Synthetic Organic Chemical Models for Transmembrane Channels

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Received April 17, 1996

Introduction

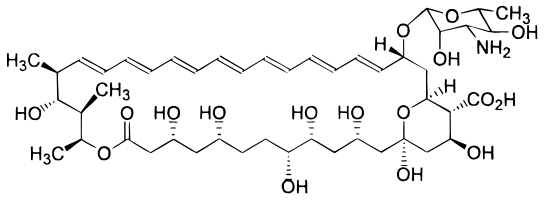
The flow of ions and molecules either into or out of a cell is a precisely regulated biological process. Specific transport systems modulate the molecular and ionic composition of the intracellular medium, and membranes control the flow of chemicals and chemical signals (information) between cells and their environment. Membrane permeability is modulated by complex proteins that repeatedly cross the bilayer and form "channels". An enormous amount has been learned about the function of transmembrane protein channels during the last decade, but details of the chemical mechanisms of transport and selectivity remain obscure.

Lipid bilayer membranes are impermeable to small ions such as Na^+ or Cl^- because considerable energy is required to transfer an ion from the aqueous phase into and through the apolar, hydrophobic interior of the membrane.[1] Ions are known to be transported in membranes both by carrier molecules and by channels.[2] A carrier compound is a molecule that binds the ion at one interface and transports it through the membrane as a complex. Carriers were viewed in the early literature as "ferryboats" diffusing back and forth across the membrane.[3] In contrast, channel molecules span the entire membrane, creating a tunnel-like pathway for ions. These different transport mechanisms are illustrated schematically in Figure 1.

Advances in molecular biology and protein chemistry using such techniques as patch-clamping, molecular cloning, and site-directed mutagenesis have permitted channel function to be probed in unprecedented detail. Structures are known for bacteriorhodopsin[4] and the acetylcholine receptor,[5] but even in cases such as the Na^+ channel of the electric eel (*Electrophorus electricus)* for which the primary structure was deduced more than a decade ago,[6] the three-dimensional arrangement can only be guessed and details of the chemical mechanism are even less certain. On the basis of the bacteriorhodopsin model, the transmembrane segments of most protein channels are thought to be α-helical,[7] although the generality of this notion has been challenged.[8] The paucity of chemical and mechanistic information concerning natural channels has inspired organic chemists to devise model systems in the hope that their function would provide insight into the function of transmembrane proteins.

Natural, Small-Molecule Channel Compounds

The thinking of modern organic chemists interested in channels has been influenced profoundly by three naturally occurring compounds: (1) nystatin and amphotericin B, polyene antibiotics;[9] (2) gramicidin,[10] a peptide that forms a cation-selective channel which is the best characterized of all channels, and (3) alamethicin, a peptide that forms voltage-gated channels.[11] These low molecular weight (MW 924−2000) compounds show characteristics of channel behavior: ion selectivity, voltage dependence, subconductance states, blocking, and modulation properties.[12] Amphotericin is an amphiphilic structure: the polar



amphotericin

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S0001-4842(96)00135-5 CCC: $12.00

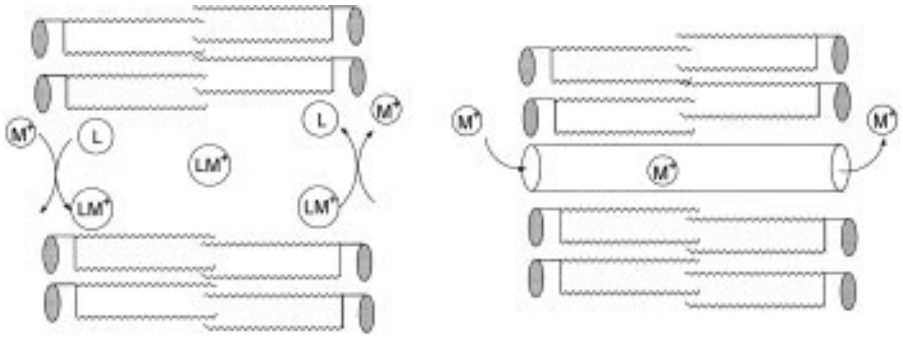

Figure 1. Schematic illustration of an ion carrier (left) and a channel former compound (right). M^+ stands for a cation and L for the free ligand (host).

