A radical pair mechanism, like that which has been proposed[5c,d] to account for low stereochemical integrity in 1,5-alkyl group migrations, is a less likely explanation because α-alkoxyalkyl radicals are not greatly stabilized relative to their primary alkyl counterparts.[16] The corresponding cations, on the other hand, are hardly comparable, for the former are oxonium species rather than carbocations.

(16) In a model reaction that involves radical pair formation ($ArCH_2HgCH_2Ar \rightarrow ArCH_2^{\bullet} + {}^{\bullet}HgCH_2Ar$),[17] *p*-methoxy is not rate enhancing, within experimental error, relative to *p*-methyl and only 3.4-fold enhancing relative to *p*-H.

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Synthetic Flux-Promoting Compounds, Exceeding the Ion-Transporting Ability of Gramicidin

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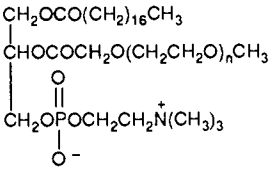
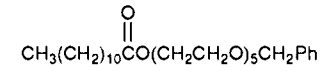
Discourses on membrane transport, such as this communication, often begin by extolling the importance of the subject. Continuing the practice, we note briefly that membrane transport underlies research into the molecular basis of diseases such as hypertension,[1] cancer,[2] epilepsy,[3] cystic fibrosis,[4] and malaria.[5] Our own specific interest in transport processes relates here to the synthesis of organic compounds that emulate the ion-channeling ability of gramicidin. Gramicidin is a pentadecapeptide antibiotic whose dimers from cation-conducting pores in lipid bilayers. So potent is gramicidin that a single channel carries a greater ion current than can an entire 1.0 × 1.0 mm gramicidin-free membrane.[6] To date only a few non-peptide mimics have been studied.[7-10] The examples reported herein are characterized by an attractive simplicity and a remarkable activity (surpassing gramicidin under certain conditions).

Discovery of our flux-promoting compounds was, admittedly, fortuitous. The original goal had been to synthesize and test phospholipids bearing a polyether chain (I). These materials proved incapable of accelerating ion movement across bilayers.



I

Such was not the case, however, with intermediates acquired in the synthesis of I having the general structure $RO(CH_2CH_2O)_nR'$. Upon discovering that these compounds facilitate ion translocation, we systematically varied the three sections of the molecules (R, *n*, and R′) to optimize the effect. As will be shown, the greatest success was achieved with II where R = dodecanoyl, *n* = 5, and R′ = benzyl.



II

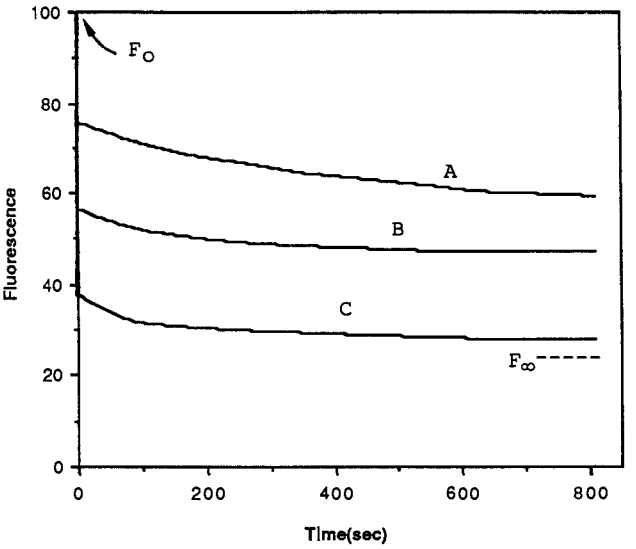


Figure 1. Fluorescence vs time data after adding HCl to pyranine-containing vesicles so as to abruptly lower the external pH from 7.7 to 6.6 (K_2HPO_4 buffer, 20.0 °C, [DSPC] = 0.3 mM). Polyether additives are all present at 45 molecules/vesicle assuming 5000 lipids/vesicle. Curve A represents plot for blank (no additive) as well as three inactive polyethers: (a) $R_1 = R_2$ = dodecanoyl, n = 5; (b) $R_1 = R_2$ = benzyl, n = 5; (c) R_1 = hexanoyl, R_2 = benzyl, n = 5. Curve B: R_1 = dodecanoyl, R_2 = benzyl, n = 3. Curve C: compound II.

