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Liquid Membrane Transport of Supramolecular Complexes of Some Amines and Amino Acids with Macrocyclic Ligands

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Abstract. The transport of some amines in protonated form was studied (viz. methylamine, dimethylamine, diethylamine and *n*-propylamine) and α-amino acids (L-leucine, L-methionine, L-isoleucine, L-phenylalanine, L-valine, L-α-alanine and L-cysteine). The following macrocyclic ligands were used as carriers throughout the experiments: 15-crown-5 (15C5), 18-crown-6 (18C6), benzo-18-crown-6 (B18C6), dibenzo-18-crown-6 (DB18C6), diazacrown ether [2.2] (1,7,10,16-tetraoxa-4,13-diazacyclooctadecane) and cryptand [2.2.2] (4,7,13,16,21,24-hexaoxa-1,10-diazabicyclo [8.8.8] hexacosane). The active transport, assisted by pH gradient, of amino acids and amines in protonated form as ion pairs in the presence of picrate anion was performed. The experiments suggested the influence of the ligand size, the donor atom type, and the substituents on the transport phenomena.

Key words: Amines, amino acids, liquid membrane, macrocyclic ligands, active transport.

1. Introduction

Macrocyclic ligands like crown ethers and cryptands can be used for the selective transport of cations through bulk and supported liquid membranes [1–6]. The ability of macrocyclic ligands to interact with biologically interesting ammonium ions via hydrogen bonds has been reported in several papers [7–13]. An understanding of the selective recognition of amines and amino acids is of both theoretical interest, as a fundamental process, and also practically, in view of the mimicking of natural biological systems. The possibility of macrobicyclic ligands forming cryptate inclusion complexes, depending on their structure, has suggested the idea of complexing protonated amines by cryptands and of extracting them in organic solvents by coupling with an inorganic anion.

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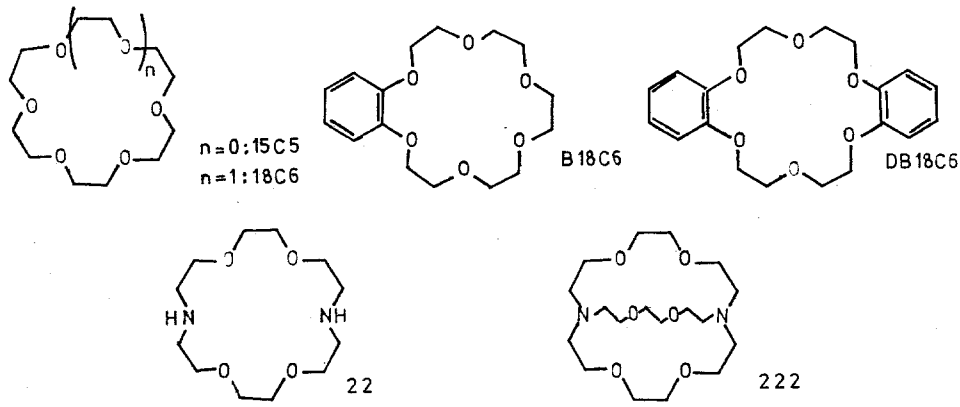

Figure 1. Chemical structures of the macrocyclic and the macrobicyclic ligands used in experiments: 15C5 (15-crown-5); 18C6 (18-crown-6); B18C6 (benzo-18-crown-6); DB18C6 (dibenzo-18-crown-6); Kryptofix [2.2] (1,7,10,16-tetraoxa-4,13-diazacyclooctadecane); Kryptofix [2,2,2] (4,7,13,16,21,24-hexaoxa-1,10-diazabicyclo[8.8.8] hexacosane).

The present study reports the transport of some amines, as well as of some amino acids, through 1,2-dichloroethane membranes containing carrier macrocyclic ligands, such as 15C5, B18C6, DB18C6, [2.2] and [2.2.2]. The active transport of amines and amino acids in protonated form as ion pairs in the presence of picrate anion, is assisted by a pH gradient. The experiments suggested the influence of the ligand size, donor atom type, and the substituents on the transport mechanism. The experimental data on transport phenomena of complexation of several protonated amines and amino acids by macrocyclic ligands were compared with the extraction constants ($\log K_{ex}$).