mycosamine head group is attached to a less polar polyol fragment and a nonpolar polyene chain. In a steroid-rich membrane, amphotericin increases the flux of water, monovalent cations, and small nonelectrolytes. Nonelectrolytes larger than glucose cannot pass through the channels formed by these polyenes, suggesting a pore with a diameter of 8 Å. The model is that of the "barrel-stave" structure, which is formed from 8−10 of these molecules, with the hydroxylated portion of each molecule facing the inside, forming a water-filled pore extending across the bilayer.[13]

The simplest "protein" channels form from the gramicidins, a family of closely-related linear polypeptide antibiotics isolated from *Bacillus brevis*. Unlike proteins, alternate amino acids in gramicidin have D-stereochemistry. The structure may be summarized as OHCNH-L-Val-Gly-L-Ala-D-Leu-L-Ala-D-Val-L-Val-D-Val-L-Trp-D-Leu-L-xxx-D-Leu-L-Trp-D-Leu-L-Trp-$CONHCH_2CH_2OH$, in which xxx has the following identities: gramicidin A (gA), Trp; gB, Phe; and gC, Tyr. Gramicidin D (Dubos), the most studied system, is a mixture of gramicidin A, B, and C (∼80:5:15).[14]

Gramicidin is a good model for protein channels because (1) it is a peptide, (2) the tail-to-tail dimer[15] spans the membrane (∼26Å), (3) it conducts monovalent cations, and (4) it is blocked by divalent cations.[16] Na^+ and K^+ are transported through the channel at rates of 10^7 ions/s.[17] The alternating L- and D-configuration results in a $\beta^{6.3}$-helix[18] in which the side chains penetrate the bilayer and the backbone carbonyls all face the channel interior. The result is a coil-like structure (diameter ∼4Å) through which cations can pass, relayed by the carbonyl groups. D-Amino acids do not occur in proteins so this cannot be the mechanism of conduction in a natural, α-helical channel.

Alamethicin is one member of a family of naturally-occurring peptides which form voltage-gated channels across membranes and, thus, provides a good model for how channel opening and closing can be controlled by the transmembrane potential. It contains 20 amino acids. The key structural feature of alamethicin and its homologues is that they readily form α-helices.

Channel Models

During the past few years, several research groups have reported the syntheses of cation-channel-forming compounds, in an attempt to elucidate the ion transport mechanism (ion selectivity and gating) through lipid membranes. Mutter and Montal[19] have used template-assembled synthetic proteins (TASPs)[20] which were designed to adopt a globular, four-helix bundle conformation that forms ion channels in lipid bilayers that may be like natural channels. DeGrado and co-workers[21] synthesized model peptides containing only leucine and serine residues. A 21-residue peptide, H_2N-(Leu-Ser-Ser-Leu-Leu-Ser-Leu)$_3$-$CONH_2$, formed ion channels, with ion permeability and lifetime (opening and closing) characteristics resembling those of the acetylcholine receptor. This work has been the subject of an Account.[22]

Synthetic Organic Ion Channels

A second group of channel model systems may be loosely classified as nonpeptidic, and this category can be further subdivided. This includes (1) "half-channel elements" that must aggregate and organize within the bilayer to achieve function and (2) membrane-length, "tunnel-like" structures.

The earliest designed, nonpeptidic channel model resulted from pioneering studies by Tabushi and co-workers.[23] The structure used β-cyclodextrin's secondary hydroxyl groups to provide the polarity required to stabilize the amphiphile within the bilayer, and four "relay"-containing side chains were attached randomly to primary hydroxyl groups (Figure 2). The channel transported copper and cobalt: $k_{Co(II)} = 4.5 \times 10^{-4}$ s^{-1}, [channel monomer] = 55 μM. This transport rate was much faster than that in the absence of a channel, but no further results appeared before Tabushi's untimely death in 1987.

Protochannels

Menger and co-workers attempted to prepare phosphatidylcholine derivatives in which the sn-1 posi-

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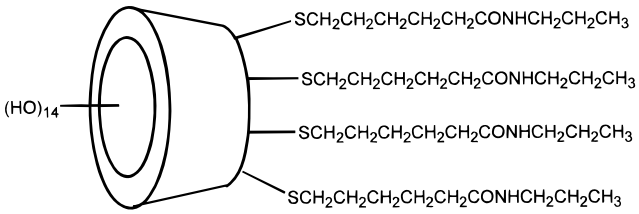

Figure 2. Semischematic illustration of Tabushi's β-cyclodextrin ion channel.

