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Communications to the Editor

Anion Carrier Mediated Membrane Transport of Phosphate: Selectivity of $H_2PO_4^-$ over Cl^-

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Since the first report by Reusch and Cussler[1] in 1973, numerous papers have appeared on the selective transport of salts through supported liquid membranes (SLMs) mediated by neutral cation carriers. In transport assisted by neutral cation carriers such as simple crown ethers, calixarenes, or natural ionophores like valinomycin, anions affect the transport rates because of the different dehydration energies. Therefore, lipophilic anions like ClO_4^-, NO_3^-, or SCN^- are often used as the counterion. Much less lipophilic anions like Cl^- or $H_2PO_4^-$ have only been transported by *charged* anion carriers (ACs) via an ion-exchange mechanism,[2-4] and the transport selectivity follows the Hofmeister series:[5] $ClO_4^- > I^- > SCN^- > NO_3^- > Br^- > Cl^- \gg CO_3^{2-}, H_2PO_4^-, SO_4^{2-}$.

Recently, we have designed neutral receptors which preferentially bind monovalent phosphate ($H_2PO_4^-$) anions in organic solvents by the combined action of Lewis and Brønsted acidic sites.[6] In this communication we report the *selective transport of $H_2PO_4^-$ over Cl^-* through an SLM, either exclusively by an anion receptor or by a combination of anion and cation receptors. The receptors exhibit selectivity opposite to the order of dehydration energies of the anions in the Hofmeister series. To the best of our knowledge, this is the first report on the phosphate anion cotransport by *neutral* anion receptors.[7]

The anion carriers **1**−**3** (Chart 1) were synthesized by reacting the appropriate ω-halide functionalized amide moieties with 2-(allyloxy)-3-hydroxybenzaldehyde, followed by deallylation and reaction with 1,2-*cis*-cyclohexanediamine.[6]

They were used in an SLM with *o*-nitrophenyl *n*-octyl ether (NPOE) as the membrane solvent immobilized in an Accurel 1E-PP support. The flux (*J*) of KH_2PO_4 was measured from buffered (pH 6.8) aqueous source phase solutions.[8]

The flux of KH_2PO_4 was measured with anion carrier concentrations in the SLM of 0.02 M (Table 1). Receptors **1** and **2** are already effective on their own as $H_2PO_4^-$ carriers in the transport of KH_2PO_4, although the fluxes are low. Surprisingly, despite strong $H_2PO_4^-$ binding detected for **3** by ^{1}H NMR (DMSO) and conductometry (MeCN:DMSO = 99:1),[6] transport of KH_2PO_4 through NPOE could not be detected.

Chart 1

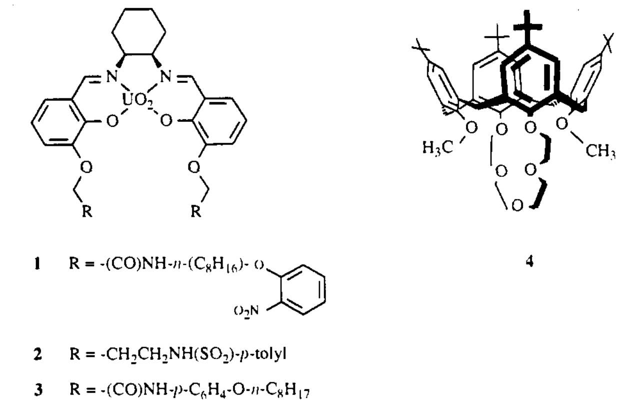

Table 1. Effect of the Combination of Anion Carriers and Cation Carrier **4** on KH_2PO_4 Flux

carrier(s)[b]	$J(KH_2PO_4)$[a] (10^{-8} mol m^{-2} s^{-1})	carrier(s)[b]	$J(KH_2PO_4)$[a] (10^{-8} mol m^{-2} s^{-1})
1	3.2	**1 + 4**	5.0
2	5.1	**2 + 4**	12.5
3	<0.2	**3 + 4**	7.2
4	0.6		

[a] $[KH_2PO_4]_s + [K_2HPO_4]_s = 0.2$ M; $pH_s = 6.8$. [b] [AC] = [CC] = 0.02 M.