The investigations carried out with protonated amino acids and amines may also assist in setting up the optimum experimental conditions for some separation techniques involving liquid membranes.

2. Experimental

Reagent grade 15C5, 18C6, B18C6, DB18C6, Kryptofix [2.2] and Kryptofix [2.2.2] were obtained from Merck and used throughout the experiments without further purification (Figure 1). The following amines and amino acids were used: methylamine (Fluka), dimethylamine (Fluka), diethylamine (Fluka), *n*-propylamine (Fluka), L-α-alanine (L-α-Ala, Fluka), L-valine (L-Val, Aldrich), L-phenylalanine (L-Phe, Sigma), L-leucine (L-Leu, Fluka), L-methionine (L-Met, Sigma), and L-isoleucine (L-Ile Aldrich), which were of the highest commercially available purity. pH was determined by a digital MV-870 Pracitronic pH-meter with glass electrode and saturated calomel electrode.

Liquid membrane transport experiments were performed using the equipment reported earlier [5]. The device consists of two concentric tubes: the inner one

contains the source phase (5 mL) and also acts as a stirrer. The receiving phase (5 mL) and the membrane phase (30 mL) are introduced in the outer tube. The phases were stirred at 180 r.p.m. for 6 h, an interval which we had previously determined to be long enough for the system to reach equilibrium [14]. Each experiment was repeated three times. Reproducibility was ± 10%. The concentrations of amino acids in the receiving phase after the transport through liquid membrane were determined chromatographically using a Carlo Erba 3A 28M amino acid analyzer. In the source phase, the amino acid solutions were 0.16 mM of the individual amino acid (the same for the amines) and 1.6 mM solution of the picric acid. The pH=2.02 was ensured by use of HCl. All membrane carrier solutions were 0.01 M. The solvent 1,2-dichloroethane was distilled before use. The pH=13.01 of the receiving phase was ensured by using 0.1 N LiOH.

The quantitative solvent extraction and the determination of the extraction equilibrium constants ($\log K_{ex}$) were carried out in the 1,2-dichloroethane–water two phase system according to the procedures previously reported [15]. Equal volumes of 1.5×10^{-3}–6.0×10^{-4} M of amino acids and 6.0×10^{-5} M of picric acid (pH = 2.02) in the aqueous phases were extracted with 2.5×10^{-3}–2.0×10^{-2} M crown ethers, with Kryptofix [2.2] and Kryptofix [2.2.2] in the 1,2-dichloroethane phase. The concentrations used for the amines were in the range of 4.0×10^{-4}–2.0×10^{-2} M. The $RNH_3L^+A^-$ ion-pair complex was extracted into 1,2-dichloroethane. The volume ratio of aqueous and organic phases, respectively, in the extraction process was 1:1. Distilled 1,2-dichloroethane and water were saturated with each other prior to use in order to prevent volume change during extraction. The concentrations of picrate extracted into the organic phase were determined spectrophotometrically. No picrate was extracted into the organic phase in the absence of crown ethers. All experiments were performed at 25 ± 1 °C. A Perkin Elmer UV-Visible Spectrometer, Model 559 was used.

The overall extraction equilibrium may be written as:

$$(R\text{—}NH_3^+)_w + (A^-)_w + (L)_s = (R\text{—}NH_3LA)_s \tag{1}$$

and it is characterized by the overall extraction constant:

$$K_{ex} = \frac{[R\text{—}NH_3LA]_s}{[R\text{—}NH_3^+]_w[A^-]_w[L]_s} \tag{2}$$

where subscripts w and s define aqueous and organic phases respectively. The partition ratio (D), of the amino acid between phases w and s, respectively, can be defined by the equation:

$$D = \frac{[R\text{—}NH_3LA]_s}{[R\text{—}NH_2]_w + [R\text{—}NH_3^+]_w} \tag{3}$$

In the case of $[R\text{—}NH_3^+]_w \gg [R\text{—}NH_2]_w$, Equation (2) may be written as:

$$K_{ex} = \frac{D}{[A^-]_w[L]_s} \tag{4}$$

Table I. Experimental data of the transport study of some protonated α-amino acids through 1,2-dichloroethane liquid membranes by macrocyclic ligands and the values of log K_{ex} of their complexes.