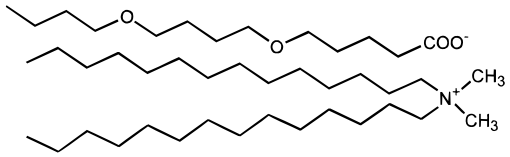

Figure 3. Kobuke's amine/carboxylate system.

tion[24] was acylated by stearic acid and sn-2 was acylated by $HOOCCH_2(OCH_2CH_2)_nOCH_3$.[25] The success or failure of this approach was not further elaborated, but interesting results were obtained from a simple ester compound, apparently an intermediate in the phospholipid synthesis: $CH_3(CH_2)_{10}COO(CH_2CH_2O)_5CH_2Ph$. Proton transport was assessed in phospholipid bilayers by using a spectroscopic technique developed originally by Fendler and Kano.[26] In this method, pyranine dye is trapped within the vesicles. Transport out of the liposomes, induced by a pH change and mediated by the channel former, is followed by decay of the fluorescence signal.

Menger's ester has three "domains" that can be identified. First, the fatty acyl polymethylene chain is hydrophobic. Second, the ester and ethyleneoxy groups are relatively hydrophilic. Third, the benzyl terminus, said to be required for proton transport, may serve as a membrane anchor.[27] Individual monomers do not appear to be long enough to span the membrane. A simple carrier compound, 18-crown-6, was inactive under identical conditions. The H^+-transport capabilities of this protochannel exceeded that of gramicidin. Transport may result from alignment of individual monomers diffusing in opposite membrane leaflets which affords a transmembrane pore.

Kobuke and co-workers[28] demonstrated transport of Na^+ or K^+ by glycolate ethers of monoalkyloligo(1,4-butylene glycol) combined with dioctadecyldimethylammonium cation. They postulated that the carboxylate group of $BuO[(CH_2)_4O]_n(CH_2)_4COO^+$ was complexed with the quaternary ammonium head group of $Me_2N^+R^1R^2$ where R is typically a C_{16} alkyl chain (Figure 3). These chain lengths are similar to leaflet thicknesses in bilayers, so it is presumed that the amphiphiles dimerize to function. We speculate that opening and closing of the channels occur when lateral relaxation in the membrane leaflets leads to complex alignment, producing a transmembrane "defect" through which cations pass. The channels were not cation selective: single-channel conductances in 0.5 M NaCl were almost the same as those in 0.5 M KCl.

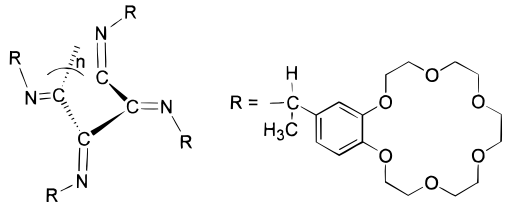

Figure 4. An isonitrile-derived, oligomeric ion channel.

A Rigid Tunnel Model

While investigating crown-substituted isonitriles, Nolte and co-workers[29] obtained an oligomer that they presumed possessed four tunnel-like tubes of crown ether rings atop each other, spaced at a (crown ether face-to-face) distance of ~4 Å. The oligomer, estimated to span about 40 Å, was incorporated into vesicles prepared from dihexadecyl phosphate; $k_{Co(II)} \approx 10^{-4}$ s^{-1}. No transport of Co^{2+} was detected by UV−vis spectrophotometry in the absence of the poly(crown).

Several bolaamphiphiles have been reported to be channel models but have been characterized only as cobalt transporters[30] (Figure 4).

Voyer and Robitaille used crown ethers to form a "tunnel" through which cations can pass on their transmembrane journey.[31] Their channel model organized adjacent benzo-21-crown-7 residues on -Leu-(CrF-Leu-Leu-Leu-CrF-Leu)$_3$-, an α-helical peptide chain, in which "CrF" stands for crown−phenylalanine (Figure 5). By placing the macrocycles on every fourth residue, they are approximately aligned in a tubular arrangement.