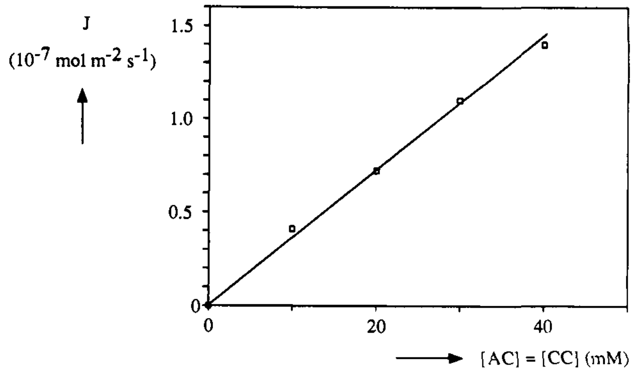

Figure 1. KH_2PO_4 flux mediated by the combination of carriers **3** and **4** as a function of the carrier concentration. [AC]/[CC] = 1; $[KH_2PO_4]_s + [K_2HPO_4]_s = 0.2$ M; $pH_s = 6.8$.

To improve the flux, a K^+ selective cation carrier, calix[4]-arene crown-5 (**4**)[9] (Chart 1), was added to the membrane solution (0.02 M) for the simultaneous facilitation of the potassium ion transport (Table 1).

The flux of KH_2PO_4 mediated by the combination of carriers **2** and **4** is the highest in the series (12.5×10^{-8} mol m^{-2} s^{-1}) and higher than obtained with any of the anion carriers alone at [AC] = 0.02 M.

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(1) Reusch, C. F.; Cussler, E. L. *AIChE J.* **1973**, *19*, 736−741.

(2) Molnar, W. J.; Wang, C. P.; Fennel-Evans, D.; Cussler, E. L. *J. Membr. Sci.* **1978**, *4*, 129−140.

(3) (a) Neplenbroek, A. M.; Bargeman, D.; Smolders, C. A. *J. Membr. Sci.* **1992**, *67*, 107−119; (b) **1992**, *67*, 121−132.

(4) Kokufuta, E.; Nobusawa, N. *J. Membr. Sci.* **1990**, *48*, 141−154.

(5) Wegmann, D.; Weiss, H.; Amman, D.; Morf, W. E.; Pretsch, E.; Sugahara, K.; Simon, W. *Mikrochim. Acta* **1984**, *3*, 1.

(6) Synthesis of **3**: Rudkevich, D. M.; Verboom, W.; Brzózka, Z.; Palys, M. J.; Stauthamer, W. P. R. V.; van Hummel, G. J.; Franken, S. M.; Harkema, S.; Engbersen, J. F. J.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **1994**, *116*, 4341−4351. Experimental details of **1** and **2** will be published later.

(7) Recently, nucleoside phosphates have been transported across bulk liquid membranes by, e.g., porphyrin and Kemp triacid derivatives; see, for example: Král, V.; Sessler, J. L.; Furuta, H. *J. Am. Chem. Soc.* **1992**, *114*, 8704−8705. Andreu, C.; Galán, A.; Kobiro, K.; de Mendoza, J.; Park, T. K.; Rebek, J., Jr.; Salmerón, A.; Usman, N. *J. Am. Chem. Soc.* **1994**, *116*, 5501−5502 and references therein.

(8) The KH_2PO_4 flux was measured from a source phase of neutral pH (0.1 M KH_2PO_4 and 0.1 M K_2HPO_4, pH = 6.8). Only KH_2PO_4 will be transported because of the stronger binding and much higher lipophilicity of $H_2PO_4^-$. The transport measurements were carried out at least in duplicate at 25 °C in a permeation cell described elsewhere.[12] Aqueous salt solutions were used as the source phase and doubly distilled and deionized water as the receiving phase. Initial steady state transport was measured up to receiving phase salt concentrations of $\leq 1 \times 10^{-4}$ M. The transport of salt was determined by monitoring the conductivity of the receiving phase as a function of time except for competitive transport experiments (see Table 2). The standard deviation in the transport measurements with anion (and cation) carriers is about 20%.