Amino acids	18-crown-6		Benzo-18-crown-6		Dibenzo-18-crown-6	
	* (%)	log K_{ex}	* (%)	log K_{ex}	* (%)	log K_{ex}
L-Methionine	83	4.70 ± 0.04	41	4.25 ± 0.03	37	3.95 ± 0.02
L-Isoleucine	81	4.90 ± 0.04	47	4.52 ± 0.05	38	4.10 ± 0.04
L-Phenylalanine	81	5.16 ± 0.02	55	4.45 ± 0.09	45	4.25 ± 0.05
L-Leucine	81	5.76 ± 0.01	43	4.30 ± 0.05	42	4.15 ± 0.05
L-Valine	79	4.36 ± 0.03	67	4.38 ± 0.20	35	3.50 ± 0.03
L-α-Alanine	74	4.32 ± 0.02	38	4.15 ± 0.12	30	3.15 ± 0.02
L-Cysteine	50	4.11 ± 0.03	30	3.85 ± 0.05	25	2.50 ± 0.07

Source phase: [amino acid] = 0.16 mM; [picric acid] = 1.6 mM; HCl 0.05 N (pH = 2.02); 5 mL.
Membrane: 1,2-dichloroethane; [macrocyclic ligand] = 10 mM, 30 mL.
Receiving phase: LiOH 0.1 N (pH = 13.01); 5 mL.
Temperature: 25 ± 1 °C.
* (%): Amino acid percentage found in the receiving phase after 6 h stirring.

The distribution ratio is represented by:

$$D = K_{ex}[A^-]_w[L]_s \tag{5}$$

Plots of $\log(D/[A^-]_w)$ as a function of $\log[L]_s$ should give straight lines with a slope of 1 in every case. This indicates that the ligand forms a 1 : 1 complex with the cation [14].

Quantitative solvent extraction studies with amino acid and amine picrates were carried out at a variety of ligand concentrations to determine the extraction equilibrium constants K_{ex}. Each log K_{ex} is the average of 6–8 measurements.

3. Results and Discussion

The experimental data of the transport of some protonated amino acids through 1,2-dichloroethane liquid membrane containing 18C6, B18C6, and DB18C6 as carriers, in comparison with log K_{ex} of the amino acids complexes with the above mentioned macrocyclic ligands, are presented in Table I.

The transport sequence 18C6 > B18C6 > DB18C6 of the complexes with amino acids (Table I) was found to be different with respect to the extraction equilibrium.

The results indicated higher values for the amino acid transport through 1,2-dichloroethane membrane containing the ligand 18C6 as carrier; the values ranged from 83% (L-Met) to 50% (L-Cys). Similarly, the results showed higher values for the extraction constants in the case of 18C6, which ranged from 5.76 (L-Met) to 4.11 (L-Cys). This is mainly due to the relative solubility of 18C6 in water, and the fit of its cavity with the cation NH_3^+.

Table II. Percentage of some amino acids transported in the receiving phase by macrocyclic ligands in 1,2-dichloroethane membranes and the values of log K_{ex} of their complexes.

	15-crown-5		18-crown-6		Kryptofix [2.2.2]	
Amino acid	* (%)	log K_{ex}	* (%)	log K_{ex}	* (%)	log K_{ex}
L-Valine	35	3.28 ± 0.02	79	4.36 ± 0.03	62	4.25 ± 0.02
L-Leucine	40	3.87 ± 0.06	81	5.76 ± 0.01	69	4.43 ± 0.04

The experimental parameters as specified in Table I.