Cation flux was assessed in unilamellar phospholipid bilayers. A proton transporter (FCCP) was added to the membrane to enhance proton flux. The change in pH was monitored using a pH-stat as various alkali metal cations were added to the medium. The authors suggested that poor selectivity within the group of cations Li^+, Na^+, K^+, and Rb^+ could be attributed to the large size of the macroring. A peptide that was too short to span the bilayer was found to function as a carrier rather than as a channel. No transport rate constant was reported, but a graphical representation suggested that maximal flux was reached in ~200 s. The transport rate remained constant for >2000 s, until the vesicles were lysed by addition of the neutral detergent Triton X-100.

Tartaric Acid as a Channel Central Unit

The ready availability of tartaric acid (HOOC−CHOH−CHOH−COOH) in optically active and *meso* forms has made it a valuable synthetic building block. Schrieber and co-workers[32] used its oppositely-oriented

(24) The three hydroxylated carbons of glycerol in lipids are designated sn-1 to sn-3. The chiral carbon is sn-2, and the lipid head group is located at sn-3.

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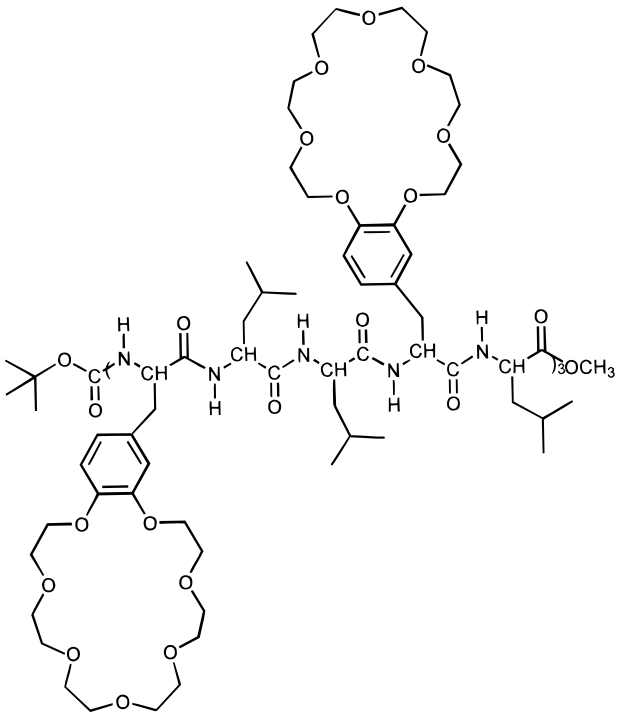

Figure 5. Voyer and Robitaille channel of benzo-21-crown-7 rings positioned on an α-helical peptide backbone. The peptide chain is α-helical, aligning the macrocycles rather than placing them on opposite sides of the chain as shown.

carboxyl groups to link two gramicidin units. By so doing, a covalent, "dimeric" arrangement was achieved that approximated the putative dimer channel conformation of gramicidin in a bilayer. Rather than being a channel model *per se*, the preparation of this system constituted a mechanistic probe in the best traditions of bioorganic chemistry.

Macrocyclic polyethers derived from tartaric acid residues were the basis for the first families of synthetic, tunnel-like channel systems. Two such systems, based upon very different design principles, were developed almost simultaneously by the groups of Lehn and of Fyles. Lehn and Jullien[33] used the six alternating carboxyl groups of hexacarboxy-18-crown-6 to anchor dendrimer-like chains which radiated from the central macrocycle. The name "chundle" (a contraction for "channel formed from a bundle of fibers") was suggested for this unique molecule. No cation transport data were reported in this preliminary communication.

The Fyles approach[34] involved building a tunnel always using crown central units but constructing the walls from bolaamphiphiles in approximate analogy to the amphotericin barrel-stave notion. The most sophisticated of Fyles' structures possessed three "walls" on either side of a central macrocycle. These

