organic phase (8), it is considered that the cationic amino acid, H_2A^+, is extracted as an m-polymerized complex into the organic phase, and the extraction equilibrium can be described by the following generalized form:

$$m\mathrm{H_2A^+} + m\left(n + \frac{1}{2}\right)\overline{(\mathrm{HR})} \overset{K_e}{\Leftrightarrow} \overline{(\mathrm{H_2AR}\cdot n(\mathrm{HR})_2)_m} + m\mathrm{H^+} \qquad (5)$$

where a component under a bar indicates the organic phase. The extraction equilibrium constant for the equation is expressed as

$$K_e = \frac{[\overline{(\mathrm{H_2AR}\cdot n(\mathrm{HR})_2)_m}][\mathrm{H^+}]^m}{[\mathrm{H_2A^+}]^m[\overline{(\mathrm{HR})_2}]^{m(m+1/2)}} \qquad (6)$$

where $[i]$ represents the molar concentration of component i.

The distribution coefficient of the amino acid between the organic and the aqueous phases is defined as

$$D = \frac{[\overline{\mathrm{H_2A^+}}]}{[\mathrm{H_2A^+}] + [\mathrm{HA}] + [\mathrm{A^-}]} = \frac{m[\overline{(\mathrm{H_2AR}\cdot n(\mathrm{HR})_2)_m}]}{[\mathrm{H_2A^+}] + [\mathrm{HA}] + [\mathrm{A^-}]} \qquad (7)$$

In this work the equilibrium pH of the aqueous phase is less than 4.0. Therefore, the concentration of the anionic amino acid, A^-, is negligible. Equation (7) can be simplified to

$$D = \frac{m[\overline{(\mathrm{H_2AR}\cdot n(\mathrm{HR})_2)_m}]}{[\mathrm{H_2A^+}] + [\mathrm{HA}]} \qquad (8)$$

Combining Eqs. (6) and (8) gives

$$D = \frac{mK_e[\mathrm{H_2A^+}]^n\,[\overline{(\mathrm{HR})_2}]^{m(n+1/2)}[\mathrm{H^+}]^{-m}}{[\mathrm{H_2A^+}] + [\mathrm{HA}]} \qquad (9)$$

Moreover, the total mass balance of amino acid in the extraction system can be expressed by

$$[\mathrm{HA}]_T = [\mathrm{H_2A^+}] + [\mathrm{HA}] + [\overline{\mathrm{H_2A^+}}] \qquad (10)$$

Substitution of Eq. (10) into Eq. (9) leads to an expression of the distribution coefficient D. That is,

$$\log D + \frac{1 - m}{m} \log \frac{D}{1 + D} = \log K_e' + \left(n + \frac{1}{2}\right) \log[\overline{HR}] + \frac{m - 1}{m} \log[HA]_T - \log([H^+] + K_1) \quad (11)$$

where K_e' is a constant,

$$K_e' = \left(\frac{mK_e}{2^{m(n+1/2)}}\right)^{1/m} \quad (12)$$

By partially differentiating Eq. (11), we can obtain the following equations:

$$\left(\frac{\partial \log D}{\partial \log[HA]_T}\right)_{[H^+][\overline{HR}]} = \frac{1 + D}{1 + mD}(m - 1) \quad (13)$$

$$\left(\frac{\partial \log D}{\partial \log[\overline{HR}]}\right)_{[H^+][HA]_T} = \frac{m(1 + D)}{1 + mD}\left(n + \frac{1}{2}\right) \quad (14)$$

$$\left(\frac{\partial \log D}{\partial pH}\right)_{[\overline{HR}][HA]_T} = \frac{[H^+]}{[H^+] + K_1} \frac{1 + D}{1 + mD} m \quad (15)$$

Furthermore, the following equation is derived by taking the ratio of the left- and right-hand sides of Eqs. (13) and (15):

$$\left(\frac{\partial pH}{\partial \log [HA]_T}\right)_{[\overline{HR}]} = \left(\frac{[H^+] + K_1}{[H^+]}\right)\frac{m - 1}{m} \quad (16)$$

Then 3the value of m can be determined by plotting the experimental data of $[HA]_T$ vs pH. As shown in Fig. 1, the slope of the $[HA]_T$ vs pH plot is nearly zero. From Eq. (16) it can be calculated that $m = 1$. Thus Eq. (5) can be simplified to the following form:

$$H_2A^+ + \left(n + \frac{1}{2}\right)\overline{(HR)_2} \overset{K_e}{\Leftrightarrow} \overline{HAR \cdot n(HR)_2} + H^+ \quad (17)$$

