ELSEVIER

www.elsevier.nl/locate/carres

Carbohydrate Research 329 (2000) 409–422

CARBOHYDRATE RESEARCH

Molecular recognition of carbohydrates by a resorcinarene. Selective transport of alditols through a supported liquid membrane

Nabila Tbeur [a,b], Touria Rhlalou [a,c], Miloudi Hlaïbi [b], Dominique Langevin [a], Michel Métayer [a], Jean-François Verchère [a,*]

[a] *Laboratoire 'Polymères, Biopolymères, Membranes', UMR 6522 du CNRS, Université de Rouen, Faculté des Sciences, F-76821 Mont-Saint-Aignan, France*

[b] *Université Hassan II, Faculté des Sciences d'Aïn Chock, BP 5366, Maârif, Casablanca, Morocco*

[c] *Université Hassan I, FST de Settat, BP 577, Settat, Morocco*

Received 3 April 2000; accepted 19 June 2000

Abstract

A supported liquid membrane (SLM) containing a resorcinarene carrier has been used for the selective transport of erythritol, threitol, ribitol and xylitol from concentrated (1.0–0.01 M) aqueous solutions. The membrane is made of a microporous polytetrafluoroethylene film impregnated with a 0.01 M solution of the carrier in CCl_4. The permeabilities of the SLM for all alditols were calculated. On the basis of the flux dependence on the initial concentrations of carrier and alditol, the rate-determining step in the transport mechanism is shown to be the migration of the 1:1 carrier–carbohydrate complex in the immobilized organic phase. The flux of sugar is related to the initial concentration of alditol in the feed phase by a saturation law, which allowed the determination of the apparent diffusion coefficients and the stability constants of the resorcinarene complexes of alditols formed in the liquid membrane. © 2000 Elsevier Science Ltd. All rights reserved.

Keywords: Alditols; Facilitated transport; Resorcinarene; Stability constants; Supported liquid membrane

1. Introduction

The separation of a mixture of carbohydrates is not an easy target. Since most sugars are isomers which only differ in the configuration of specific CHOH groups, methods based on differences of chemical reactivities are generally unsuitable [1]. On the contrary, separative methods based on *molecular recognition* appear as promising tools. The principle relies on the use of a selective reagent (the host) that can form host–guest complexes with carbohydrates (the guests). When the stability of the complex varies with the structure of the sugar, the components of the mixture are complexed in different proportions, and various techniques such as extraction may be used for the isolation of the resulting complexes.

In the case of carbohydrates, a possible process is extraction into organic solvents, which is made possible by forming complexes with lipophilic guests. Such complexes, contrary to uncomplexed sugars, can be extracted from aqueous solutions. However, for use on the industrial scale, extraction methods should

* Corresponding author. Tel.: +33-235-146698; fax: +33-235-146704; http://www.univ-rouen.fr/pbm.

E-mail address: jf.verchere@univ-rouen.fr (J.-F. Verchère).

0008-6215/00/$ - see front matter © 2000 Elsevier Science Ltd. All rights reserved.
PII: S0008-6215(00)00188-9

be preferably adapted to *liquid membrane processes*, which currently offer the best strategy for environmental-friendly separations [2]. Most liquid membranes consist in an organic phase that separates two aqueous (feed and receiving) phases [2]. In this case, a double extraction process allows the overall *transport* of water-soluble species across the membrane. Selective transport occurs when the liquid membrane contains a *carrier agent* which forms complexes of different stabilities with the various sugars present in the feed phase. The main advantage of liquid membranes over classical extraction is that a very small volume of organic solvent is necessary, and since this solvent is trapped in the membrane, the quantity of harmful volatile chemicals released in the atmosphere is reduced. Moreover, operations take place at ambient temperature, with considerable reduction of the energy cost.

Supported liquid membranes (SLMs) are the preferred class of liquid membranes for practical applications, although other types of membranes are sometimes used, such as bulk liquid membranes or emulsion liquid membranes [2]. In SLMs, the organic phase is immobilized in a thin, microporous polymer film (the support) that gives mechanical toughness to the device. SLMs have been recognized as useful tools for the recuperation of valuable compounds or the elimination of pollutants, mainly metal ions from aqueous solutions [2]. The first commercial use of SLMs was for chromium removal from waste water, under the trademark SLiM of Commodore Separation Technologies, Inc. [3].

Our long-term project is aiming at the separation of carbohydrates by means of SLM processes. For this purpose, the membrane should contain a lipophilic carrier that can specifically recognize sugars by forming host–guest complexes. The chosen host belongs to the family of resorcinarenes, that are analogues of calix[4]arenes in which the four phenol units are replaced by resorcinol units [4,5]. Calixarene derivatives have been increasingly used as carriers in liquid membranes, essentially for the extraction of metal ions [6,7]. More generally, the use of various types of macrocyclic carriers in SLMs is well documented [8].

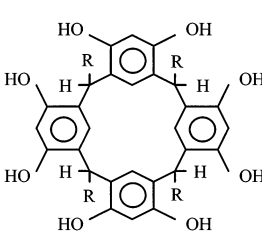

Fig. 1. Structure of resorcinarene **1** ($R = CH_3(CH_2)_{10}$).

Our initial guideline was the discovery, by Aoyama et al. [9–11], that resorcinarene **1** (Fig. 1) selectively extracts several sugars from concentrated aqueous solutions into an anhydrous CCl_4 layer. The selectivity of the extraction is due to the differences in the stabilities of the host–guest complexes formed between **1** and the sugars in the organic solvent. Our subsequent studies have shown that extraction also takes place, with a different selectivity pattern, when the organic solvent is not dried before re-extraction of the sugar into water [12]. It may indicate that the species present in the organic layer are indeed resorcinarene–sugar–water ternary complexes. This finding opened the way for the first preparation of SLMs containing the resorcinarene carrier **1**, which selectively effect the transport of sugars between two aqueous solutions [13]. We also noticed that whereas alditols are not extracted into a dried organic layer [10], they are extracted into an organic solvent saturated with water [12]. This suggested that SLMs containing **1** could be used for the transport of alditols.

Such SLMs offer an alternative to various membranes proposed in the literature for the separation of mixtures of sugars. Ion-exchange membranes have been suggested for the separation of aqueous solutions of sugars as borate complexes [14–16]. Following the pioneering work of Shinbo et al. [17], liquid membranes containing arylboronic acids as carriers were also designed for transport of carbohydrates [18–21]. Finally, plasticized cellulose triacetate (PCTA) membranes containing a quaternary ammonium salt as the active carrier have been patented for the separation of mixtures of carbohydrates (sucrose, fructose and glucose) by facilitated transport [22,23].