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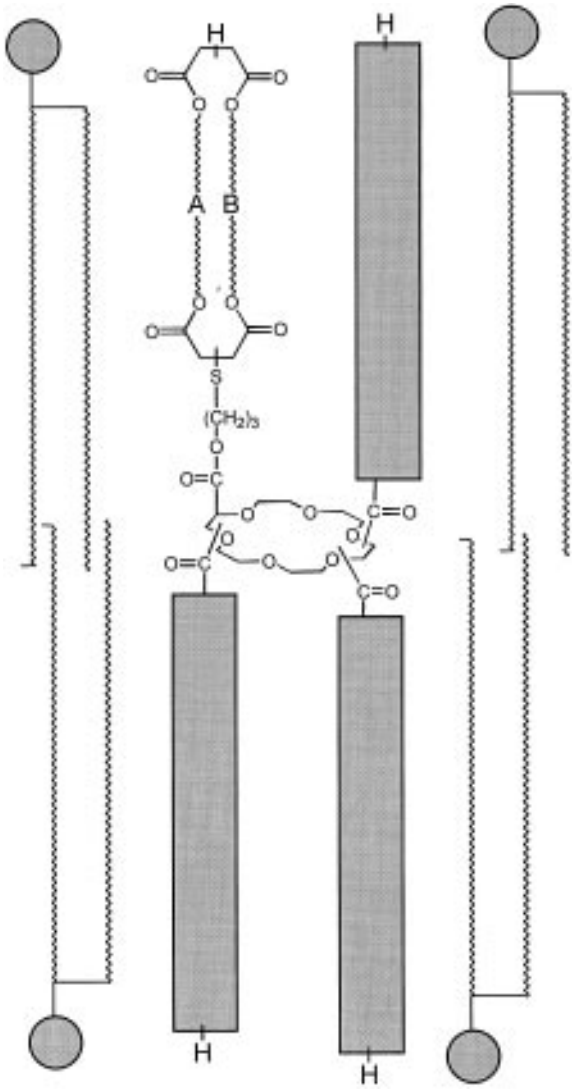

Figure 6. General structure representing the family of channel models prepared by Fyles and co-workers. The gray rectangles represent the bolaamphiphile structure illustrated. See the text for the identities of chains A and B and head groups H.

two different approaches require separate discussion, which follows.

Fyles Family of Macrocyclic Channels

The channels synthesized for this first major systematic study can be represented as shown in Figure 6. The design involved a relatively nonpolar "molecular tunnel", anchored in the membrane by polar head groups, of appropriate distance to completely span the bilayer (~30 Å). The carboxyl groups attached to the crowns were part of one, two, or three tartaric acid units so two, four, or six "wall units" were ultimately attached. Each wall was connected to a "head group" such as glucose, 2-mercaptoacetic acid, or 3-mercaptopropanol. The length and polarity of the wall units could be varied by changes in the units shown in the Figure as "A" and "B", but in all cases, they remained bolaamphiphiles.

Cation flux was assessed using a pH-stat method developed for this purpose and also used by others.[31] This method permitted an assessment to be made of the Li^+, Na^+, K^+, Rb^+, and Cs^+. In an otherwise identical group of three structures, varying the head group from glucose to mercaptoacetic acid to ethanol