(9) Ghidini, E.; Ugozzoli, F.; Ungaro, R.; Harkema, S.; El-Fadl, A. A.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **1990**, *112*, 6979−6985 and references therein.

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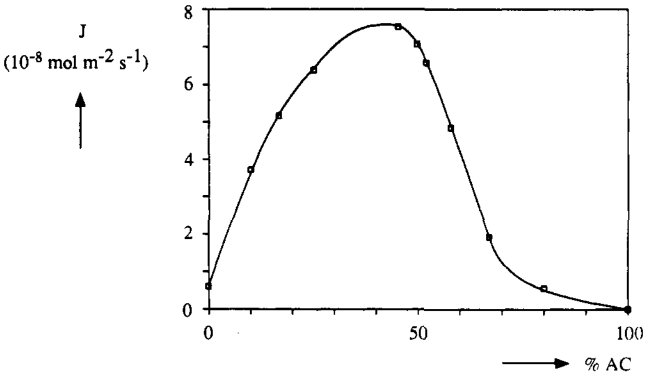

Figure 2. KH_2PO_4 transport mediated by the combinations of carriers **3** and **4** as a function of the percentage of anion carrier. [AC] + [CC] = 0.04 M; $[KH_2PO_4]_s + [K_2HPO_4]_s = 0.2$ M; $pH_s = 6.8$.

Table 2. Transport Selectivity for Dihydrogen Phosphate over Chloride

AC[a]	$J(H_2PO_4^-)$[b] (10^{-8} mol m^{-2} s^{-1})	$J(Cl^-)$[c] (10^{-8} mol m^{-2} s^{-1})	S[d]
1	1.81	4.75	38
2	7.52	5.25	143
3	2.19	7.60	29

[a] [**4**] = [AC] = 2×10^{-2} M. [b] $[KH_2PO_4]_s = 1 \times 10^{-3}$ M, no K_2HPO_4 added. [c] $[KCl]_s = 1 \times 10^{-1}$ M. [d] The fluxes of dihydrogen phosphate and chloride were determined by titrating samples from the receiving phase. Chloride was determined by addition of 5×10^{-4} M $Hg(NO_3)_2$ (for low concentrations of the anion) or 1×10^{-1} M $AgNO_3$ and total phosphate by addition of 1×10^{-2} M HCl followed by titration with 1×10^{-2} M tetrabutylammonium hydroxide.

The absolute concentrations of the 1:1 combination of carriers, **3** and **4** have also been varied (Figure 1). A steady increase of the flux is observed up to carrier concentrations of [**3**] = [**4**] = 4×10^{-2} M. The fact that receptor **3** alone was *not* effective as a carrier for KH_2PO_4, but only in combination with **4**, prompted us to measure the KH_2PO_4 flux as a function of their concentration ratio [**3**]/[**4**] (Figure 2). The optimum flux is reached at about equal concentrations of **3** and **4**.

Finally, we have investigated the transport selectivity for $H_2PO_4^-$ over Cl^- in competition experiments from a source phase that contained 1×10^{-3} M of KH_2PO_4 and 1×10^{-1} M of KCl by the combination of anion receptors **1**–**3** and carrier **4** (Table 2).[10] The selectivity, *S*, is defined as the ratio of the fluxes divided by the ratio of source phase anion concentrations. All anion carriers show transport selectivity for H_2PO_4 over Cl^- which is opposite to the dehydration energy according to the Hofmeister series. The transport selectivity for phosphate over chloride increases as a function of the anion receptor in the order **3** < **1** < **2** up to a value of about 140.

In nature, orthophosphate anions can be specifically transported into cells and organelles by the phosphate binding protein.[11] We have demonstrated that synthetic phosphate receptors are effective on their own or in combination with a cation receptor to transport KH_2PO_4 across a lipophilic membrane.

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(10) In this case, no K_2HPO_4 was added, as was the case in the single ion $H_2PO_4^-$ transport experiments (see ref 8).

(11) Leucke, H.; Quiocho, F. A. *Nature* **1990**, *347*, 402–406.

(12) Stolwijk, T. B.; Sudhölter, E. J. R.; Reinhoudt, D. N. *J. Am. Chem. Soc.* **1987**, *109*, 7042–7047.