CARRIER ANTIBIOTICS AND MODEL COMPOUNDS AS COMPONENTS OF SELECTIVE ION-SENSITIVE ELECTRODES

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ABSTRACT

The ion selectivity exhibited by different types of membrane electrodes is rationalized by a theoretical treatment of the membrane potential. An equation, which is admirably suited for the interpolation of e.m.f.-values, is derived for the electrode response of membrane electrodes of the electrically neutral ligand type (antibiotics, model compounds) to mixed solutions of monovalent and divalent cations. In contrast to the classical Nicolsky equation it describes the contribution of interfering ions to the e.m.f. by an activity-independent selectivity term.

Electrically neutral ligands selective for calcium ions to be used as components for liquid membrane electrodes were designed and prepared. The corresponding electrodes are in all respects superior to the calcium ion liquid membrane electrodes described so far.

In the past decade, a great number of selective ion-sensitive electrodes have been designed and have found application in all branches of routine analysis[1-4]. Usually, a membrane electrode assembly (shown schematically in *Figure 1*) is employed. The e.m.f. of this electrochemical cell is related to the ion activities in the sample solution by the extended Nicolsky equation

$$E = E_0 + \frac{RT}{z_iF} \ln \left[a_i + \sum_{j \neq i} K_{ij}^{\text{Pot}} (a_j)^{z_i/z_j} \right] \tag{1}$$

E: cell potential (e.m.f.)
E_0: constant reference potential
a_i: activity of a primary ion I^{z_i} in the sample solution
a_j: activity of an interfering ion J^{z_j} in the sample solution
K_{ij}^{Pot}: selectivity factor, characteristic of a given membrane
RT/F: Nernst factor

The theoretical treatment of the membrane potential† has led to an understanding of the ion specificity exhibited by different electrode types, the

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† The so-called membrane potential is defined as the electrical potential difference between the two outside solutions (*Figure 1*) and may be broken down into a boundary potential and a diffusion potential, i.e. internal membrane potential. For all practical purposes, the membrane potential can be regarded as the only activity-dependent contribution to the cell potential.
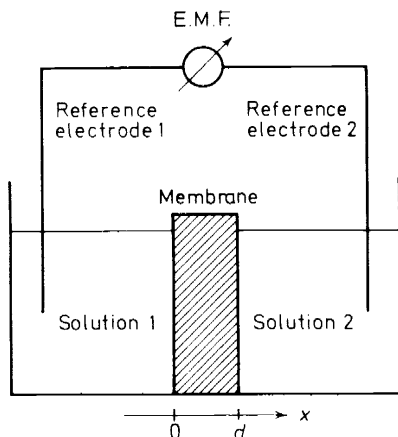
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Figure 1. Schematic representation of a membrane electrode assembly. In practical applications only the composition of solution 1 (aqueous sample solution) is varied. The half-cell comprising membrane, solution 2 (internal filling solution), and reference electrode 2 (internal reference electrode) remains unchanged and constitutes the membrane electrode.

derivable selectivity factors K_{ij}^{Pot} being in perfect agreement with experimental data[5-9]. Using the model assumptions given in *Table 1* and evaluating the

Table 1. Model assumptions necessary to derive expressions corresponding to equation 1[5]

i	The same solvent is used for the electrolyte solutions on either side of the membrane.
ii	A thermodynamic equilibrium exists between the membrane and each of the outside solutions at the respective phase boundaries.
iii	Every cell component is homogeneous with respect to a direction perpendicular to the cell axis; therefore, concentration gradients and the concomitant potential differences are possible only along this cell axis (x-coordinate, *Figure 1*). There is no pressure gradient.
iv	The membrane phase approximates ideal behaviour, i.e. the activity of any component is equal to its concentration.
v	Within the membrane, the chemical standard potentials and the mobilities of all particles present are invariant with space and time.
vi	The system is in a zero current steady-state.
vii	There is no convection current across the membrane.

Assumptions i to vii are invoked simultaneously; in addition, one of the assumptions (a) to (c) holds:

(a)	Within the membrane, the individual mobilities and/or concentrations of either all anions or all cations are negligible. All the particles whose fluxes give rise to a diffusion potential are of the same charge z.
(b)	Throughout the membrane, the concentration gradients are negligible for all ionic species not of charge z. The electroneutrality condition holds.
(c)	Within the membrane, the concentration gradients and/or the mobilities are negligible for all ions, i.e. the diffusion potential is nearly zero. Furthermore, the total concentration of all forms of cations respectively anions is constant.

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e.m.f.-response to a primary ion and an interfering species of the same charge z according to

$$E = E_0 + \frac{RT}{zF} \ln \left[a_i + K_{ij}^{Pot} a_j \right] \tag{1a}$$

the expressions for K_{ij}^{Pot} compiled in *Table 2* are obtained.

As can be gathered from *Table 2*, theory predicts for a solid-state electrode with, for example, a silver halide type of membrane a correlation between the selectivity factors and the solubility products. *Figure 2* (see also References
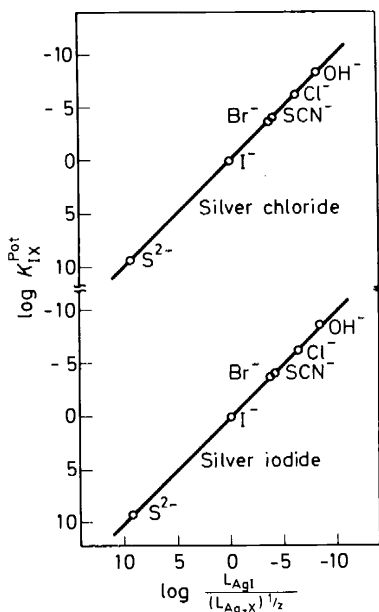

Figure 2. Correlation between the selectivity factors and the solubility products for silver halide solid-state membrane electrodes[12].

1–4, 10–12) confirms that the solubility equilibria are, indeed, responsible for the ion-selective behaviour of solid-state membrane electrodes.