Substituting $m = 1$ into Eq. (9) gives

$$D = \frac{K_e[H_2A^+][\overline{(HR)_2}]^{(n+1/2)}[H^+]^{-1}}{[H_2A^+] + [HA]} \quad (18)$$

By rewriting Eq. (18) and taking the logarithm of both sides of the rewritten

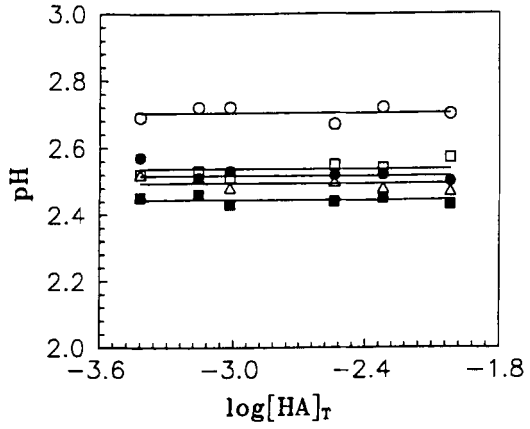

FIG. 1 Effect of total L-Trp concentration on pH at different D2EHPA concentrations. D2EHPA concentrations (mol/dm^3): 0.4844 (○), 0.605 (□), 0.756 (△), 1.058 (●), 1.361 (■).

equation, the following equation is obtained:

$$\log \{K_1 + [H^+]D\} = \log K_e + \left(n + \frac{1}{2}\right) \log[\overline{(HR)_2}] \qquad (19)$$

The relationship of $\log\{(K_1 + [H^+])D\}$ and $\log[(HR)_2]$ on the Cartesian coordinate is shown in Fig. 2. Linearly least-square fittings give the slopes, i.e., the values of n in Eq. (19). When the total amino acid concentrations are less than 3.0 mM, the slope of the plots is 2.03, nearly equal to 2.0. Thus, the value of n is 1.5. The result suggests that at lower amino acid concentrations the stoichiometry of the extraction can be expressed as

$$H_2A^+ + 2\overline{(HR)_2} \overset{K_e}{\Leftrightarrow} \overline{H_2AR \cdot 1.5(HR)_2} + H^+ \qquad (20)$$

Equation (20) is the same as that proposed by Teramoto et al. (10) and Liao et al. (12) where n-dodecane (10) and kerosene (12) were employed as the diluent, respectively. Thus, as in metal extraction, one molecule of the cationic amino acid is carried into the organic phase by four monomeric D2EHPA molecules. From the molecule structure of amino acid, we propose the structure of the complex as illustrated in Fig. 3. In this figure the four D2EHPA molecules are numbered from I to IV. The first D2EHPA molecule substitutes the hydrogen ion on the $NH^+{}_3$ group of the amino acid by ion exchange. The other three D2EHPA molecules form hydrogen bonds with the cationic amino acid. Moreover, the third and

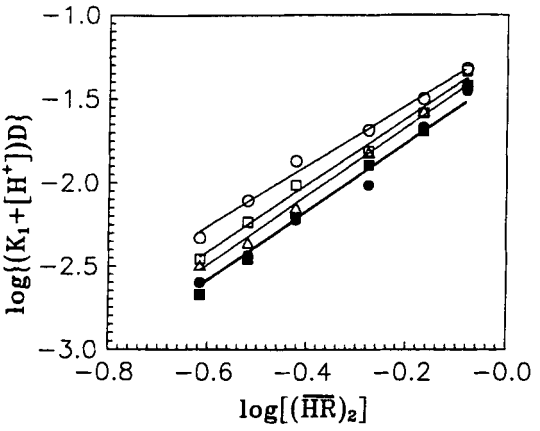

FIG. 2 The relationship between $\log[(\overline{HR})_2]$ and $\log\{(K_1 + [H^+])D\}$. Total L-Trp concentrations (mmol/dm^3): 9.59 (○), 4.80 (□), 2.90 (△), 0.97 (●), 0.70 (■).

fourth form a hydrogen bond to each other in the organic phase. The range of amino acid concentrations where Eq. (20) holds will be further discussed in the next subsection.