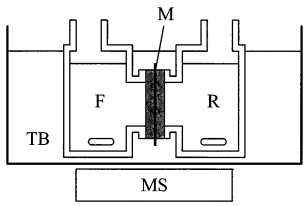

Fig. 2. Scheme of the transport cell. M is the SLM. F is the feed phase. R is the receiving phase. TB is the thermostated bath. MS is the multiple magnetic stirrer. Phases F and R are stirred using magnetic bars.

A drawback of the systems involving borate or boronate complexes is that the aqueous phases must be buffered, because the carbohydrate complexes are formed only in strongly alkaline media. Moreover, contrary to the newer PCTA membranes [22,23], most SLMs described in the literature for the transport of sugars suffer from insufficient stability and/or small fluxes.

In this study, we examined SLMs containing carrier **1** for the transport of small alditols (C_4 and C_5) that have the opposite all-erythro or all-threo configurations. Using a model previously designed for the transport of sugars [13], the stability constants of the migrating complexes were determined, in order to relate the macroscopic permeability of the membrane to the microscopic characteristics of the complex.

2. Experimental

The alditols and other chemicals were commercial products (Aldrich or Fluka) of the purest available grade, used as received. Resorcinarene **1** was synthesized according to a published procedure [10,12,13,24]. Its ^1H and ^{13}C NMR spectra, measured with a Bruker ARX-400 spectrometer, were identical with those of a commercial sample (purchased from Fluka).

The SLM support was a microporous PTFE film (Goodfellow) of thickness 63 μm. Characteristic values are porosity 84% and pore size 0.45 μm. The membrane area available for diffusion was 19.6 cm^2 (diameter, 5.0 cm).

The transport cell (Fig. 2) is made of two compartments of equal volumes (100 mL) separated by the SLM prepared by soaking a square portion of the polymer film (8 × 8 cm), into a 0.01 M solution of resorcinarene **1** in pure carbon tetrachloride, during 15 h. The cell is immersed into a thermostated bath (T, 298 K). The solutions in both compartments are stirred with magnetic bars, using a Variomag apparatus.

Two different techniques were used. In the first one (mode a), a new support, taken from two different batches, was used for each experiment. Initially, the feed compartment contained the alditol solution ($c_0 = 0.01–1.0$ M) and the receiving compartment contained pure water. In the second one (mode b), the same support was used for all runs. At equilibrium after each run, the contents of both compartments were withdrawn and mixed together, then 100 mL of the resulting solution were introduced in the feed compartment and 100 mL of water in the receiving compartment and the following run was started. This procedure was typically repeated four times. In such cases, the same membrane was used during 5 days without showing any sign of failure.

In both modes, small aliquots ($V = 1.0$ mL) of the receiving phase were withdrawn at known intervals. Some experiments were continued for 24 h, when samples of both aqueous phases were withdrawn and analyzed to ensure that equal concentrations were present, indicating that equilibrium was reached.

The samples were analyzed using a HPLC apparatus equipped with a 30-cm Phenomenex Rezex column in calcium form, maintained in an oven at 85 °C. The eluent was pure water, degassed and filtrated with a cellulose ester membrane (Millipore, pore size 0.45 μm). The flow rate was 0.6 mL/min. The pump was a Shimadzu LC-9A model. Detection was achieved with a Varian RI-4 refractometer. Typical retention times (in min) for the alditols were 19.0 (erythritol), 20.6 (threitol), 17.4 (ribitol) and 24.0 (xylitol). The alditol concentrations were determined by analyzing chromatographic data with the Varian Star software. All experiments were duplicated and were reproducible within 3% accuracy. Calculations were made with the Sigmaplot 4 software.

3. Results

Transport experiments.—Beside their numerous advantages, the main drawback of most SLMs is instability, as the organic solution is eventually displaced from the support by the aqueous feed and receiving phases. Various causes for membrane instability have been discussed elsewhere [25]. However, the SLMs studied in this work seem to represent an ideal case, since the transported carbohydrates are almost insoluble in the organic phase, whereas the carrier is insoluble in aqueous medium, avoiding thus its washing-out from the organic layer.

The high stability of our membranes was previously demonstrated [13]. In the case of the transport of sugars, the SLMs could be used for 10 days. In the present study with alditols, most experiments were carried out during 5 days with the same membrane (mode b), without any observation of leaking. Another advantage of our system is that only facilitated transport of the sugars takes place, as passive diffusion of the sugars across the SLM cannot be detected in the absence of carrier. This result is consistent with the hydrophobic nature of the PTFE film, and with the fact that uncomplexed alditols are not soluble in carbon tetrachloride.

A special attention was given to the synthesis of carrier **1**, because this macrocycle is known to possess several diastereoisomeric forms [26,27]. We ensured the exclusive formation of the thermodynamically more stable crown form by heating the crude condensation mixture at reflux for at least 4 h [26]. The stereochemical implications of the complexation of chiral guests with the achiral host **1** have been discussed elsewhere [28].

Measurements of permeabilities.—The principle of the calculation of permeabilities and fluxes has been developed elsewhere [13]. The transport rate is measured by determining the increase of the alditol concentration c_R in the receiving phase versus time t. This rate is related to the flux J of alditol by Eq. (1),

$$dc_R/dt = JS/V \quad (1)$$

where S is the membrane area and V is the volume of the receiving phase.

When the system reaches a quasi-steady state, the flux J is related to Δc, the difference between the concentrations of alditol in the feed (c_F) and the receiving phases (c_R), and the membrane thickness l by Eq. (2) derived from Fick's First Law,

$$J = P\Delta c/l \quad (2)$$

where P is the permeability of the membrane.

Since the flux of alditol is very large, the concentration (c_R) of the receiving phase is not negligible versus the concentration (c_F) of the feed phase. Thus, Δc is calculated using Eq. (3) where c_0 is the initial concentration of alditol in the feed phase:

$$c_F = c_0 - c_R \text{ and } \Delta c = c_0 - 2c_R \quad (3)$$

Combining Eqs. (1)–(3) yields differential Eq. (4):

$$P\ dt = (lV/S)dc_R/(c_0 - 2c_R) \quad (4)$$

Integration of both terms of Eq. (4) yields Eq. (5):

$$P(t - t_L) = (lV/S)(1/2)\ln\ c_0/(c_0 - 2c_R) \quad (5)$$

which shows that, after an induction period (t_L) that may last up to several hours, the term $-\ln(c_0 - 2c_R)$ is a linear function of t. The plots drawn (Fig. 3) for the transport of the four alditols were indeed straight lines for $t > t_L$. The permeability P values for the various alditols were calculated (Tables 1 and 2), using Eq. (6), from the slopes a of the plots.