For a liquid membrane composed of a water-immiscible solvent of sufficiently high dielectric constant, lipophilic anions S^-, and counterions as strongly dissociated solute species, the electrochemical selectivity between cations of the same charge is given by (*Table 2*)

$$K_{ij}^{Pot} \approx \frac{k_j}{k_i} = A_{ij} \tag{2}$$

The ratio of the partition coefficients, A_{ij}, represents the exchange equilibrium constant for the reaction

$$I^{z+} \text{ (membrane)} + J^{z+} \text{ (solution)} \rightleftharpoons J^{z+} \text{ (membrane)} + I^{z+} \text{ (solution)} \quad (3)$$
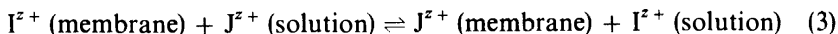
and is, therefore, independent of the chemical properties of any membrane components other than the solvent. It is known that a great number of organic solvents exhibit a selectivity for large ions. Thus, a useful sensor for large organic onium-ions has been designed using 2-nitro-p-cymene as selective membrane solvent and tetraphenylborate as cation-exchange sites[13]. *Figure 3*, according to equation 2 ,shows a satisfactory agreement of
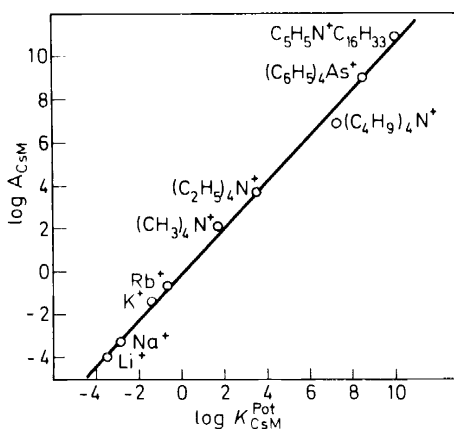

Figure 3. Correlation between the selectivity factors and the ion-exchange equilibrium constants for a liquid membrane electrode based on the solvent 2-nitro-p-cymene[13].

the selectivity factors K_{ij}^{Pot} of this sensor with the exchange constants A_{ij}, as obtained from salt extraction data[13]. A similar correlation between selectivity factors and extraction parameters is also to be expected for associated ion exchangers if the association between counterion and ion-exchange site has little influence on the ion selectivity of the system, i.e. the lipophilic exchanger ion exhibits no efficient ion specificity. Such a behaviour has been reported for anion-sensitive electrodes[14–16] which are based on a membrane of decanol containing tetraalkylammonium ions as anion-exchange sites.

A more complex behaviour is to be expected for liquid membrane electrodes incorporating negatively charged ligands S^- capable of selective complexation of cations. As is shown in *Table 2* for this type of membrane, two completely different expressions for the selectivity between corresponding ions can be derived ,depending on which assumptions are made

$$K_{ij}^{Pot\ 1} \approx \frac{k_j}{k_i} \quad (4)$$

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Table 2. Expressions for the selectivity between a primary ion I^z and an interfering ion J^z for different types of membrane electrodes[5].

Type of membrane	Species within the membrane	Selectivity factor $K_{ij}^{Pot} = 1/K_{ji}^{Pot}$	Limiting case (see *Table 1*)	References
Liquid membrane with lipophilic ions S⁻ (dissociated cation-exchanger)	I^{z+}, J^{z+}, S^-	$\dfrac{u_j k_j}{u_i k_i}$	(a)	6, 7
		$\dfrac{(zu_j + u_s) k_j}{(zu_i + u_s) k_i}$	(b)	
		k_j/k_i	(c)	
Liquid membrane with lipophilic electrically charged ligands S⁻ (associated cation-exchanger)	IS_z, JS_z (I^{z+}, J^{z+}, S^-)	$\dfrac{u_j k_j}{u_i k_i}$	(a) $[u_s \ll u_i, u_j]$	6, 7
		$\dfrac{(zu_j + u_s) k_j}{(zu_i + u_s) k_i}$	(b)	
		$\dfrac{u_{js} K_j}{u_{is} K_i}$	(a) $[u_s \gg u_i, u_j]$ total flux of all ligand forms negligible	6, 7
		$\dfrac{K_j}{K_i}$	(c) $c_s(x) = const.$	
		cf. equation 9	(c) electroneutrality holds	
Liquid membrane with lipophilic electrically neutral ligands S (complex forming membrane component)	$IS_n^{z+}, JS_n^{z+}, X^-$ (I^{z+}, J^{z+}, S)	$\dfrac{u_{js} K_j}{u_{is} K_i}$	(a) $c_s(x) = const.$	7, 8
		$\dfrac{(zu_{js} + u_x) K_j}{(zu_{is} + u_x) K_i}$	(b) $c_s(x) = const.$	
		$\dfrac{K_j}{K_i}$	(c) $c_s(x) = const.$	
Solid-state membrane of the silver halide type	$M^+, I^{|z|-}, J^{|z|-}$	$\dfrac{L_{mi}}{L_{mj}}$	(c)	9, 10

k: partition coefficients of cations between outside solution and membrane
K: partition coefficients of cations between outside solution and the respective complexes in the membrane
L_{mi}, L_{mj}: solubility products of the pure silver halides
u: mobilities in the membrane
$c(x)$: concentrations in the membrane
z: ionic charge

and

$$K_{ij}^{Pot\,2} \approx \frac{K_j}{K_i} \qquad (5)$$

Whereas the first expression again represents the selectivity as a function of the membrane solvent alone, the second relation, on the other hand, de-

scribes the ion-selective behaviour of the ligand. Thus, the ratio K_j/K_i is equal to the equilibrium constant for the ion-exchange reaction

$$JS_z \text{ (membrane)} + J^{z+} \text{ (solution)} \rightleftharpoons JS_z \text{ (membrane)} + I^{z+} \text{ (solution)} \quad (6)$$
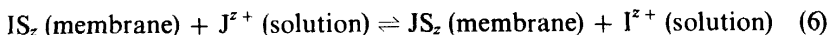
It follows:

$$\frac{K_j}{K_i} = \frac{K_{js}\,k_j}{K_{is}\,k_i} \approx \frac{K_{js}^*}{K_{is}^*} \quad (7)$$

where K_{is} and K_{js} are the stability constants of the respective complexes in the membrane, K_{is}^* and K_{js}^* are the membrane-independent complex stability constants in the outside solution.