From the intersections of the straight lines on the coordinate in Fig. 2, the values of K_e are estimated. The results, together with the values of n, are shown in Table 1. Clearly, the equilibrium constant and n at different

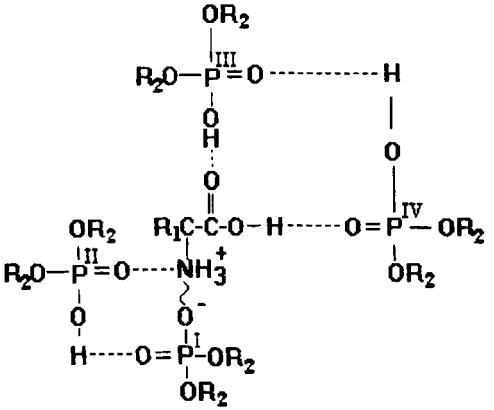

FIG. 3 Supposed structure of L-Trp-D2EHPA complex in the organic phase: (~) ionic bond, (- - -) hydrogen bond.

TABLE 1
The Values of n and K_c Obtained by Combining Eq. (19) and Fig. 2

$[HA]_T$ (mmol/dm^3)	0.70	0.97	2.90	4.79	9.59
$(n + 1/2)$	2.05	2.05	2.00	1.99	1.80
n	1.55	1.55	1.50	1.49	1.30
$K_c \times 10^2$ (dm^3/mol)	4.53	4.45	5.77	6.07	6.65

amino acid concentrations do not remain constant. This will be further discussed in the following subsection. From Table 1 and Fig. 2, the value of K_e approaches a constant, i.e., 0.045 dm^3/mol, only when the amino acid concentration is less than 1.0 mmol/dm^3.

Limit of the Extraction Equilibrium Formula

It has been shown in the above subsection that n and K_e are not constant at high total amino acid concentrations. Thus, we compared the relationship between the loading ratio of the amino acid in the organic phase and the apparent equilibrium constant. The apparent extraction equilibrium constant K_a is defined as

$$K_a = \frac{[\overline{H_2A_+}][H^+]}{[H_2A^+][\overline{(HR)_2}]^2} \tag{21}$$

while the loading ratio L is defined as the ratio of the equilibrium amino acid concentration in the organic phase to the total dimeric D2EHPA concentration, i.e.,

$$L = \frac{[\overline{H_2A^+}]}{[\overline{(HR)_2}]_T} \tag{22}$$

As indicated in Fig. 4, the apparent extraction equilibrium constant K_a remains constant when the loading ratio is less than about 3×10^{-3}. Then it increases with an increase of the loading ratio.

From the result that n declines from 1.5 with increasing amino acid concentration (Table 1), it is known that the complex $H_2AR \cdot 1.5(HR)_2$ exists in the organic phase only in the range of lower amino acid loadings. The phenomenon may be explained as follows: The hydrogen bonds in the complex (Fig. 3) are much less stable than the ionic bond. With the increase of amino acid concentration in the organic phase, the polarity of the organic phase increases and the hydrogen bonds become weaker. This results in a decrease of the polymerization tendency of D2EHPA mole-

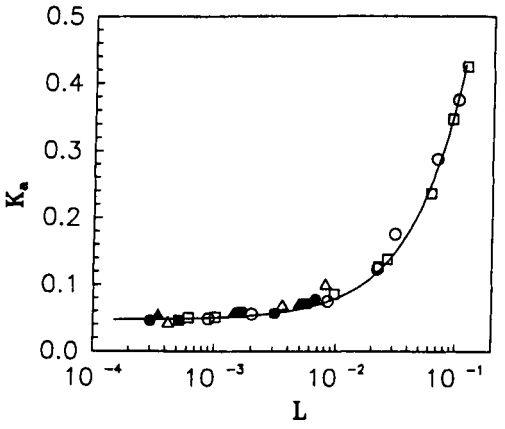

FIG. 4 Determination of the limit of extraction equilibrium formulation. D2EHPA concentrations (mol/dm^3): 0.4844 (○), 0.605 (□), 0.756 (△), 1.058 (●), 1.361 (■), 1.663(▲).