$$P = alV/2S \quad (6)$$

The intersection of the linear section with the time axis defines the 'true initial time' t_L at which the alditol concentrations in the aqueous phases are $c_F \approx c_0$ and $c_R \approx 0$. At this instant, the initial value of the flux, J_i, can be calculated by Eq. (7) derived from Eq. (2).

$$J_i = Pc_0/l \quad (7)$$

Modeling and parameters of the complex-forming reaction.—The mechanism (scheme in Fig. 4) for the overall transport of an alditol S by the carrier C across the liquid membrane is known to involve five consecutive steps [13]:

1. diffusion of the alditol through the diffusion layer, from the bulk of feed phase to the interface,

2. reaction between the alditol and the carrier at the feed phase–SLM interface, to form a complex soluble in the membrane organic phase,
3. diffusion into the SLM, where the complex migrates towards the SLM-receiving interface and the carrier migrates back towards the feed–SLM interface,
4. dissociation of the complex at the SLM-receiving phase interface, to release the alditol into the receiving phase and the carrier into the membrane,
5. diffusion of the liberated alditol through the diffusion layer to the bulk of receiving phase.

Steps (1) and (5) are presumably fast, because both aqueous phases are stirred. Steps (2) and (4) correspond to the fast formation and dissociation of the complex at the interfaces. Hence, as in most studies with SLMs, the rate-determining step is expected to be step (3), i.e., the diffusion of the complex into the organic solvent that fills the pores of the polymeric support.

The complex was assumed to be a 1:1 alditol–resorcinarene species. The stoichiometry of such complexes could not be found in the literature. However, in the case of ribose, the

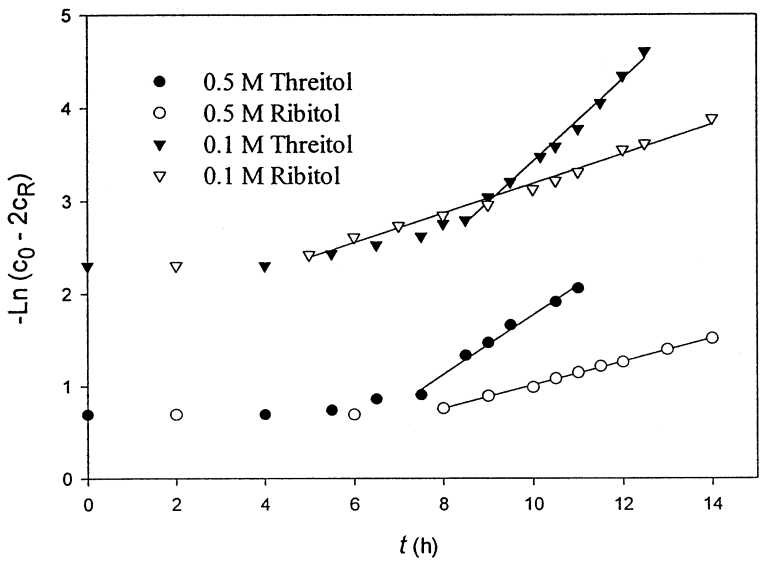

Fig. 3. Plots of $-\ln(c_0 - 2c_R)$ vs. time t for the transport of threitol and ribitol.

Table 1
Transport of tetritols across a SLM. Influence of the alditol concentration [a]

Alditol	c_0 (feed phase) (mmol cm^{-3})	$a \times 10^5$ (s^{-1})	$P \times 10^7$ (cm^2 s^{-1})	$J_i \times 10^5$ (mmol cm^{-2} s^{-1})
Erythritol	1.00	3.89	6.25	9.92
Erythritol	0.75	4.42	7.10	8.45
Erythritol	0.50	5.64	9.06	7.19
Erythritol	0.20	7.42	11.92	3.78
Erythritol	0.10	8.56	13.75	2.18
Threitol	1.00	7.14	11.47	18.21
Threitol	0.50	8.89	14.28	11.34
Threitol	0.25	10.74	17.26	6.85
Threitol	0.10	11.75	18.89	3.00
Threitol [b]	0.10	11.94	19.19	3.05

[a] Conditions: t, 25 °C, $[C]_0 = 0.010$ mol L^{-1}, C is the carrier, uncertainties $\pm 5\%$. a is the slope of a linear plot of Eq. (5). Permeability P is calculated from Eq. (6). J_i, flux at the end of the induction period, is calculated using Eq. (7). For the PTFE film, the area S is 19.6 cm^2 and the thickness l is 0.0063 cm. Porosity 84%, pore size 0.45 μm. All experiments were carried out following mode (a).

[b] The membrane was equilibrated with water for 12 h before starting the experiment.

Table 2
Transport of pentitols across a SLM. Influence of the alditol concentration

Alditol	Mode [a]	c_0 (feed phase) (mmol cm^{-3})	$a \times 10^5$ (s^{-1})	$P \times 10^7$ (cm^2 s^{-1})	$J_i \times 10^5$ (mmol cm^{-2} s^{-1})
Xylitol	a	1.00	3.64	5.85	9.28
Xylitol	a	0.80	3.91	6.28	7.98
Xylitol	a	0.40	4.56	7.32	4.65
Xylitol	a	0.25	4.75	7.63	3.03
Xylitol	b	0.40	4.68	7.53	4.78
Xylitol	b	0.20	5.14	8.26	2.62
Xylitol	b	0.10	5.21	8.38	1.33
Xylitol	b	0.05	5.31	8.53	0.677
Ribitol	a	1.00	2.78	4.47	7.09
Ribitol	a	0.50	3.53	5.67	4.50
Ribitol	a	0.25	4.04	6.50	2.58
Ribitol	a	0.10	4.41	7.09	1.12
Ribitol	b	0.10	4.41	7.09	1.12
Ribitol	b	0.05	4.53	7.28	0.577
Ribitol	b	0.025	4.61	7.41	0.294
Ribitol	b	0.0125	4.64	7.46	0.148

[a] Mode a: as in Table 1. Mode b: same conditions as in Table 1, except that the whole series of runs was performed with a single membrane, by stepwise dilution of the feed phase.

existence of a 1:1 complex with **1** in CCl_4 was reported by Aoyama et al. [9]. Our own results on the extraction of sugars and alditols by **1** also supported the formation of a 1:1 complex [12].

In the transport experiments, the initial alditol concentration c_0 in the feed phase is much larger (50–100 fold) than the initial carrier concentration ($[C]_0 = 1 \times 10^{-2}$ M) in the organic phase. Therefore, the concentration [CS] of the 1:1 carrier–alditol complex in the membrane will be limited by the initial concentration of the carrier.