The transport values and the extraction constants obtained with the ligand B18C6 were relatively good but lower than those corresponding to 18C6.

The lowest values for the extraction constants were obtained using the ligand DB18C6. The reason is twofold: first, its low aqueous solubility (9×10^{-5} M) [16] and, secondly, the influence of the benzo groups, which confer a higher rigidity on the ligand's structure. The inefficient transport was attributed to the much lower values of log K_{ex} for cation–DB18C6 [17] interaction, due to the electron withdrawing effect of the benzo groups.

The transfer of α-amino acids from aqueous phases to organic solvents was performed by transforming them into hydrophobic entities, such as ion pairs, as a result of the formation of a supramolecular complex. The complexation was carried out by macrocyclic ligands, the cavity of which may accommodate the cation — NH_3^+ of the —NH_2 group previously protonated in an acid medium, if they fit in shape and size, and a hydrophobic pair anion [18].

The experimental data for the transport of L-valine and L-leucine complexes with 15C5, 18C6 and Kryptofix [2.2.2] through a 1,2-dichloroethane liquid membrane and the corresponding values of log K_{ex} for the respective complexes are presented in Table II.

The complexes of L-valine and L-leucine with the ligand 18C6 and the Kryptofix [2.2.2] are characterised by higher values of log K_{ex} than the corresponding ones when using the ligand 15C5. The explanation is a better fit to the cavity size of the ligand 18C6 (r = 1.4 Å) and the Kryptofix [2.2.2] (r = 1.4 Å) with the —NH_3^+ group (r = 1.42 Å), as compared with the ligand 15C5 (r = 0.9 Å).

In most cases, macrobicyclic ligands such as cryptand [2.2.2] form stronger complexes with organic species, as compared with macrocyclic ligands [6]. The cryptate inclusion complexes which are formed depend on the macrobicyclic structure and can be extracted into organic solvents by coupling with an organic or inorganic anion.

The experimental data for the transport of some protonated amines through a 1,2-dichloroethane liquid membrane containing 18C6, DB18C6, Kryptofix [2.2] and Kryptofix [2.2.2] as carriers, and the corresponding values of log K_{ex} for the respective complexes are presented in Table III.

Table III. Percentage of some protonated amines transported in the receiving phase by macrocyclic ligands through 1,2-dichloroethane liquid membranes and the values of log K_{ex} of their complexes.

Amines	18-crown-6		Dibenzo-18-crown-6		Kryptofix [2.2]		Kryptofix [2.2.2]	
	* (%)	log K_{ex}	* (%)	log K_{ex}	* (%)	log K_{ex}	* (%)	log K_{ex}
$CH_3NH_3^+$	25	4.76 ± 0.08	20	2.45 ± 0.06	48	4.04 ± 0.01	65	4.47 ± 0.05
$(CH_3)_2NH_2^+$	38	4.20 ± 0.05	22	2.20 ± 0.05	44	3.91 ± 0.02	63	4.25 ± 0.03
$(C_2H_5)_2NH_2^+$	33	3.75 ± 0.02	19	2.05 ± 0.03	42	3.85 ± 0.03	58	4.02 ± 0.02
n-$C_3H_7NH_3^+$	40	5.12 ± 0.07	25	3.49 ± 0.06	52	4.34 ± 0.05	78	4.45 ± 0.03

* (%): Amine percentage found in the receiving phase after 6 h stirring.

The transport yields of the methylamine, diethylamine, dimethylamine and n-propylamine complexes with 18C6 and DB18C6 are relatively low, namely 25–40% (methylamine and n-propylamine, respectively, with 18C6 as carrier) and 19–25% (diethylamine and n-propylamine, respectively, with DB18C6 as carrier). In contrast, the transport yields of the abovementioned amine complexes with the Kryptofix [2.2] and Kryptofix [2.2.2] are relatively high, namely 42–52% (diethylamine and n-propylamine, respectively, with the Kryptofix [2.2]) and 65–78% (methylamine and n-propylamine, respectively, with the Kryptofix [2.2.2]).