cules by hydrogen bonds (15). Thus the hydrogen bonds tend to break up and other complex forms with less D2EHPA molecules, such as $H_2AR{\cdot}HR$ and H_2AR, will appear in the organic phase at higher amino acid loadings. It has been demonstrated that the independent existence of $H_2AR{\cdot}1.5(HR)_2$ and $H_2AR{\cdot}HR$ or $H_2AR{\cdot}1.5(HR)_2$ and H_2AR was not able to explain the experimental results; we assume that the both $H_2AR{\cdot}HR$ and H_2AR appear in the range of higher loadings. Hence we consider that three parallel extraction equilibria exist at the same time, i.e., Eq. (20) and the following two equations:

$$H_2A^+ + \overline{H_2AR{\cdot}1.5(HR)_2} \overset{K_{e,1}}{\Leftrightarrow} 2\overline{H_2AR{\cdot}HR} + H^+ \tag{20a}$$

and

$$H_2A^+ + \overline{H_2AR{\cdot}HR} \overset{K_{e,2}}{\Leftrightarrow} 2\overline{H_2AR} + H^+ \tag{20b}$$

The equilibrium constants for Eqs. (20), (20a), and (20b) are expressed as

$$K_e = \frac{[\overline{H_2AR{\cdot}1.5(HR)_2}][H^+]}{[H_2A^+][\overline{(HR)_2}]^2} \tag{23}$$

$$K_{e,1} = \frac{[\overline{H_2AR{\cdot}HR}]^2]H^+]}{[H_2A^+][\overline{H_2AR{\cdot}1.5(HR)_2}]} \tag{24}$$

and

$$K_{e,2} = \frac{[\overline{H_2AR}]^2[H^+]}{[H_2A^+][\overline{H_2AR \cdot HR}]} \tag{25}$$

respectively. Because

$$[\overline{H_2A^+}] = [\overline{H_2AR \cdot 1.5(HR)_2}] + [\overline{H_2AR \cdot HR}] + [\overline{H_2AR}]$$

taking Eqs. (23), (24), and (25) into Eq. (21) gives

$$K_a = K_e + \frac{(K_eK_{e,1})^{1/2}}{[\overline{(HR)_2}]} + \frac{(K_eK_{e,1})^{1/4}K_{e,2}^{1/2}}{[\overline{(HR)_2}]^{3/2}} \tag{26}$$

Because the concentrations of the three kinds of H_2A–D2EHPA complex cannot be separately measured, the values of $K_{e,1}$ and $K_{e,2}$ could not be determined from the experimental data. However, it is considered that Eq. (26) may qualitatively describe Fig. 4 because the increase in loading ratio corresponds to the decrease in the equilibrium extractant concentration $[\overline{(HR)_2}]$, which leads to the increase of K_a in Eq. (26).

Very low amino acid loading corresponds to very high equilibrium extractant concentration. Thus, in the range of very low amino acid loadings, the second and third terms on the right-hand side of Eq. (26) are negligible and K_a remains constant ($= K_e$). Hence the applicability of Eq. (20) is limited by the loading ratio. As shown in Fig. 4, the apparent equilibrium constant remains nearly constant up to an L-Trp loading ratio of 3×10^{-3}. This means that the extraction equilibrium formula, Eq. (20), is only applicable to the range of monomeric D2EHPA conversion less than 0.6%. In fact, the loading ratio of 3×10^{-3} was less than 3% of the whole loading ratio range (>0.1, Fig. 4). Hence the parallel reactions, Eqs. (20a) and (20b), occurred at the very low equilibrium aqueous phase concentrations.

Increases of the extraction equilibrium constant at higher loading ratios were also reported in zinc extraction with D2EHPA (11). However, K_e remained constant when the loading ratio was less than 1×10^{-1}, corresponding to a monomeric D2EHPA conversion of 25%. Hence the applicable range of Eq. (20) for amino acid extraction is much smaller than that of metal extraction. Two reasons may be considered.

1. The hydrogen bonds of the amino acid–D2EHPA complex are much weaker than the coordinate bonds of zinc–D2EHPA.
2. For amino acid extraction, dissociation equilibria exist in the aqueous phase beside the extraction equilibrium.

In Fig. 4 the value of the apparent extraction equilibrium constant, K_a, is 0.0469 (dm^3/mol) when the loading ratio approaches to zero. The value

is nearly the same as that shown in Table 1 at total amino acid concentrations less than 1.0 mmol/dm^3.