The relationship between the flux J and the alditol concentration $[S]_t$ in the feed phase, at any time, was established using a kinetic model in which the diffusion step (3) is assumed to be rate-determining [13]. The reaction between the alditol and the carrier at both interfaces, may be considered as a fast heterogeneous equilibrium:

$$C\ (org) + S\ (aq) \rightleftharpoons CS\ (org)$$

where (org) and (aq) refer to the organic phase and feed phase, respectively.

The concentration $[CS]_i$ of complex at the interface obeys the mass action law, Eq. (8),

$$[CS]_i = K[C]_i[S]_i \quad (8)$$

where K is the formation constant of the complex, $[C]_i$ is the concentration of the carrier in the membrane, near the interface, and $[S]_i$ is the concentration of alditol in the feed phase, near the interface.

In the rate-determining step (3), the flux J is determined by Eq. (9), derived from Fick's First Law by assuming that the concentration of complex is practically nil at the receiving phase–membrane interface.

$$J = (D^*/l)[CS]_i \quad (9)$$

D^* is the apparent diffusion coefficient and l is the membrane thickness.

Because the alditol S is in excess in the feed phase, $[CS]_i \ll [S]_i$ and $[S]_i = c_0$, the initial concentration of alditol. The small concentration $[C]_0$ of carrier immobilized in the SLM is constant, but the free carrier is in equilibrium

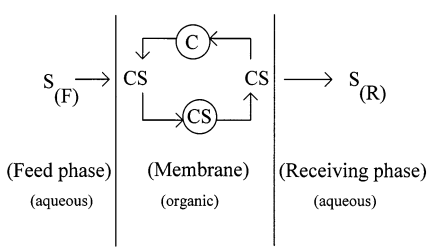

Fig. 4. Scheme of the facilitated transport of a carbohydrate (S) by resorcinarene **1** (C) through the SLM.

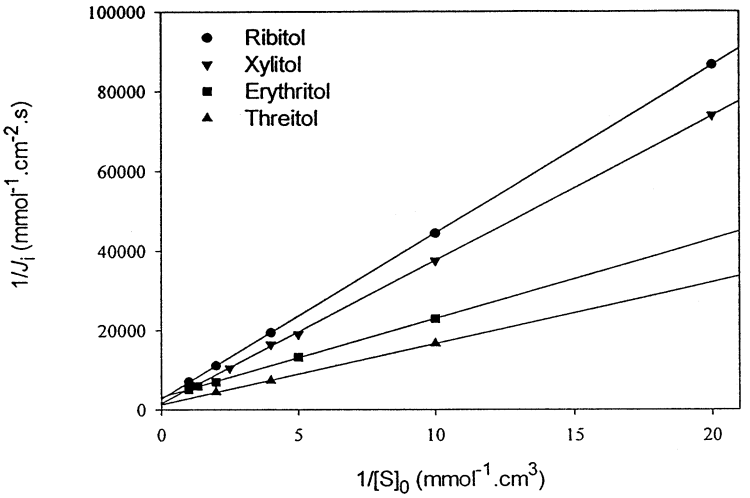

Fig. 5. Plots of $1/J_i$ vs. $1/c_0$ for the transport of four alditols across the SLM.

Table 3
Apparent diffusion coefficients and stability constants for the alditol complexes of **1** [a]

Alditol	$10^4 \times D^*$ ($cm^2\ s^{-1}$)	K (mol^{-1} L)	$10^3 \times$ slope	$10^3 \times$ intercept	r
Erythritol	1.00	1.58	3.97	6.27	0.999
DL-Threitol	2.53	0.81	3.08	2.49	0.997
Ribitol [b]	1.11	0.68	8.33	5.68	0.9999
Xylitol [b]	1.92	0.46	7.21	3.28	0.9998
L-Arabinose [13]	5.95	0.18	5.88	1.06	0.999

[a] Slope and intercept were obtained from a plot of $1/J_i$ vs. $1/c_0$, Eq. (11). r is the correlation coefficient. Accuracy: $K \pm 0.1$; $D^* \pm 0.1 \times 10^{-4}$. K is the equilibrium constant for the heterogeneous reaction: C (org) + S (aq) $\rightleftharpoons$ CS (org).
[b] All data in Table 2 were used to draw a single plot for xylitol or ribitol.

with the complex. The flux under the initial conditions J_i is given by Eq. (10):

$$J_i = (D^*/l)[C]_0 K c_0/(1 + K c_0) \quad (10)$$

The postulated mechanism requires that J_i is proportional to the initial concentration of carrier $[C]_0$ and obeys a saturation law with respect to c_0. In order to test the proposed relationship, Eq. (10) was linearized as a Lineweaver–Burk plot, Eq. (11):

$$1/J_i = (l/D^*[C]_0 K)(1/c_0) + l/D^*[C]_0 \quad (11)$$

Transport experiments with the four alditols were carried out in which c_0 varied in the ranges 0.1–1.0 M for the tetritols and 0.0125–1.0 M for the pentitols. Eq. (11) was checked by plotting the values of $1/J_i$ versus $1/c_0$, which yielded very good linear relationships in every case (Fig. 5). From the slopes and intercepts of the above plots, the apparent diffusion coefficients and stability constants of the complexes were calculated through linear regressions (Table 3):

$$K = \text{intercept/slope and}$$

$$D^* = l/[C]_0 \text{ intercept.}$$

To our knowledge, this is the first report on the determination of stability constant of the complexes formed between alditols and resorcinarene **1**. A value was reported for a sugar–resorcinarene complex in the case of arabinose [13].

Examination of Fig. 5 shows that the pair of threo alditols yields plots with nearly the same intercept. The same phenomenon is observed with the pair of erythro alditols. This should be related to the close values of the corresponding apparent diffusion coefficients D^*, since intercept $= l/D^*[C]_0$.

Hence, the experimental results support a mechanism in which diffusion of the CS com-

plex is the rate-determining step. However, the values of D^* seem very high, compared to usual diffusion coefficients for carbohydrates. It suggests that the migration of the complex is not a pure diffusion process. This point is discussed hereafter.

Induction period and mechanism of transport.—In our first experiments, transport of the alditol began only after a lengthy induction period which lasted 4–7 h and did not depend on the nature of the alditol and its concentration. The hypothesis that transport would occur as a consequence of membrane breakdown, and thus correspond to passive transport, can be rejected on the following grounds:

1. the transport of different alditols takes place at different rates. This may obviously be attributed to differences in the values of the diffusion coefficients of alditols, but in this case, both tetritols, which have the same size and likely possess similar diffusion coefficients, would be expected to migrate at similar rates, which is not true, as threitol migrates faster than erythritol,
2. one may expect that, for passive transport of alditols, the initial flux J_i is proportional to the initial alditol concentration c_0. On the contrary, our results clearly establish that a saturation law is obeyed, since $1/J_i$ is proportional to $1/c_0$. Such a law can only be explained if the reversible formation of a carrier–alditol complex occurs before the transport step.