In practice, however, neither of the limiting cases as defined by equations 4 and 5 is realistic (cf. stipulations in *Table 2*). A rather reasonable approach is to formulate the electrode response as follows

$$E = E_0 + \frac{RT}{(z+1)F} \ln\left[a_i + K_{ij}^{\text{Pot 1}}\,a_j\right] + \frac{RT}{z(z+1)F} \ln\left[a_i + K_{ij}^{\text{Pot 2}}\,a_j\right] \quad (8)$$

This equation can be derived from theory when the diffusion potential is assumed to be negligible (cf. assumption (c) in *Table 1*) and the electroneutrality condition is valid†. Starting from equation 8, the usual selectivity factor defined by the Nicolsky equation (1a) is found to be

$$K_{ij}^{\text{Pot}} = \left(K_{ij}^{\text{Pot 1}} + \frac{a_i}{a_j}\right)^{\{z/(z+1)\}} \left(K_{ij}^{\text{Pot 2}} + \frac{a_i}{a_j}\right)^{\{1/(z+1)\}} - \frac{a_i}{a_j} \quad (9)$$

With this it is proved that the electrochemical selectivity between cations of the same charge displayed by liquid membranes with charged ligands is a function of the activities in the sample solution. Furthermore, it is evident that a sufficiently high discrimination of interfering ions can be achieved only if both membrane components, solvent and ligand, exhibit a comparable ion specificity. Thus, for example, liquid membranes consisting of a solution of the potassium-selective carrier antibiotic nigericin‡ (ratio of the complex stability constants $K_{KS}^*/K_{NaS}^* \approx 50$[17]) or the sodium-selective antibiotic monensin‡ ($K_{NaS}^*/K_{KS}^* \approx 25$[17]) in decanol ($A_{NaK} \approx 2$) are not suited for the preparation of sensors for the respective ions. In agreement with experimental results[19], the selectivity of these systems come out to be rather poor: nigericin in decanol: $K_{KNa}^{\text{Pot}} \sim 0.1 - 0.25$ and monensin in decanol: $K_{NaK}^{\text{Pot}} \sim 0.3 - 1$.

As a consequence of the expected activity dependence of the selectivity factors, especially for liquid ion-exchange membranes, the unqualified use of the Nicolsky equation in assessing the electrode response to mixed electrolyte solutions can be misleading. A serious difficulty arises if the electrode

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† For monovalent ions, a more general description is available owing to the theoretical work of Sandblom, Eisenman and Walker[6].

‡ The carrier antibiotics of the so-called nigericin group have a dissociated carboxylate group at physiological pH and, therefore, form electrically neutral 1:1 complexes with alkali metal ions[17, 18].

potential is produced by a mixture of ions of different charge; for instance, for a divalent cation I^{2+} and a monovalent cation J^+, both of which are in equilibrium with the respective lipid-soluble complexes IS_2 and JS at the membrane surface, the fundamental equation is

$$IS_2 \text{ (membrane)} + 2J^+ \text{ (solution)} \rightleftharpoons 2JS \text{ (membrane)} + I^{2+} \text{ (solution)} \quad (10)$$
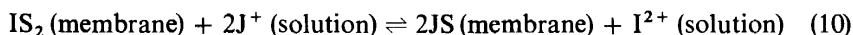
Since the differentiation between a primary ion and an interfering ion is arbitrary in this case, two inconsistent expressions of the Nicolsky type (1) are admissible:

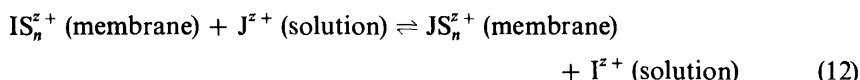$$E = E_0 + \frac{RT}{2F} \ln \left[a_i + K_{ij}^{Pot} a_j^2 \right] \quad (1b)$$

and

$$E = E_0' + \frac{RT}{F} \ln \left[a_j + K_{ji}^{Pot} a_i^{\frac{1}{2}} \right] \quad (1c)$$

It is obvious that the weighting factors K_{ij}^{Pot} and K_{ji}^{Pot}, both characteristic of the ion selectivity, must again be to a large degree activity-dependent. A detailed theoretical study of the monovalent–divalent ion selectivity demonstrates that the response of liquid membrane electrodes incorporating charged ligands can generally not be reproduced by any explicit expression containing only sample solution-activities and constant selectivity parameters. It can be shown, however, that equation 1b constitutes a reasonable approximation for liquid membrane electrodes highly responsive to monovalent cations, whereas equation 1c more successfully delineates the behaviour of liquid membrane electrodes highly sensitive to divalent cations[20]. In practice, the alternative representation is commonly preferred; hence one has to cope with large variations in the selectivity factors.

In striking contrast to liquid membrane electrodes with negatively charged complexing agents, the behaviour of membranes based upon electrically neutral ligands S is fully resolved by theory. As can be gathered from *Table 2*, the selectivity of neutral ligand electrodes between cations of the same charge is for every limiting case listed determined as

$$K_{ij}^{Pot} \approx \frac{K_j}{K_i} \quad (11)$$

Again, the ratio K_j/K_i is identical to the equilibrium constant for the exchange reaction:

$$IS_n^{z+} \text{ (membrane)} + J^{z+} \text{ (solution)} \rightleftharpoons JS_n^{z+} \text{ (membrane)}$$
$$+ I^{z+} \text{ (solution)} \quad (12)$$

and is found to be:

$$\frac{K_j}{K_i} = \frac{K_{js} k_j}{K_{is} k_i} = \frac{K_{js}^* k_{js}}{K_{is}^* k_{is}} \quad (13)$$

where K_{is}, K_{js} and K_{is}^*, K_{js}^* are the complex stability constants in the membrane and in the outside solution respectively, and k_{is}, k_{js} are the partition

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coefficients of the positively charged complexes between the outside solution and the membrane. Equation 13 suggests that extremely high selectivities can be achieved by using neutral ion-specific ligands as membrane components. Suitable lipophilic ligands have been available since the discovery of highly selective carrier antibiotics, such as valinomycin or the macrotetrolides†, and of related synthetic ligands, for example crown compounds. (For a review see Reference 18).