The effect of the loading ratio on the distribution coefficient at lower loading ratios is shown in Fig. 5. Although the extraction equilibrium constant remains constant at the loading range (Fig. 4), the distribution coefficient ascends with an increasing loading ratio. This can be explained by using Eq. (18). Because of the increase of $[H_2A^+]/([H_2A^+] + [HA])$ with increasing amino acid concentration $[HA]_T$ at constant D2EHPA and pH (Fig. 1), D will increase with the loading ratio.

Discussion

Teramoto et al. (10) reported the extraction equilibrium of L-Trp by D2EHPA in a similar experimental conditions. In their work the loading ratio was about 3.6×10^{-3}. The value of K_e was 0.0554 (dm^3/mol). At the same loading ratio, the extraction equilibrium constant in Fig. 4 is 0.0586 (dm^3/mol), only slightly larger than that reported by Teramoto et al.

Using $n = 1.5$ and $m = 1$ in Eqs. (13), (14), and (15), these equations are rewritten to the following equations:

$$\left(\frac{\partial \log D}{\partial \log[HA]_T}\right)_{[H^+][\overline{HR}]} = 0 \tag{27}$$

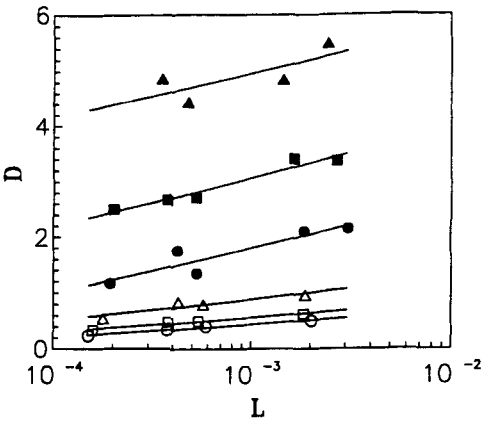

FIG. 5 Effect of amino acid loading on distribution coefficient. D2EHPA concentrations (mol/dm^3): 0.4844 (○), 0.605 (□), 0.756 (△), 1.058 (●), 1.361 (■), 1.663(▲).

$$\left(\frac{\partial \log D}{\partial \log[\overline{HR}]}\right)_{[H^+]} = 2 \tag{28}$$

$$\left(\frac{\partial \log D}{\partial pH}\right)_{[\overline{HR}]} = \frac{[H^+]}{[H^+] + K_1} \tag{29}$$

In Eq. (27) the distribution coefficient is not related to the total amino acid concentration. Then, based on Eqs. (27) and (28), plots of log D vs log[HR] at different hydrogen ion concentrations are obtained, as shown in Fig. 6. The results agree to those shown in Fig. 5. Namely, the distribution coefficient increases with increasing total amino acid concentration. In addition, the distribution coefficient increases with an increase of the extractant concentration in the organic phase. The slope of the straight lines at $[HA]_T$ less than 1.0 mM is 1.94 in Fig. 6, nearly the same as the theoretical value (2) expressed by Eq. (28). This also agrees with the result in Table 1, i.e., $n = 1.5$. At higher total amino acid concentrations, however, the slopes of the straight lines decline. The result further supports the conclusion that only at very low amino acid concentrations or amino acid loadings does L-Trp exists in the organic phase in the form of $H_2AR \cdot 1.5(HR)_2$, while at higher amino acid loadings it exists in other forms such as $H_2AR \cdot HR$ and H_2AR.

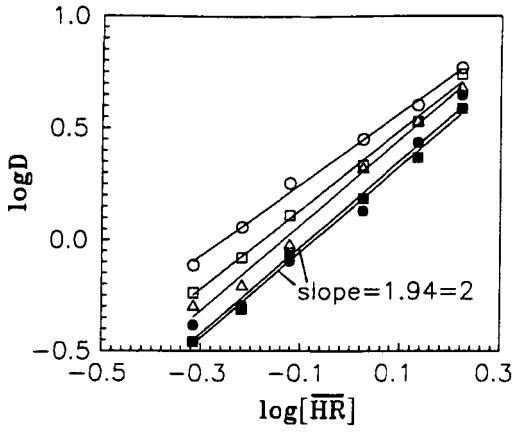

FIG. 6 The relationship of log D and log$[HR]_T$. Total L-Trp concentrations (mmol/dm^3): 9.59 (○), 4.80 (□), 2.90 (△), 0.97 (●), 0.70 (■).