Nevertheless, since the proposed mechanism does not account for the existence of the induction period, another slow event or reaction must precede the transport step. We reasoned that such a reaction could be the formation of a water–resorcinarene complex, since a proton NMR spectroscopic study by Aoyama et al. [9] showed that **1** forms in CCl_4 a complex with water formulated $C(H_2O)_4$. The kinetics were not studied, but it was reported that the mixing of both phases was maintained for 24 h. Accordingly, under our standard conditions, the membrane contains the anhydrous solution of **1** when the experiment starts, and since the concentration of water in the feed phase is much larger than that of carbohydrate, a likely preliminary step would be the uptake of water by the membrane.

In order to test this hypothesis, an experiment (A) was performed in which both compartments of the cell were filled with water, and equilibration of the SLM (prepared as usual) was allowed overnight (12 h). Then, water in the feed compartment was replaced by an aqueous 0.1 M solution of threitol and transport was monitored. Under these conditions, the induction period was found to be short (0.8 h) and afterwards, the permeability and flux were almost similar to those found for a 0.1 M threitol solution studied under our standard conditions (Table 1). This result demonstrates that the induction period is due to uptake of water by the membrane.

The small induction period observed when the membrane is initially equilibrated with water was assumed to represent the time necessary for exchanging water for the alditol in the $C(H_2O)_4$ complex. In order to test this idea, experiment (B) was carried out. A classical transport experiment was performed with a ribitol solution ($c_0 = 0.25$ M) in the feed compartment. When equilibrium was reached (after 24 h), the concentrations of both aqueous phases were determined to be 0.125 M, as expected. Then, the receiving phase was replaced by pure water and the transport was monitored. The results were very close to those calculated for $c_0 = 0.125$ M, using the plots drawn with Eq. (11), but with the important difference that the induction period completely disappeared:

$$\text{Calcd: } P = 6.98 \times 10^{-7} \text{ cm}^2 \text{ s}^{-1} \text{ and } J_i = 1.38 \times 10^{-5} \text{ mmol cm}^{-2} \text{ s}^{-1}$$

$$\text{Obsd: } P = 6.71 \times 10^{-7} \text{ cm}^2 \text{ s}^{-1} \text{ and } J_i = 1.33 \times 10^{-5} \text{ mmol cm}^{-2} \text{ s}^{-1}$$

The difference between experiments (A) and (B) is that in (B), when the transport of 0.125 M alditol begins, the membrane is equilibrated with the aqueous solution of alditol, and not with pure water as in (A). Thus, the small induction period observed for (A) indeed corresponds to the time necessary for saturating the membrane with the alditol.

In consequence, we repeated the complete sets of experiments with xylitol and ribitol

under the same conditions as in (B). In the first run, the membrane was equilibrated overnight with water before introducing the alditol solution in the feed compartment. The transport was monitored until completion of equilibrium, where both compartments contained identical solutions of the alditol. Then, the solution in the receiving compartment was discarded and replaced by pure water, and the transport was monitored. This procedure was repeated for 4 days, allowing the study of transport of one alditol at four decreasing concentrations with a single membrane. The results presented in Table 2 show that data obtained with both techniques are in good agreement. Moreover, the suppression of the induction period greatly facilitates the determination of the slopes of plots of the function $-\ln(c_0 - 2c_R)$ versus t.

Finally, these series of experiments are valuable tools for checking that (i) the membrane transport of alditols is a reproducible process and (ii) no breakdown of the membrane takes place during 4 days.

In conclusion, step (2) of the transport mechanism is indeed the combination of three successive steps occurring at the feed phase–membrane interface. In the first step (5–7 h), the free carrier C present in the organic phase reacts with water of the feed phase to form the $C(H_2O)_4$ complex reported by Aoyama et al. [9]. This slow step is followed by a faster exchange step in which the alditol of the feed phase competes with water for the carrier in the organic phase, to form the carrier–sugar complex CS that is the migrating species. However, if the alditol does not replace all four water molecules in $C(H_2O)_4$, the transported species may be a ternary carbohydrate–water–resorcinarene species. Finally, in the third step, transport of the carbohydrate occurs as the CS complex migrates towards the receiving phase, while uncomplexed carrier molecules liberated in the SLM migrate back towards the feed phase.

Transport of mixtures of alditols.—In order to explore the possible application of membrane transport to the separation of carbohydrates, we studied the transport of mixtures of alditols through our SLM. Two cases were selected. The first example, a mixture of the isomeric xylitol and ribitol, is useful as a model for a separation based on different configurations. The second example, a mixture of erythritol and xylitol, was considered because the permeabilities of these alditols are very different and suggested that separation was possible. In both cases, the feed phase was an aqueous solution of both 0.10 M alditols. The concentrations of the alditols in the receiving phase were monitored as usual. The retention times are sufficiently different for a good separation of the chromatographic peaks of each alditol. In both experiments (carried out in mode a) transport occurred after a long induction period. The suppression of this induction period (by using mode b) was not attempted, because the existence of different induction periods might be favorable for the expected separations.

The results (Fig. 6) show that migration of the alditols occurs at different rates. The rates vary in the order: erythritol > xylitol > ribitol, in agreement with the results found for alditols alone. The permeabilities for alditols in mixtures are a little smaller than those calculated for the single alditols (Table 4). In the case of xylitol, which was present in both mixtures, the fluxes are of the same magnitude, either alone or in mixture with erythritol or ribitol. Whereas xylitol competes either with a faster alditol (erythritol) or a slower alditol (ribitol), the fluxes of xylitol are always smaller than for xylitol alone.

This decrease of the flux may be due to (i) the presence of the second alditol that increases the overall concentration of alditols to 0.2 M (this is known to decrease P) and (ii) the fact that the unchanged amount of carrier is now shared between both alditols. Since the transported species is a 1:1 alditol–carrier complex, competition between both alditols may take place at the interface, making a smaller effective concentration of the carrier available for each alditol.