In the case of carrier antibiotics that are capable of enveloping monovalent cations completely and evenly[18], the complex mobilities and partition coefficients are entirely independent of the nature of the central ion. According to *Table 2* and equation 13, the alkali ion selectivity of the respective liquid membranes is then determined solely by the complexing properties of the carrier ligands[5, 7, 8]

$$K_{ij}^{Pot} = \frac{K_{js}^*}{K_{is}^*} \tag{14}$$

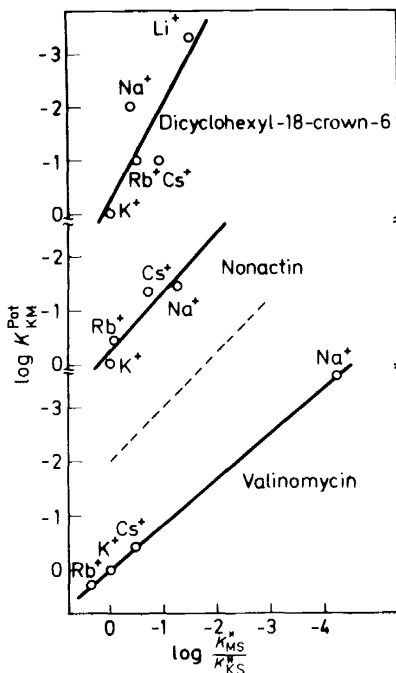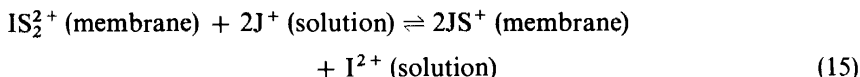*Figure 4* demonstrates impressively that e.m.f.-measurements are a welcome

Figure 4. Correlation between the selectivity factors and the complex stability constants for liquid membrane electrodes based on neutral carrier ligands[5] (see also references 21, 22).

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† In contrast to the carrier antibiotics of the nigericin group, the representatives of the valinomycin group behave as electrically neutral ligands.

method for directly assessing the relative stabilities of the complexes of the valinomycin group antibiotics in the outside solution, i.e. in water and water-like solvents. The selectivity factors of the liquid membranes are in this case independent of the membrane solvent used[21].

If, however, the ligands used as membrane components are not able to completely displace the solvent shell of the cation, as is the case with many of the synthetic crown compounds, equation 14 is not valid, since now the membrane solvent has a remarkable influence[23] on the ion-exchange reaction, equation 12. The rather poor correlation between the selectivity factors and the ratios of complex formation constants found for crown compounds (see *Figure 4* and Reference 22) is therefore not surprising.

As is demonstrated above, it is possible to successfully rationalize the selectivity of membrane electrodes between ions of the same charge if the diffusion potential, i.e. the internal membrane potential, is assumed to be negligible (see *Table 2*). Under such conditions, a theoretical treatment of the monovalent–divalent ion selectivity of liquid membrane electrodes based on neutral ligands can be carried out[24]. In analogy to equation 10, the following ion-exchange equilibrium is assumed to determine the electrode response

$$IS_2^{2+} \text{ (membrane)} + 2J^+ \text{ (solution)} \rightleftharpoons 2JS^+ \text{ (membrane)}$$

$$+ I^{2+} \text{ (solution)} \tag{15}$$

It is helpful to define the equilibrium constant for reaction 15 as

$$K_{ij}^M = \frac{a_i\, x_j^2}{a_j^2\, x_i} \tag{16}$$

where x signifies the fraction of the complexed ligand concentration at the membrane surface, c^{tot}, that is coordinated to one cation species. The constant K_{ij}^M is related to the complex stability constants and partition coefficients as follows

$$K_{ij}^M = \frac{K_{js}^{*2}\, k_{js}^2}{2K_{is}^*\, k_{is} c^{tot}} \tag{17}$$

As presented elsewhere[24], an appropriate expression for the electrode response to a mixed solution of I^{2+} and J^+ can be derived

$$E = E_0 + \frac{RT}{F} \ln \left[(a_i + \tfrac{1}{4} K_{ij}^M\, a_j^2)^{\frac{1}{2}} + (\tfrac{1}{4} K_{ij}^M\, a_j^2)^{\frac{1}{2}}\right] \tag{18}$$

The superiority of this formulation over either of the commonly employed Nicolsky equations 1b and 1c will be impressively substantiated below.

As shown in the previous treatment, the electrically neutral antibiotics of the valinomycin group[25] and related lipid-soluble ligands are predestined to be incorporated as the active components in highly selective liquid membrane electrodes because the remarkable specificity they exhibit in their complexation behaviour towards cations can be fully exploited in this manner. The analytical potential of these ion-selective sensors has been recognized and forms incorporating electrically neutral ligands specific for

K^+, NH_4^+, Ba^{2+} and Ca^{2+} have found acceptance in practical applications[18, 26].

At present the roster of suitable carrier molecules is limited to a small number of natural and synthetic compounds. The results of the model calculations described earlier[27] establish a framework of most likely desirable characteristics, from which the synthesis of new neutral ligands can proceed. According to these calculations the requirements governing the suitability of electrically neutral ligands as carriers for alkali or alkaline-earth metal ions including ammonium ions in membranes are[27]

(a) A carrier molecule should be composed of polar and nonpolar groups.
(b) Among the polar groups there should be preferably 5 to 8, but not more than 12 coordinating sites such as oxygen atoms.
(c) Ideally, all cations should be forced into accepting the same given number of coordinating groups.
(d) The carrier should be able to assume a stable conformation that provides a cavity, surrounded by the polar groups, suitable for the uptake of cations, while the nonpolar groups form a lipophilic shell around the coordination sphere.
(e) High selectivities are achieved by locking the coordinating sites into a rigid arrangement around the cavity. Such rigidity can be enhanced by the presence of bridged structures or hydrogen bonds. Within one group of the periodic system, the cation that best fits into the offered cavity is preferred.
(f) Notwithstanding requirement (e), the ligand should be flexible enough to allow a sufficiently fast exchange. This is possible only with a stepwise substitution of the solvent molecules by the ligand groups. Thus, a compromise between stability (e) and exchange rate (f) has to be found.
(g) The overall dimensions of a carrier should be rather small but consistent with high lipid solubility.
(h) Electrically neutral Li^+- or Na^+-specific carriers should contain no more than 6 coordination sites.
(i) Electrically neutral ligands designed to be specific for large alkaline-earth cations must be as small as possible and should be used in membrane solvents of high dielectric constant. A neutral carrier specific for Mg^{2+} seems unlikely.