Ion flux was monitored by a simple method that Kano and Fendler[11] developed a decade ago. Thus, 0.3 mM distearoylphosphatidylcholine (DSPC) and an acid-responsive fluorescent dye (the tetraanion of 8-hydroxy-1,3,6-pyrenetrisulfonic acid) were cosonicated[12] at 60 °C in a weak pH = 7.7 buffer containing 0.1 M KCl. This created unilamellar vesicles (diameter = 30 ± 5 nm according to our QELS measurements)[13] with dye situated both in the enclosed volume and in the bulk water. The latter was readily removed by gel filtration through a Sephadex G-25-80 column. A flux-promoting compound (gramicidin D or one or our mimics) was then added at the micromolar level and allowed to equilibrate at 20.0 °C with the vesicles. When an HCl pulse lowered the bulk pH from 7.7 to 6.6, protons entered the vesicles, dye with its pK_a = 7.2 became protonated, and the fluorescence was thereby depleted. Note that the dye, being polyanionic, cannot escape the vesicles,[11] nor will the dye bind to the membrane walls since all bilayers were provided with 10% anionic distearoylphosphatidate.[13]

Plot C in Figure 1 shows the fluorescence vs time dependence when there are only 45 molecules of II/"5000-DSPC" vesicle. It is seen that the fluorescence drops precipitously from an initial value of F_0 = 100% to a value somewhat greater than the theoretical F_∞ = 24%. This can be explained in a manner totally consistent with the literature[14-16] by assuming the following: (a) Any vesicle with even a single functional channel suffers an instantaneous and electrically neutral ion flux. (b) A certain number of vesicles lack a channel at the concentrations of the experiment. Increasing the concentration of II to roughly 75 molecules/vesicle

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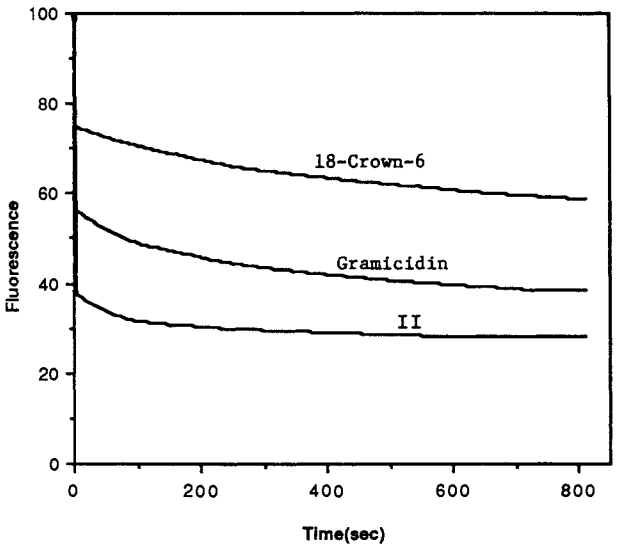

Figure 2. Comparison of 18-crown-6, gramicidin D, and II under conditions given in the caption of Figure 1. 18-Crown-6 is inactive, while the activity of II exceeds that of gramicidin D at 45 channel compounds/vesicle.

sufficed for total and immediate equilibration between external and internal ions. Greater than 1 molecule/vesicle is required for II because its ion-conducting segment (the polyether) is sufficiently long to traverse only a single leaflet. A *minimum* of two molecules (one in each leaflet) must, therefore, become aligned to permit passage across the entire membrane. This is also true of gramicidin.

Figure 1 compares the behavior of II (plot C) with that of related structures (plots A and B). Plot A, identical with the blank,[17] shows that all activity is lost if $n = 5$ and *both* R and R′ are dodecanoyl or *both* R and R′ are benzyl. Activity also disappears if R is II is shortened to a hexanoyl group. Plot B shows that activity is impaired if the polyether unit of II is shortened to $n = 3$. We conclude that spanning a polyether moiety across a leaflet to form a channel component requires (a) a benzyl group (presumably to associate with a DSCP quaternary nitrogen by an ion–dipole attraction)[18] and (b) a distal hydrocarbon tail that embeds itself in the apolar region of the membrane.