For the xylitol–ribitol mixture, the ratio of permeabilities is 1.15. For the single alditols, it is 1.18 and therefore, both permeabilities decrease by the same factor (0.89) in the mixture. The conditions of the xylitol–ribitol experiment were very unfavorable, because both alditols have quite similar permeabilities, and in this case, an efficient separation could not be expected in a single-step process. However, a significant enrichment of the receiving phase in

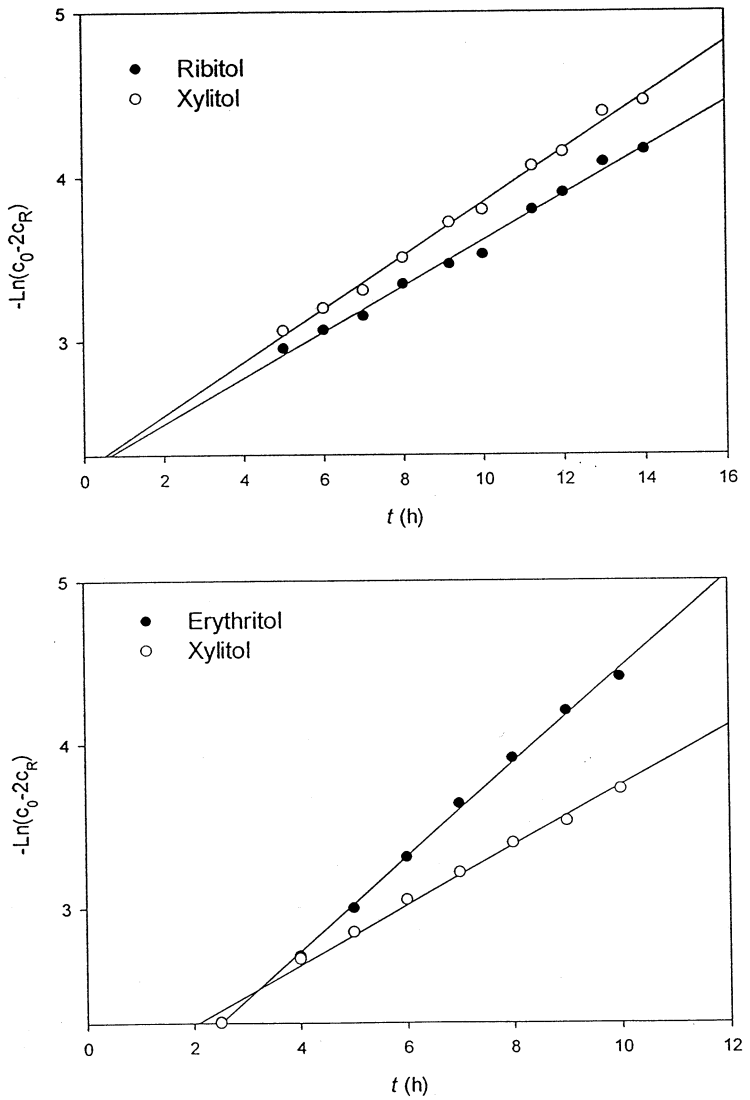

Fig. 6. Transport of mixtures of alditols across the SLM. (a) Mixture of ribitol and xylitol (both 0.1 M). (b) Mixture of erythritol and xylitol (both 0.1 M).

Table 4
Transport of equimolar mixtures of two alditols [a]

Alditols [b]	c_0 (feed phase) (mmol cm^{-3})	$a \times 10^5$ (s^{-1})	$P \times 10^7$ (cm^2 s^{-1})	$J_i \times 10^5$ (mmol cm^{-2} s^{-1})
Ribitol (m)	0.10	3.97	6.38	1.01
Xylitol (m)	0.10	4.58	7.36	1.17
Erythritol (m)	0.10	8.03	12.90	2.05
Xylitol (m)	0.10	4.75	7.63	1.21
Ribitol (s)	0.10	4.41	7.09	1.12
Erythritol (s)	0.10	8.56	13.75	2.18
Xylitol (s)	0.10	5.21	8.38	1.33

[a] Conditions: t, 25 °C, $[C]_0 = 0.010$ mol L^{-1}, C is the carrier, uncertainties $\pm$ 5%. The cell and membrane are the same as in Tables 1–3.

[b] (m) mixture of two alditols; (s) single alditols.

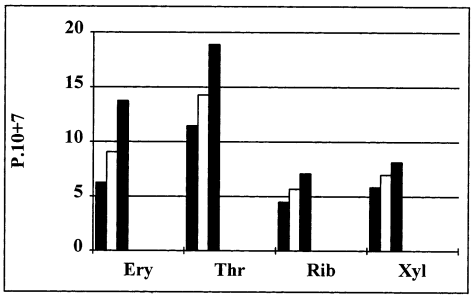

Fig. 7. Compared permeabilities of the SLM for the alditols. From left to right: $c_0 = 1.0$, 0.5, and 0.1 M, respectively.

xylitol was indeed observed. The practical enrichment ratio of xylitol into the receiving phase, defined as:

$$([\text{xylitol}] - [\text{ribitol}])/([\text{xylitol}] + [\text{ribitol}])$$

was 5.5–5% when the transport was carried out during 4–9 h. This enrichment ratio decreased with time. For a transport time of 7 h, the enrichment ratio was 5.1%, corresponding to extraction yields of 63% (xylitol) and 57% (ribitol).

For the erythritol–xylitol mixture, the ratio of permeabilities is 1.69. For the single alditols, it is 1.64. The permeabilities in the mixture are smaller than the permeabilities for the single alditols (ratio 0.94). The receiving phase is enriched in erythritol. When the transport was carried out during 5–9 h, the practical enrichment ratio of erythritol was 9–10%. Contrary to the ribitol–xylitol mixture, the enrichment ratio increased during 7 h, then decreased. For a transport time of 7 h, the larger enrichment ratio was 10.3%, corresponding to extraction yields of 74% (erythritol) and 60% (xylitol). As expected, the separation is more efficient when the alditols have very different permeabilities.

Another result was that the induction period was different for both alditols. For example, the migration of xylitol, which migrates slower than erythritol, starts before that of erythritol (Fig. 6). This observation provides additional evidence that the fast migration of alditols is not due to membrane breakdown. If it was the case, both carbohydrates would begin to migrate at the same time, probably at similar rate. Further studies on various mixtures of carbohydrates are in progress, in order to assess the utility of the method.

4. Discussion

Comparison with extraction data.—In their pioneering studies, Aoyama et al. reported that ribitol was not extracted by an *anhydrous* solution of **1** in carbon tetrachloride [9,10]. Instead, Verchère et al. demonstrated that by using a solution of **1** in CCl_4 *saturated with water* [12], several alditols were extracted as well as sugars, although the extraction selectivity was not so good as in anhydrous conditions. Comparison of these results indicates that the extraction of carbohydrates by **1** is considerably modified in the presence of water, probably because the carrier can complex water as well as carbohydrates [10]. This effect of water is obviously present in our SLM transport process. Whether it implies the formation of true ternary complexes, or the formation of water pockets that dissolve the sugar in the organic solvent, cannot be ascertained at this stage of the program. However, the selectivity observed in the transport process suggests that ternary complexes with quite different stability constants may be involved.