The amine structure is obviously important in producing the above results, but to a lesser extent.

4. Conclusions

The experiments allow us to conclude that both the transport yields and the values of log K_{ex} for the complexes of the amino acids with macrocyclic ligands of the polyether type decrease with the number of benzo groups added to the ring of the polyether 18C6. The structures of the macrocyclic ligands, as well as the hydrophilicity of the amino acids are important to the efficiency of the transport process.

The values of log K_{ex} for various α-amino acids are not so significantly different as to offer relevant information for a similar interpretation of the experimental data. We could not find any relevant correlation between log K_{ex} of the complexes of the amino acids or amines and the macrocyclic ligands or macrobicyclic ligands on one hand, and their transport through liquid membranes on the other.

The transport yields and the values of log K_{ex} for the amines under study were relatively higher in the case of the complexes formed by the Kryptofix [2.2] and Kryptofix [2.2.2], as compared with the complexes formed by the crown ethers, respectively.

Finding the optimum separation conditions of amines and amino acids through liquid membranes by macrocyclic ligands is a challenging task for further research in the field of both molecular recognition and supramolecular chemistry.

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References

1. R.M. Izatt, B.L. Nilsen, J.J. Christensen and J.D. Lamb: *J. Membr. Sci.* **9**, 263 (1981).
2. R.M. Izatt, J.D. Lamb, N.E. Izatt, B.E. Rossiter, J.J. Christensen and B.L. Haymore: *J. Am. Chem. Soc.* **101**, 6273 (1979).
3. J.D. Lamb, J.J. Christensen, J.L. Oscarson, B.L. Nilsen, B.W. Asay and R.M. Izatt: *J. Am. Chem. Soc.* **102**, 6820 (1980).

4. J.M. Timko, R.C. Helgeson, M. Newcomb, G.W. Gokel and D.J. Cram: *J. Am. Chem. Soc.* **96**, 7097 (1974).
5. L. Mutihac, R. Mutihac, T. Constantinescu and C. Luca: *J. Incl. Phenom.* **17**, 45 (1994).
6. T.B. Stolwijk, E.J.R. Sudholter and D.N. Reinhoudt: *J. Am. Chem. Soc.* **111**, 6321 (1989).
7. R.M. Izatt, K. Pawlak, J.S. Bradshaw and R.L. Bruening: *Chem. Rev.* **91**, 1721 (1991).
8. J.M. Lehn: *Pure Appl. Chem.* **51**, 979 (1979).
9. J.P. Behr, J.M. Lehn and P. Vierling: *Helv. Chim. Acta* **65**, 1853 (1982).
10. H.-J. Buschmann and L. Mutihac: *Thermochim. Acta* **237**, 203 (1994).
11. H.-J. Buschmann and L. Mutihac: *Rev. Roum. Chim.* **39**, 563 (1994).
12. J. Takeda, N. Ikbo and N. Samata: *Talanta* **38**, 1325 (1991).
13. R.M. Izatt, J.D. Lamb and R.L. Bruening: *Sep. Sci. Technol.* **23**, 1645 (1988).
14. L. Mutihac and R. Mutihac: *Acta Chim. Hung. – Models in Chemistry* **129**, 573 (1992).
15. L. Mutihac, D.O. Popescu and R.I. Stefan: *Anal. Lett.* **28**, 835 (1995).
16. C.J. Pedersen: *J. Am. Chem. Soc.* **89**, 7017 (1967).
17. R.M. Izatt, D.W. McBride, Jr., J.J. Christensen, J.S. Bradshaw and G.A. Clark: *J. Membr. Sci.* **22**, 31 (1985).
18. L. Mutihac, C. Luca, T. Constantinescu and M. Radu: *Rev. Roum. Chim.* **36**, 91 (1991).