Ligands were designed[28, 29] which, in accordance with the model set forth above, should exhibit a preference for divalent cations (see *Figure 5*). The ligand most thoroughly studied, and the first of this series to be synthesized, contains six potential coordination centres, namely two etheric oxygens as well as the carbonyl oxygens of two amide and two ester groups (ligand I). Four of these coordination sites are separated by three segments of two atoms each, thereby predetermining five-ring chelates (optimal size of chelate rings[30]). The high flexibility provided by the acyclic structure ensures a step-wise substitution of the solvate shell of the cation by ligand coordination groups. Thus, the rapid complexation and decomplexation necessary for the incorporation in sensors is given[31]. The coordination sites are interconnected by an aliphatic skeleton. It is intended that the small ligand-layer thickness achieved herewith shall lead to a preference of divalent

I R^1 $: - CH_3$

   R^2 $: - (CH_2)_{10} - COO - CH_2 - CH_3$

II R^1 $: - CH_3$

   R^2 $: -(CH_2)_6 - CH_3$

III $R^1 = R^2$ $: - CH_2 - CH_2 - CH_3$

IV R^1 $: - CH_2 - CH_2 - CH_3$

   R^2 $: - CH_2 - C-(CH_3)_3$

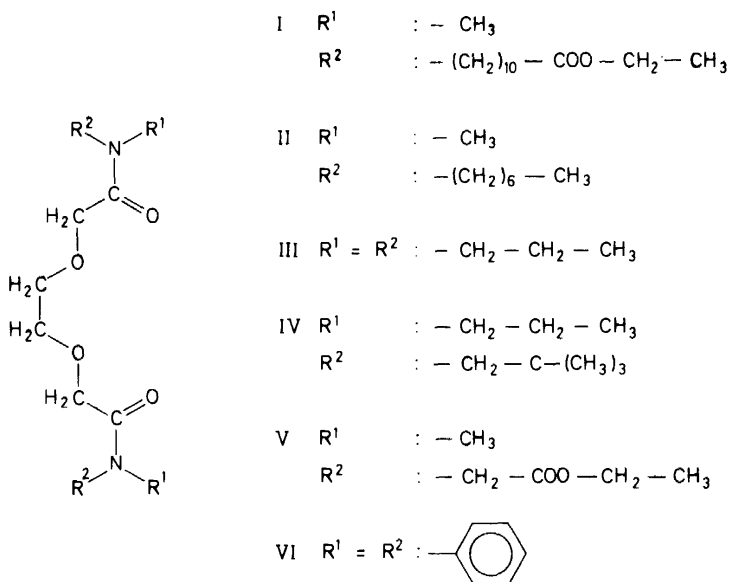V R^1 $: - CH_3$

   R^2 $: - CH_2 - COO - CH_2 - CH_3$

VI $R^1 = R^2$:

Figure 5. Structures of some synthetic carriers specific for alkaline-earth metal ions.

over monovalent cations. Compound I, in fact, does show[28, 32] a high Ca^{2+}-selectivity (*Table 3*). The properties of this calcium sensor are discussed below.

Providing that there is a large excess of calcium ions a 1:1 complex exists in methanol. More recent investigations employing ^{13}C-n.m.r.-spectroscopy[33] indicate that the esteric carbonyl groups do not participate as co-ordination sites. If the Ca^{2+}-induced chemical shifts are plotted against the Ca^{2+}:ligand concentration ratio, the resonance frequencies of a number of carbon atoms are shifted by amounts that attain a maximum and a minimum

Table 3. Electrochemical selectivity behaviour of the synthetic ligands I–VI presented in *Figure 5*. The e.m.f.-responses of the respective sensors (PVC matrix) to 10^{-12} M solutions of different cations are given (referred to Ca^{2+}).

Cation M^{z+}	$E_M - E_{Ca}$, mV for ligand					
	I	II	III	IV	V	VI
Mg^{2+}	-110	-112	-101	-111	-95	-81
Ca^{2+}	0	0	0	0	0	0
Ba^{2+}	-1	-3	-40	-50	-65	$+73$
Na^+	-73	-77	-91	-103	-92	$+13$
K^+	-82	-86	-82	-75	-66	$+3$
Cs^+	-103	-109	-103	-107	-89	-34

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respectively for a concentration ratio of about 0.5. Accordingly, the stoichiometry of the complexes should be different for small (probably 1 :2) and for large metal concentrations (1 :1, four coordination sites occupied by the ligand, all others by methanol).

Ligand II does not contain the terminal ester groups. Since this ligand behaves quite similarly to ligand I in sensor systems (see *Table 3*), these ester functions surely do not contribute to the high Ca^{2+}-selectivity. Both ligands probably form 1 :2 complexes with Ca^{2+} in the sensor. Investigations carried out with compounds III and IV (*cf. Table 3*) indicate that the *N*-alkyl groups are of minor importance as concerns the selectivity behaviour of the ligands.

The similarity in ion-selective behaviour of ligands I–IV and ligand V (*Table 3*) gives rise to the assumption that it is the same coordination sites in V as in I–IV that are actively involved in complexation. It is, indeed, deducible from ^{13}C-n.m.r.-spectra that the esteric carbonyl groups do not participate in complexation[33].