The transport of xylitol is faster than that of ribitol (Table 2). On the other hand, the percentage of extraction of 2.5 M aqueous solutions of alditols by an organic solution of **1** in CCl_4 saturated with water, after 24 h, was reported to be 53% for ribitol and 25% for xylitol [12]. Such a lack of correlation between permeabilities and extraction data was already mentioned in the case of sugars [13] and may be related to the absence of the kinetic factor in the extraction experiments, in which measurements are only performed at equilibrium.

Moreover, the stability constant of the complex is known to influence the transport phenomenon in a two-fold way, because transport is hampered when the complex is very weak *and also* when it is very strong. Maximum rates are observed for complexes of medium stability, because the complex must form in sufficient amount at the feed phase–membrane interface, but it must also readily dissociate at the receiving phase–membrane interface [29].

Parameters of the permeabilities.—The permeabilities of the SLM for the four alditols at three initial concentrations are compared in Fig. 7. Two important parameters are visualized: the size of the molecule and its configuration. The order of permeabilities is: threitol > erythritol > xylitol > ribitol, indicating that favorable factors for the transport of alditols are shortness of chain length ($C_4 > C_5$) and threo configuration (threo > erythro), as tetritols are transported faster than pentitols and threo compounds migrate faster than erythro compounds. The effect of the molecule size was already reported in a study of the transport of aldoses, in which larger permeabilities were found for pentoses (xylose, ribose and arabinose) than for hexoses (glucose and mannose) [13]. In the same study, the variations of the permeabilities in a series of pentoses and in a series of hexoses indicated another specific effect due to the sugar configuration, but no clear relationship was found at this time.

The determination of the stability constants and apparent diffusion coefficients for the complexes of four alditols (Table 3) offers the possibility of dissecting the influence of these parameters on the transport of alditols across the SLM.

The *influence of chain length* can be appreciated by comparing tetritol–pentitol couples of identical configurations. The complex of threitol is stronger than that of xylitol (K, 0.81 vs. 0.46) and the complex of erythritol is stronger than that of ribitol (K, 1.58 vs. 0.68). Thus, the stability constant K noticeably depends on chain length and decreases when chain length increases. On the other hand, the diffusion coefficient D^* depends little on chain length, as demonstrated by the comparison of the threitol–xylitol and erythritol–ribitol couples. It suggests that variations in the chain length only influence K.

The *influence of the configuration* can be appreciated by comparing alditols with identical chain lengths. In this case, the complex of erythritol is stronger than that of threitol, (K, 1.58 vs. 0.81) and the complex of ribitol is stronger than that of xylitol, (K, 0.68 vs. 0.46). A rule-of-thumb is that complexes of erythro alditols are nearly twice as strong as those of threo alditols. A similar difference is found for the diffusion coefficient D^*, since the values for threo compounds threitol and xylitol (which are nearly insensitive to chain length) are twice as large as those for erythro compounds erythritol and ribitol.

Contrary to chain length, the alditol configuration influences both K and D^* values. Moreover, the effects of chain length and configuration appear to be additive for the K values, i.e., the ratio of stability constants for the complexes of erythritol and xylitol is close to 4 (K, 1.58 vs. 0.46).

Because the diffusion coefficients do not vary with chain length, the *permeabilities* for threo alditols are generally twice as large as those for erythro alditols, at identical number of carbons. On the other hand, when the number of carbons increases, the variations of permeability with chain length are directly related to the decrease in complex stability. Accordingly, for the threitol–xylitol and erythritol–ribitol couples, the variations of permeabilities are only related to the variations of the stability constants, i.e., the complexes that migrate faster are the strongest ones.

The influence of the stability constant K is more complex, because K depends on both chain length and configuration, and is related to P via a saturation law. In fact, threo alditols migrate faster than erythro alditols, whereas their K values are smaller.

The origin of the variations of the stability constants with the alditol configuration can be inferred from the results obtained with the tetritols. Erythritol and threitol only differ by their configuration at C-2,3 and thus, the interaction of **1** with an alditol appears to be stronger when an erythro diol site is available. Moreover, the weaker interactions observed with both pentitols indicate that the presence of a third vicinal hydroxyl group is not a favorable factor. Thus, it is unlikely that **1** binds with a triol site, and we believe that the site of binding is HO-2,3 for erythritol and threitol, and also for ribitol and xylitol.

Comparison with the transport of arabinose.—The values of the apparent diffusion coefficients D^* and stability constants K of the complexes are displayed in Table 3. A comparison is possible with arabinose [13]. All

complexes of alditols are stronger than the arabinose complex (K, 0.18 mol L^{-1}). On the other hand, D^* is much larger for the arabinose complex than for all the alditol complexes. The difference may be due to the larger stiffness of the pyranose cycle of arabinose. In this case, the hydroxyl groups of the binding diol site are probably unable to adopt the same conformation as in the acyclic alditols.

The problem of the determination of the stability constants of the complexes formed between **1** and polyols has been addressed by Aoyama et al. in two ways [30,31]. In both cases, only 1:1 complexes were detected.

First, the stability constants of the complexes of **1** with various diols in anhydrous $CDCl_3$ have been determined by NMR spectroscopy [30]. In this case, the reported values are true equilibrium constants for the formation of CS species. The drawback is that this method cannot be applied to sugars that are not soluble in organic solvents. In a second approach, the stability constants have been determined in aqueous medium, also from NMR spectroscopic studies, using a sulfonated resorcinarene that is water soluble [31].

The reported values cannot be compared with those found in this work, which refer to the formation of the carrier–carbohydrate species at the SLM interface. Thus, the reaction is a heterogeneous equilibrium involving the aqueous alditol, together with the carrier and the complex dissolved in the organic phase. Moreover, the value of K is probably influenced by the presence of the polymeric support (PTFE) that surrounds the pores. Finally, if the 'CS' complex is indeed a ternary species such as CSW_3, where W stands for water, the formation constant is an apparent constant that includes a constant term for the activity of water.

Nature of the migration step.—The apparent diffusion coefficients (Table 3) determined for the complexes seem quite high, in view of the much smaller diffusion coefficients of the constituents of the complexes. The diffusion coefficient of glucose in aqueous solution is $D = 6.4 \times 10^{-6}$ cm^2 s^{-1} [16], whereas that of a calix[4]arene in chloroform is $D = 6.8 \times 10^{-6}$ cm^2 s^{-1} [32].

The large values found with our SLM for alditols and also arabinose [13] probably indicate that the migration of the complex is not purely diffusion-controlled and may include a convective contribution due to moves of the organic solvent in the pores. It would not be unlikely that the organic solution is not completely immobilized in our SLM, which possesses unusually large pores with respect to membranes commonly used for transport experiments. The fundamental point is that, even if the mechanism does not consist in a pure diffusion process, the migration of the various carbohydrate–carrier complexes occurs at different rates and exhibits a saturation law, demonstrating that the resorcinarene carrier actually participates in the transport process.