Effect of D2EHPA Concentration on Distribution Coefficient

At a constant total L-Trp concentration, the equilibrium L-Trp concentration in the aqueous phase decreases with an increase of the D2EHPA concentration, as shown in Fig. 7. This is ascribed to the enhancement of the extraction capacity of the organic phase. Therefore, the increase of the extractant concentration increases the distribution coefficient D (Fig. 7b). It can be found that in addition to extractant concentration, the

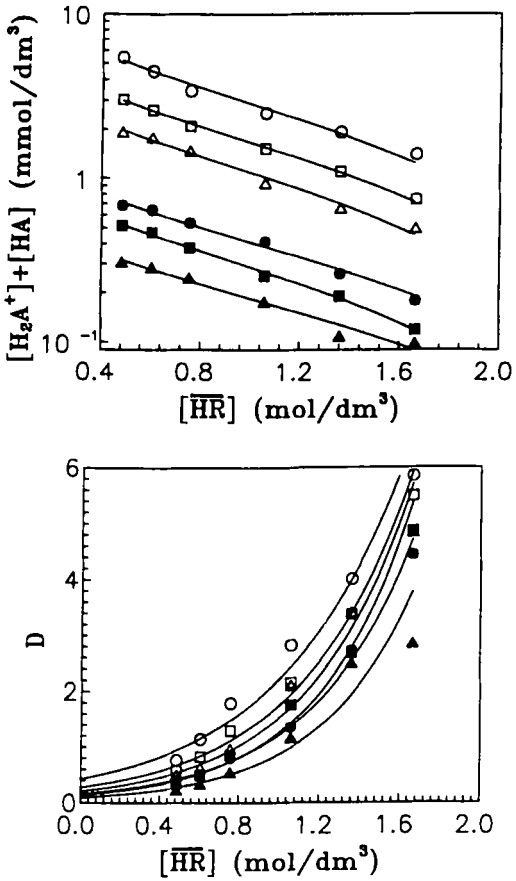

FIG. 7 Effect of D2EHPA concentration on aqueous L-Trp concentration (a) and distribution coefficient (b). Total L-Trp concentrations (mmol/dm^3): 9.59 (○), 4.80 (□), 2.90 (△), 0.97 (●), 0.70 (■), 0.38 (▲).

distribution coefficient is also affected by the total amino acid concentration. This is attributed to the increase of the apparent equilibrium constant by increasing the amino acid loading in the organic phase as stated in the above section (Figs. 2 and 6). At the range of high amino acid concentrations, Eq. (27) does not hold because the equation was derived based upon Eq. (5), i.e., Eq. (20).

Effect of pH on Distribution Coefficient

To adjust the pH value at a constant sulfate concentration (0.2 mol/dm^3), sulfuric acid and sodium sulfate were introduced to the aqueous phase at different ratios. The dependence of the distribution coefficient on H^+ concentration is shown in Fig. 8. Clearly, the distribution coefficient increases with a decrease of H^+ concentration or an increase of pH. It is obvious from the extraction equilibrium equation, Eq. (20), that higher pH is beneficial to amino acid extraction.

According to the results for $n = 1.5$ and $m = 1$, the following equation is derived from Eq. (11):

$$\log D = \log K_e' + 2 \log[\overline{HR}] - \log([H^+] + K_1) \qquad (30)$$

This shows that the slope of the log D vs log($[H^+] + K_1$) plot for Eq. (30) is negative unity. The straight line in Fig. 8 is just the expression of Eq. (30). The experiment data present good agreement to the equilibrium theory in the pH range of 0.8 to 3.29. It should be noticed that the form

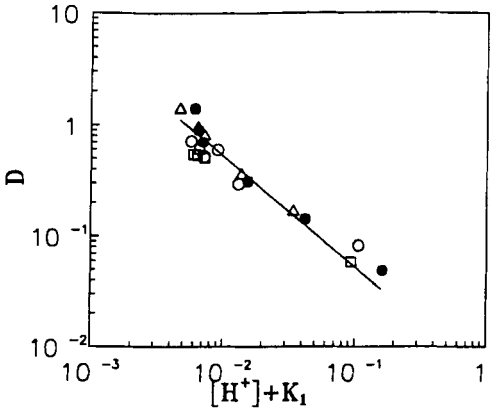

FIG. 8 Effect of hydrogen ion concentration (pH) on distribution coefficient. Total L-Trp concentrations (mmol/dm^3): 0.79 (○), 1.06 (□), 3.13 (△), 5.01 (●).

of Eq. (30) for amino acid is different from that for metal ion extraction with D2EHPA (11), in which log D was linearly proportional to log[H^+]. This is because amino acid is a zwitterion and represents dissociation equilibria in aqueous solutions.