The substitution of the *N*-alkyl groups by *N*-phenyl groups (ligand VI) leads to a preference for larger ions ($Ba^{2+} > Ca^{2+}$, cf. *Table 3*). This can be rationalized by assuming that the steric interactions among the phenyl groups result in a larger cavity between the coordination sites. The introduction of phenyl groups also degrades the discrimination of monovalent cations. A possible explanation is the marked swelling of the ligand layer. On the other hand, it should be kept in mind, that a decrease of the electron density at the carbonyl oxygens invariably produces the same effect.

Recently, and in accordance with the concepts set forth above, a crystalline
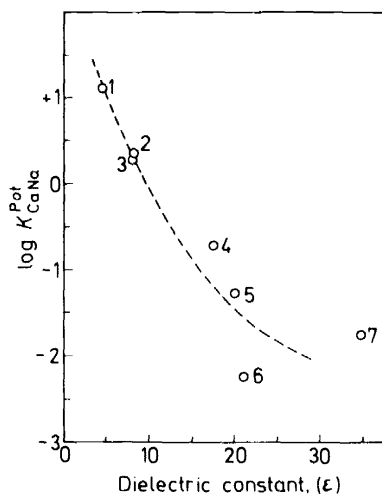

Figure 6. Selectivity for Na^+ relative to Ca^{2+} of a liquid membrane electrode (filter paper matrix) based on ligand I as a function of the dielectric constant of the membrane solvent. Membrane solvents: (1) dibutylsebacate, (2) tris-(2-ethylhexyl)-phosphate, (3) 1-decanol, (4) acetophenone, (5) 2-nitro-p-cymene, (6) *p*-nitroethylbenzene, (7) nitrobenzene. The selectivity factors K_{CaNa}^{Pot} were obtained from e.m.f.-measurements using 10^{-1} M calcium chloride and 10^{-1} M sodium chloride respectively as sample solutions.

Ba^{2+}-complex of this ligand was obtained that an elemental analysis showed to be of 1 :2 stoichiometry.

In the course of the development of a useful alkaline-earth ion specific sensor based on electrically neutral ligands, such as those presented above, particular attention is to be paid to the selection of a suitable water-immiscible membrane solvent. As has been realized only recently[32], the dielectric constant of the chosen membrane solvent is not a matter to be casually overlooked. Theoretical studies indicate that the higher it is, the better should monovalent cations be discriminated by the neutral ligands incorporated in the sensor. This prediction is handily confirmed by experimental evidence presented in *Figure 6*. Furthermore, the lifetime and the stability of sensor systems can be greatly modified through variation of the membrane matrix[1].

An electrode incorporating the calcium-selective ligand I has been developed[34] that is superior to a previously described electrode[32] in terms of lifetime and stability, mainly because of the PVC-matrix[1] employed. The membrane is prepared from a solution of 3 per cent ligand I and 30 per cent PVC in 67 per cent *o*-nitrophenyl octyl ether. The lifetime of this sensor comes to more than a half year while the potentials drift at a rate of 0.1 mVd^{-1}. The final membrane had a thickness of about 0.5 mm and was mounted in a Philips electrode body type IS 560. Membranes of this type must be conditioned for 5 to 10 days (*Figure 7*), whereas sensors incorporating either
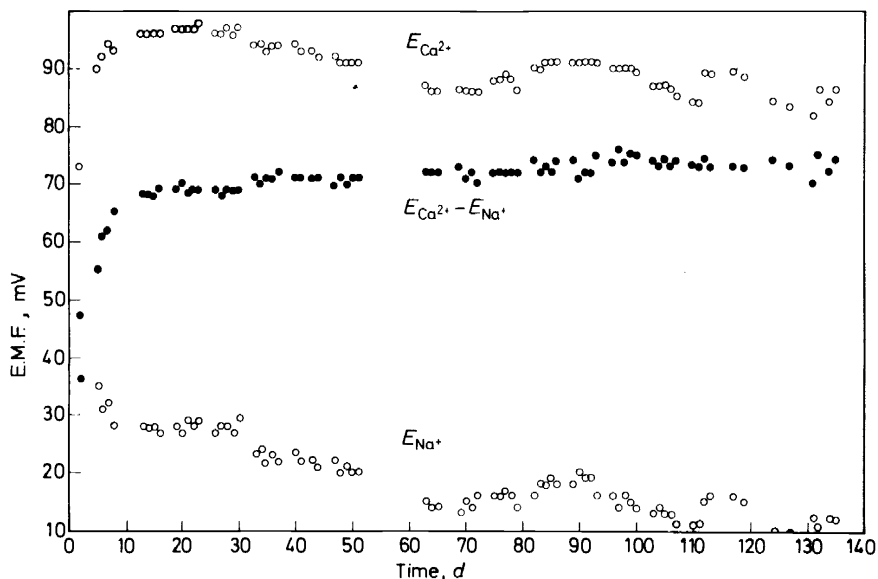

Figure 7. Response of the PVC electrode incorporating ligand I as a function of time. The e.m.f.-values were measured using unbuffered 10^{-2} M solutions of the metal chlorides at room temperature.

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thinner membranes of similar composition or membranes saturated in respect to calcium chloride can be used immediately without impairing the selectivity, the stability, or the lifetime. Using i.r.-spectroscopy, it has been shown that the ligand resides in these membranes in practically undiminished concentration for periods of over 100 days (*Figure 8*).
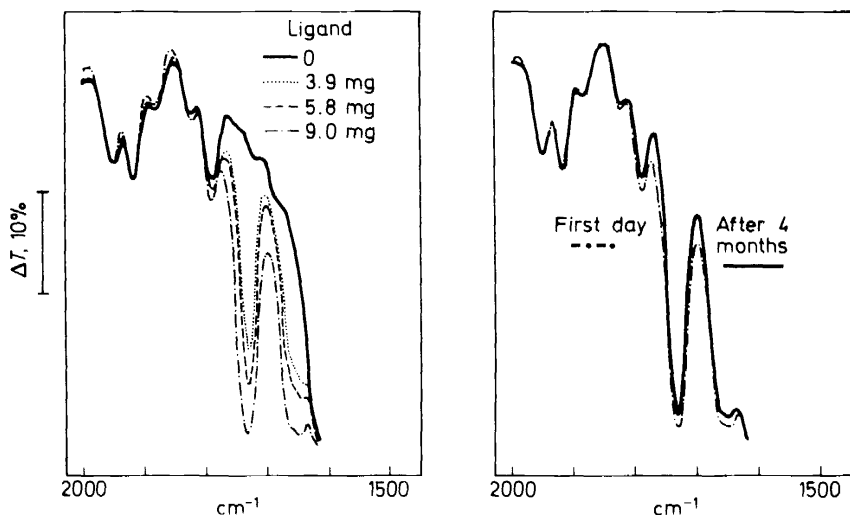

Figure 8. Infrared spectra of PVC membranes incorporating ligand I. The ester carbonyl absorption band of the ligand was used for reference purposes. The plot on the left shows the dependence on ligand concentration in the membrane, while the other graph compares the spectrum of an unused membrane with the spectrum of the same membrane after 4 months of continuous use in a sensor.