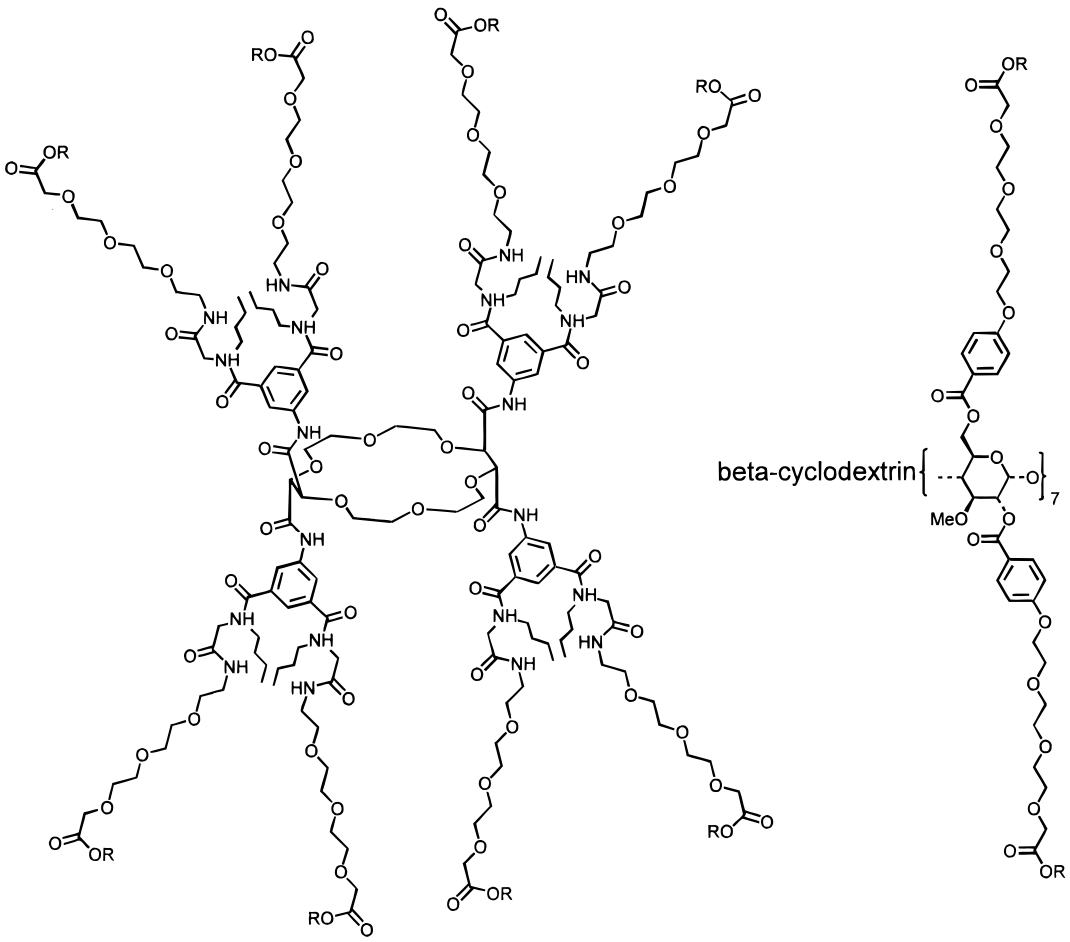

Figure 7. Structures of bouquet molecules, based on an 18-crown-6 derivative (left) and on β-cyclodextrin (right).

led to a decrease in proton flux. Among the compounds having glucose head groups, four (rather than two or six) wall units proved most conducive to channel activity in otherwise identical systems. The six-wall structure was apparently poorer than the two-wall system. Such comparisons must be made with care as one pair of channels which differed by whether the four side arms were arranged up, up, down, down *vs.* up, down , up, down were reported to alter from channel-like to carrier-like in their mode of action.

When the diol chain (A or B) was eight carbons (1,8-octanediol), the overall channel length was appropriate to span the bilayer. Thus, K^+ transport was higher than when the chain was shorter (1,5-pentanediol, worse) or longer (1,12-dodecanediol, worst). It might be expected that altering an $-O(CH_2)_8O-$ chain to $-(OCH_2CH_2)_3O-$, which has essentially the same length but provides more potential polar interactions, would lead to a more effective channel. In fact, when A = B = octyl, flux was higher than when additional oxygens were present. The most effective channel of the three possessed one chain of each type (*i.e.*, A = octyl, B = ether).

In biological studies, channels and carriers are usually distinguished in one of two ways. First, channel function is not temperature dependent whereas carrier transport slows as the temperature drops. Second, flux rates for channels are typically 10^3-10^4 greater than for carriers in black lipid membranes.[35] Fyles and co-workers used a combination of kinetic and inhibition studies to categorize their models. They concluded that, of 10 structures actually classified, half functioned as carriers. It was not obvious how small variations in generally similar molecules led to this change in mechanism. For at least two of the compounds, the cation selectivity order, namely, $K^+ > Rb^+ > Cs^+ > Na^+ > Li^+$, remained the same even though the mechanism of action was reported to differ.

It should be noted that the data obtained for this family of compounds used a coupled proton−metal cation assay. It is assumed that the method is not limited by the effectiveness of FCCP,[36] the proton coupling agent. It is also assumed that the method is not compromised by the use of K_2SO_4, although this has been noted as a potential problem.[37]

Lehn's Channel Structures

Lehn's design approach also used a central crown ether having rigidly-oriented connectors (carboxyl groups). Instead of wall units, a dendrimer-like array of amides, anchored by terminal hydroxyl groups, radiated from the crown. The concept was elaborated in later work from the same group with a change from crown ether to β-cyclodextrin as the central element (Figure 7). The structural alteration led to a change in name from "chundle" to "bouquet" molecules. In the β-cyclodextrin case, poly(oxyethylene) or polyalkyl chains, terminated by carboxylate end groups, were

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(36) FCCP is carbonyl cyanide [4-(trifluoromethoxy)phenyl]hydrazone.