Effect of Ionic Strength on Distribution Coefficient

In the experiments mentioned above, sodium sulfate and sulfuric acid were added into the aqueous solution to adjust the ionic strength of the system. The ionic strength was fixed at 0.2 M $(Na,H)SO_4$. Because the extraction equilibrium constant of sodium ion with D2EHPA in kerosene is 0.00048 dm^3/mol when the saponification ratio is less than 25% (16), the effect of sodium ion distribution on amino acid extraction equilibrium can be neglected. Then we introduced more sodium sulfate to the aqueous phase to study the effect of ionic strength on amino acid extraction. Figure 9 illustrates that D is increased by increasing the ionic strength. Because the solubility of L-Trp is far less than that of sodium sulfate in the aqueous phase, the addition of the salt results in a decrease of the free water concentration in the aqueous phase, Thus, it becomes difficult for L-Trp to be solubilized in the aqueous phase, and L-Trp is more easily distributed to the organic phase at higher ionic strength.

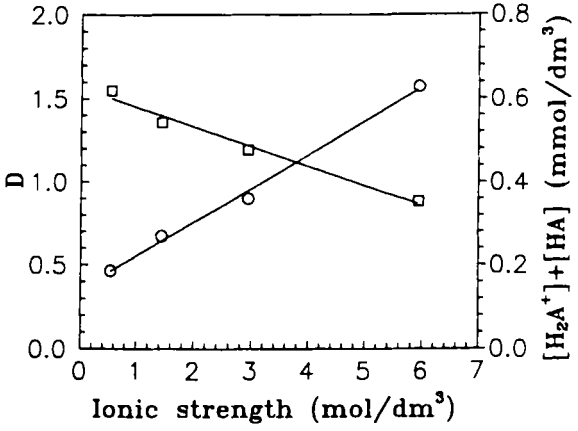

FIG. 9 Effect of ionic strength on distribution coefficient (○) and equilibrium L-Trp concentration in the aqueous phase (□). D2EHPA concentration: 0.4844 mol/dm^3. Total L-Trp concentration: 0.91 mmol/dm^3.

CONCLUSIONS

The extraction equilibrium constant remains constant only when the amino acid loading ratio is less than 3×10^{-3}. This value is much smaller than that for zinc extraction, indicating that the bonding of amino acid with D2EHPA is much weaker than that of zinc. At this range of loading ratio, the extraction equilibrium can be expressed by Eq. (20). The extraction equilibrium constant of Eq. (20) is 0.045 (dm^3/mol). Above this loading ratio, Eq. (20) does not hold, i.e., the complex $H_2AR \cdot 1.5(HR)_2$ tends to be broken up and the other complex forms with less D2EHPA molecules will appear. This causes an increase of the apparent equilibrium constant and the distribution coefficient of amino acid. Therefore, the distribution coefficient increases with an increase of the total amino acid concentration. In addition, the distribution coefficient increases with increasing D2EHPA concentration, ionic strength, as well as aqueous phase pH. Thus, in a practical extraction process, the parameters can be used to selectively isolate a desired amino acid.

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NOMENCLATURE

A^-	anion of L-Trp
D	distribution coefficient defined by Eq. (7)
H^+	hydrogen ion
HA	zwitterion of L-Trp
H_2A^+	cation of L-Trp
HR	D2EHPA monomer
$(HR)_2$	D2EHPA dimer
K_a	apparent extraction equilibrium constant defined by Eq. (21)
K_1, K_2	dissociation constant (mol/m^3)
K_e	extraction equilibrium constant
$K_{e,1}$, $K_{e,2}$	extraction equilibrium constant defined by Eqs. (24) and (25)
K_z'	constant defined by Eq. (12)
L	loading ratio defined by Eq. (22)

m	aggregation degree of the extracted species
n	number of free dimeric D2EHPA involved in the extracted species
[ ]	concentration of the species in the brackets

Superscript

$\overline{\text{(overbar)}}$	denoting the organic phase species or organic phase concentration

Subscript

T	total concentration of species

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