Figure 2 compares II with gramicidin D and 18-crown-6 at concentrations of 45 molecules/vesicle. Under these conditions, 11 actually exceeds gramicidin in activity! 18-Crown-6, on the other hand, manifests no enhancement above background. The latter is noteworthy because a U-tube system (a so-called "liquid membrane")[19] gives exactly the opposite behavior: K^+ was transported through $CHCl_3$ by 18-crown-6 at a concentration where 11 was totally inert. The simplest explanation is that the crown ether is a good ion carrier through $CHCl_3$,[20] whereas II (a poor K^+ ligand according to spectrophotometric studies)[21] is able to form channels in bilayers but not, obviously, in bulk $CHCl_3$.

Does 11 actually forge a channel, or does it merely create a defect? Although this is a difficult question to answer (especially since the distinction between a "channel" and "defect" is ill-defined), we prefer to view 11 as a channel element for the following reasons: (a) 11 is extremely active at nanomole levels. (b) Flux enhancement is structurally specific; several analogs of II that were both more and less hydrophobic lost all activity. (c) Leakage induced by II is not general; dye, for example, could not escape the channelized vesicles.[22]

(17) The fluorescence decrease for DSPC without channels corresponds to an electrically uncompensated proton diffusion.[15]

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Registry No. II, 125109-54-8; $H_3C(CH_2)_{10}CO_2(CH_2CH_2O)_5CO(CH_2)_{10}CH_3$, 92411-21-7; $PhCH_2O(CH_2CH_2O)_5CH_2Ph$, 60389-48-2; $CH_3(CH_2)_4CO_2(CH_2CH_2O)_5CH_2Ph$, 125109-55-9; $CH_3(CH_2)_{10}CO_2(CH_2CH_2O)_3CH_2Ph$, 125109-56-0.

(22) A further account of work in the area will appear in an issue of the *Boletin de la Sociedad Chilena de Quimica* honoring the important Chilean chemist Professor Lucho Sepulveda.

Production and Study in the Gas Phase of Multiply Charged Solvated or Coordinated Metal Ions

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Determinations of the gas-phase equilibria involving singly charged ions and solvent molecules such as H_2O (see eq 1), or other ligands, providing the sequential bond enthalpies $\Delta H°_{n-1,n}$ and entropies $\Delta S°_{n,n-1}$ were initiated some 20 years ago.[1,2] Such

$$M^+(H_2O)_{n-1} + H_2O = M^+(H_2O)_n \qquad (n-1,n) \qquad (1)$$

studies have provided a wealth of data[3] on ion–solvent and ion–ligand interactions.[4] Extensive theoretical work[5,6] and experimental studies such as laser spectroscopy of ion clusters in molecular beams[7] were also stimulated.

The above studies were limited to singly charged ions M^+, yet doubly charged ions M^{2+} are of paramount importance in chemistry and biochemistry. The general method used for ion clusters like $M^+(H_2O)_n$ is to generate M^+ in a gas phase in which ligand molecule vapor is present. The formation of $M^+(H_2O)_n$ then proceeds spontaneously by third body dependent association reactions like reaction 1. This method may not work[8] for M^{2+} when the second ionization energy of M, $IE(M^+)$, is larger than the ionization energy of the ligand, e.g., $IE(Mg^+) = 15.0$ and $IE(H_2O) = 12.6$ eV.

Doubly and triply charged ions exist in solution, but preparation of gas-phase ions like $M^{2+}(H_2O)_n$ by ion transfer from solution to the gas phase would not have been considered possible until recently. However, new mass spectrometric research has shown[9-11]

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(8) Little information is available on the outcome of gas-phase reactions: $M^{2+} + L$ where $IE(M^+) > IE(L)$. Charge transfer to the ligand, leading to M^+ and L^+ and formation of $M^+OH + H_3O^+$ for cases where the ligand and the third body are H_2O, are reaction channels that could compete with formation of stabilized $M^{2+}·L$.

(9) In electrospray, a solvent such as methanol containing low concentrations of an electrolyte (10^{-6}–10^{-4} M) flows through a narrow metal capillary. When the capillary is placed at a high voltage relative to ground (~5–10 kV), a fine spray of charged droplets is emitted from the capillary tip. The evaporation of the droplets in the ambient air leads to formation of gaseous ions. Electrospray has been known since the 1940s, but ion detection with a mass spectrometer, first reported by Fenn,[10] led to a renewed interest in this phenomenon.

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