The e.m.f.-response of this sensor to sample solutions containing calcium chloride and sodium chloride respectively is represented in *Figure 9*. As activity standards the fundamental values set forth by Bates[35] were employed; the activity coefficients γ_{Ca} were found to be related to the ionic strength I as

$$\log \gamma_{Ca} = -2.04I^{\frac{1}{2}}(1 + 1.55I^{\frac{1}{2}})^{-1} + 0.2I \qquad (19)$$

It is evident from *Figure 9* that the described electrode exhibits a perfectly Nernstian response over a $10^0 \rightarrow 10^{-5}$ M calcium range, as well as a linear response to sodium down to about 5×10^{-3} M (see *Table 4*). Deviations from a purely Nernstian response are most likely explained by local deviations from the bulk equilibrium that is assumed to exist between the surface of the membrane and the bulk of the sample solution, especially so in the proximity of the phase interface. In *Table 5*, the selectivity factors K_{CaM}^{Pot} for a considerable number of cations M^{2+} are compiled. Each of these values was calculated
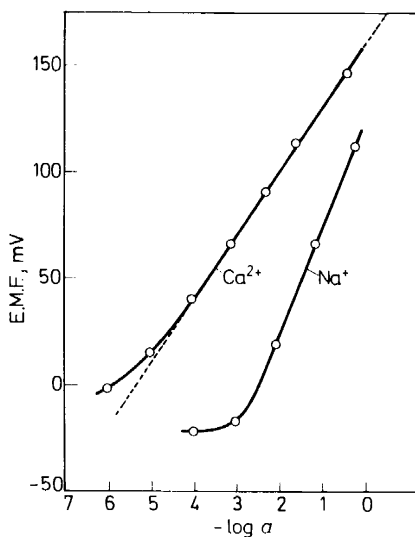
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Figure 9. E.M.F.-response of the sensor using ligand I in PVC to unbuffered aqueous solutions of calcium chloride and sodium chloride respectively (25°C). Philips electrode body IS 560.

Table 4. Calculated slopes of the e.m.f.-response in *Figure 9* at 25°C.

Electrode response to calcium		Electrode response to sodium	
Concentration range	Slope in mV/log a_{Ca} (linear regression)	Concentration range	Slope in mV/log a_{Na} (linear regression)
10^0–10^{-5} M	28.6	10^0–10^{-3} M	45.8
10^0–10^{-4} M	29.2	10^0–10^{-2} M	49.4
10^{-1}–10^{-4} M	29.6	Theoretical	59.2
Theoretical	29.6		

Table 5. Selectivity factors K_{CaM}^{Pot} characteristic of the described sensor based on ligand I (from e.m.f.-measurements using 10^{-2} M metal chloride solutions).

M^{z+}	K_{CaM}^{Pot}	M^{z+}	K_{CaM}^{Pot}	M^{z+}	K_{CaM}^{Pot}
Mg^{2+}	2×10^{-4} 2×10^{-4}†	Li^+	6×10^{-2}	NH_4^+	1×10^{-1}
Ca^{2+}	1	Na^+	3×10^{-1} 1×10^{-1}†	Al^{3+}	2×10^{-4}
Sr^{2+}	1×10^{-1}	K^+	1×10^{-1} 4×10^{-2}†	Cu^{2+}	2×10^{-3}
Ba^{2+}	9×10^{-1}	Rb^+	3×10^{-2}	Zn^{2+}	6×10^{-4}
		Cs^+	1×10^{-2}	Ce^{3+}	2×10^{-2}

† From e.m.f.-measurements using 10^{-1} M solutions.

from the electrode response to a 10^{-2} M solution of the respective chloride salt by applying a simplified form of Nicolsky equation 1

$$E = E_0 + \frac{RT}{2F} \ln \left[K_{CaM}^{Pot} a_M^{2/z} \right] \tag{1d}$$

The discrimination of potassium and sodium ions by the sensor incorporating ligand I† is somewhat reduced as compared to a PVC-electrode containing the Orion liquid ion-exchanger 92-20-02 (*Figure 10*). One of the
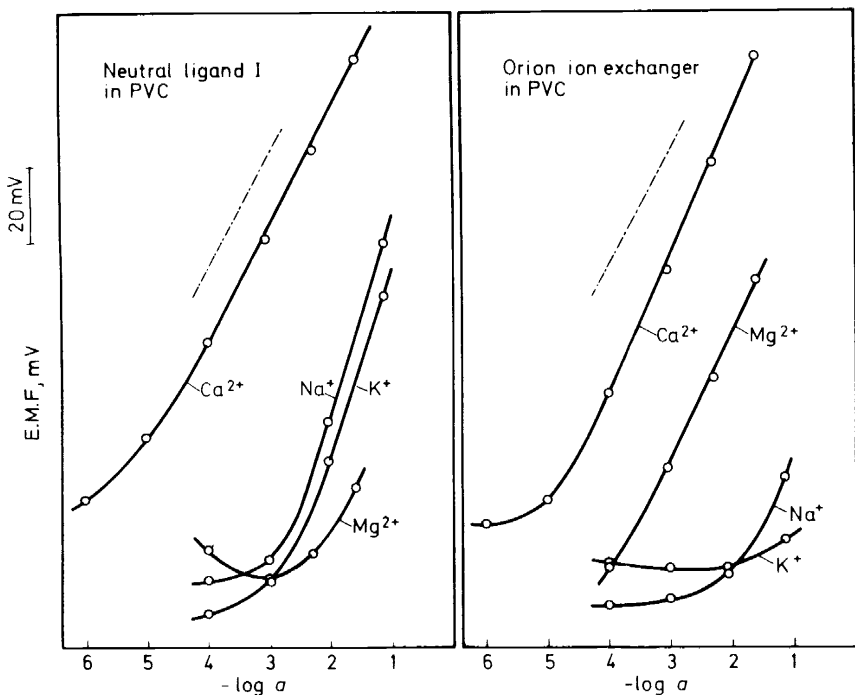