5. Conclusions

The transport of four alditols was studied through a SLM made of a porous PTFE film containing a solution of resorcinarene **1** in CCl_4. The membrane is exceptionally permeable, allowing a complete transport of all alditols within 12–15 h. The dependence of the flux versus the initial concentrations of alditol and carrier shows that the rate-determining step of the transport mechanism is the migration of 1:1 alditol–carrier complexes within the membrane, which is not purely diffusion controlled. The apparent diffusion coefficients and the stability constants of the complexes were calculated, affording insight of the nature of the resorcinarene complexes. Although all carbohydrate–resorcinarene complexes are very weak (stability constants, $K \approx 1$ mol^{-1} L), alditols are more firmly bound to the carrier than arabinose [13].

The fluxes depend both on the size and configuration of the alditols: migration is faster for tetritols than for pentitols, and for threo than erythro compounds. It suggests that our SLM may perform the separation of mixtures of alditols, and accordingly, enrichment of the receiving phase with the 'fast' alditol was demonstrated for two mixtures. Considering the remarkable stability and large permeability of the SLM for aldoses and aldi-

tols, these findings open a new perspective on membrane processes for the separation of aqueous mixtures of carbohydrates.

Acknowledgements

Professor Nguyen Quang Trong (UMR 6522, Université de Rouen, France) is gratefully acknowledged for stimulating discussions and helpful advice in the design of some experiments.

References

[1] M.F. Chaplin, J.F. Kennedy (Eds.), *Carbohydrate Analysis. A Practical Approach*, IRL, Oxford, UK, 1986.
[2] (a) R.D. Noble, J.D. Way (Eds.), *Liquid Membranes: Theory and Applications*, ACS Symp. Ser., 347 (1987). (b) R.A. Bartsch, J.D. Way (Eds.), *Chemical Separations with Liquid Membranes*, ACS Symp. Ser., 642 (1996) 1–408.
[3] SLiM, corporate communication from Commodore Separation Technologies, Inc.
[4] V. Böhmer, *Angew. Chem., Int. Ed. Engl.*, 34 (1995) 713–745.
[5] P. Timmerman, W. Verboom, D. Reinhoudt, *Tetrahedron*, 52 (1996) 2663–2704.
[6] C. Wieser, C.B. Dieleman, D. Matt, *Coord. Chem. Rev.*, 165 (1997) 93–161.
[7] M.R. Yaftian, M. Burgard, C.B. Dieleman, D. Matt, *J. Membr. Sci.*, 144 (1998) 57–64.
[8] W.F. van Straaten-Nijenhuis, F. de Jong, D.N. Reinhoudt, *Recl. Trav. Chim. Pays-Bas*, 112 (1993) 317–324.
[9] Y. Aoyama, Y. Tanaka, H. Toi, H. Ogoshi, *J. Am. Chem. Soc.*, 110 (1988) 634–635.
[10] Y. Aoyama, Y. Tanaka, S. Sugahara, *J. Am. Chem. Soc.*, 111 (1989) 5397–5404.
[11] Y. Tanaka, Y. Ubukata, Y. Aoyama, *Chem. Lett.*, (1989) 1905–1908.
[12] T. Rhlalou, M. Labbé, M. Ferhat, J.-F. Verchère, *Carbohydr. Lett.*, 3 (1999) 263–269.
[13] T. Rhlalou, M. Ferhat, M.A. Frouji, D. Langevin, M. Métayer, J.-F. Verchère, *J. Membr. Sci.*, 168 (2000) 63–73.
[14] Y. Shigemasa, S. Okamoto, H. Sahiwa, H. Saimoto, *Chem. Lett.*, (1990) 433–436.
[15] M. Igawa, T. Sekimoto, H. Okochi, *J. Membr. Sci.*, 98 (1995) 177–180.
[16] S. Ouahid, M. Métayer, D. Langevin, M. Labbé, *J. Membr. Sci.*, 114 (1996) 13–25.
[17] T. Shinbo, K. Nishimura, T. Yamagushi, M. Sugiura, *J. Chem. Soc., Chem. Commun.*, (1986) 349–351.
[18] B.D. Smith, in R.A. Bartsch, J.D. Way (Eds.), *Chemical Separations with Liquid Membranes*, ACS Symp. Ser., 642 (1996) 194–215.
[19] B.D. Smith, *Supramol. Chem.*, 7 (1996) 55–60.
[20] M.F. Paugam, J.A. Riggs, B.D. Smith, *J. Chem. Soc., Chem. Commun.*, (1996) 2539–2540.
[21] M.J. Karpa, P.J. Duggan, G.J. Griffin, S.J. Freudigmann, *Tetrahedron*, 53 (1997) 3669–3678.
[22] J.A. Riggs, B.D. Smith, *J. Am. Chem. Soc.*, 119 (1997) 2765–2766.
[23] B.D. Smith, J.A. Riggs, US Patent 5,800,624 (1998); *Chem. Abstr.*, 129 (1998) 218290r.
[24] L.M. Tunstadt, J.A. Tucker, E. Dalcanale, J. Weiser, J.A. Bryant, J.C. Sherman, R.C. Helgeson, C.B. Knobler, D.J. Cram, *J. Org. Chem.*, 54 (1989) 1305–1312.
[25] A.M. Neplenbroek, D. Bargeman, C.A. Smolders, *J. Membr. Sci.*, 67 (1992) 121–132.
[26] L. Abis, E. Dalcanale, A. Du vosel, S. Spera, *J. Org. Chem.*, 53 (1988) 5475–5479.
[27] L. Abis, E. Dalcanale, A. Du vosel, S. Spera, *J. Chem. Soc., Perkin Trans. 2*, (1990) 2075–2080.
[28] Y. Kikuchi, K. Kobayashi, Y. Aoyama, *J. Am. Chem. Soc.*, 114 (1992) 1351–1358.
[29] J.D. Lamb, J.J. Christensen, J.L. Oscarsson, B.L. Nielsen, B.W. Asay, R.M. Izatt, *J. Am. Chem. Soc.*, 102 (1980) 6820–6824.
[30] Y. Kikuchi, Y. Kato, Y. Tanaka, H. Toi, Y. Aoyama, *J. Am. Chem. Soc.*, 113 (1991) 1349–1354.
[31] K. Kobayashi, Y. Asakawa, Y. Kato, Y. Aoyama, *J. Am. Chem. Soc.*, 114 (1992) 10307–10313.
[32] O. Mayzel, O. Aleksiuk, F. Grynszpan, S.E. Biali, Y. Cohen, *J. Chem. Soc., Chem. Commun.*, (1995) 1183–1184.