Figure 10. Comparison of the response of PVC electrodes with ligand I respectively classical ion exchanger to different cations in non-buffered aqueous solutions (25°C). Philips electrode body IS 560.

improvements[34] over the PVC-sensor using the Orion exchanger, and even more so the Orion 92-20 electrode, consists in the minimized hydrogen ion sensitivity. This low pH-interference was achieved through the use of neutral ligands devoid of any functional groups that could enter acid–base reactions. A further noteworthy trait of the calcium-sensor described here is its selectivity among divalent cations; for example, the marked discrimination of magnesium or zinc ions observed is unprecedented.

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† It is evident from *Table 3* that ligand IV, for example, exhibits a considerably higher specificity for calcium ions.

A potentially important application of calcium-sensors is the determination of the ionic calcium level in human blood serum. Since in this liquid the sodium concentration (~ 0.15M) is about two orders of magnitude greater than the calcium level to be measured, a grave interference of the former ion with the response to the latter would be expected. Deviations from the perfect calcium-response accruing from the presence of large amounts of sodium are of little importance, however, as is shown by e.m.f.-measurements conducted on mixed solutions of calcium chloride and sodium chloride. It would be unwise, though, to interpret this as incontrovertible evidence for a lack of sodium-interference, i.e. that the selectivity factors K^{Pot}_{CaM} representative for mixed solutions are very small. Rather, it seems as if the interfering ion's direct influence on the electrode potential is very nearly compensated by the induced change in the activity coefficient of calcium. If the sensor is to be judiciously employed to determine the Ca^{2+}-concentration in a sample such as blood serum, it is imperative that the electrode be calibrated against solutions of similar composition.

For sensors of the type propounded here, the expression 18 derived from theory is admirably suited for the interpolation of e.m.f.-values for samples resembling the electrolyte composition of blood serum ($\gamma_{Ca} \approx 0.33$, from 19). In principle, two calibration measurements are all that is needed. The e.m.f.-response towards a solution of pure calcium chloride of a given activity will serve to fix a value for E_0, and from the response, in the absence of cal-
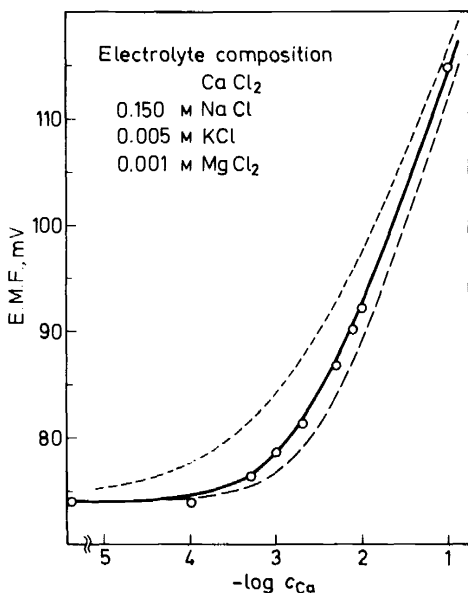

Figure 11. Comparison between calculated and measured response to Ca^{2+} of the PVC electrode using ligand I ($25°C$); Na^+-, K^+- and Mg^{2+}-level comparable to blood serum.

Upper curve: equation 1c $\Big\}$ K^{Pot}_{ij} respectively K^{Pot}_{ji} assumed to be constant
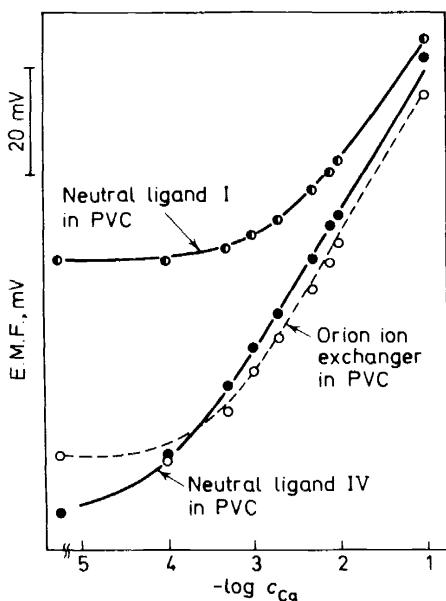lower curve: equation 1b
solid line: equation 18

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Figure 12. Response of different electrodes to Ca^{2+} in an electrolyte comparable to blood serum (NaCl 0.150M, KCl 0.005M, $MgCl^2$ 0.001M), 25°C; Philips electrode body IS 560.
  solid lines: calculated response according to equation 18.
  dotted line: calculated response according to equation 1b assuming K_{ij}^{Pot} to be constant.

cium, towards the presumably constant background of interfering ions it is possible to calculate the interfering-ion term $K_{CaNa}^M a_{Na}^2$ in equation 18. The electrode potentials calculated according to equation 18 and those measured were found to coincide to a remarkable degree (*Figure 11*). In the range 10^{-4} M to 10^{-1} M Ca^{2+} the average deviation turns out to be of the order of only 0.5 mV. The same level of precision was attained with interfering-ion backgrounds of 0.1 M and even 1 M sodium chloride.

The ligand IV prepared recently shows a discrimination of sodium in PVC-sensors which is at least ten times larger than the one obtained using compound I (*Table 3*). *Figure 12* clearly demonstrates that the corresponding electrode is in all respects far superior to the ones prepared earlier. A clinical application, for example, is certainly within reach.

ACKNOWLEDGEMENT

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