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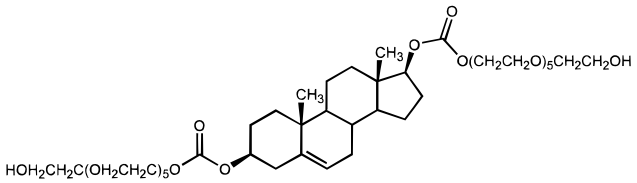

Figure 8. A steroidal amphotericin channel mimic.

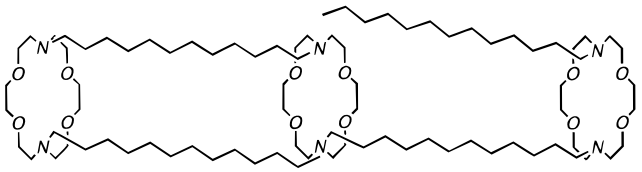

Figure 9. A tris(macrocyclic) channel model system.

attached to the toroidal heptasugar. The internal diameter of β-cyclodextrin is large enough (~7 Å) to allow the passage of metal ions as well as small organic molecules.

Cation transport was assessed by using a "counter-current" NMR method. Vesicles containing LiCl were prepared from egg phosphatidylcholine and the appropriate channel model. Opposing gradients of Li^+ (inside) and Na^+ (outside the vesicles) permitted transport of the cations in opposite directions (down their concentration gradients). Transport was followed directly by either ^7Li or ^{23}Na NMR. Channel function was nicely confirmed for the bouquets which mediated one-for-one exchange of Na^+ for Li^+ (antiport) in a process that was monitored over ~8000 min (4.8×10^5 s).[38] Under similar conditions, gramicidin equilibrated (~100%) in <10 min.

Lehn's Li^+/Na^+ NMR assay was used by Regen and co-workers[39] to assess flux through a phospholipid bilayer mediated by a 5-androstene derivative esterified in the 3β and 17β positions by $OCO(CH_2CH_2O)_nH$ groups (Figure 8). The compound was designed to mimic the function of amphotericin B by placing the steroid at the bilayer midplane and creating a channel with the ethyleneoxy groups extending from either end. Like the bouquet molecules, these compounds equilibrated cations within ~4000 min.

Tris(macrocyclic) Channel Compounds

Our own work to develop a channel model system began before the results from Lehn and Fyles appeared. Our approach was similar in the sense that a central macrocycle was used but different because our model was designed to be flexible. The essential elements of the design included a head group anchor, a central (bilayer midplane) relay, and connective units. Diaza-18-crown-6 was chosen to enforce a rudimentary cation selectivity on the system, but its principal advantage is that connectors can be attached at nitrogen and they remain conformationally flexible. The crown was envisioned as the entry point for the cation as well as the membrane anchor.[40]

At the outset, it was unclear whether crown ethers could function generally as head groups in membranes. Work by Kuwamura[41] and by Okahara[42] suggested that alkyl-substituted crowns could readily form micelles, but the formation of stable vesicles would have been more encouraging to us. That crowns can serve as head groups for the formation of stable vesicles was demonstrated in a separate series of studies.[43]

The "tunnel" notion was prominent in our original thinking about channel models. The first target is shown in Figure 9. It has three macrocyclic rings connected by 12-carbon, alkyl chains. Two such chains and three macrocycles give the compound an overall length in the 25−30 Å range (gramicidin ~26 Å[15]). The 12-carbon flexible tails were expected to insinuate themselves into the membrane to ensure an opening throughout the bilayer. The compound is shown in the Figure in the tunnel-like conformation in which it was originally envisioned.

Studies conducted using the H^+-flux fluorescence technique mentioned above[26] on two structures that differed only in the covalent chains proved enlightening. When the covalent spacer units were altered from $-(CH_2)_{12}-$ to $-(CH_2CH_2O)_3CH_2CH_2-$, proton flux was significantly diminished. This seemed counter-intuitive since the availability of a larger number of donor atoms within the channel structure was expected to enhance cation flux by lowering the transport energy required in each step.

An alternate interpretation is possible, however. One might expect that the larger number of donor groups would lead to stronger cation binding. This property may be desirable in complexation chemistry, but the dynamics of cation channel function requires that weaker binding should lead to faster transport. 18-Crown-6 is selective for K^+ over Na^+ in water. The equilibrium binding constant may be expressed as K_{eq} (usually called K_S for the stability constant) = $k_{complex}/k_{release}$ or k_1/k_{-1}. In water, the rates (k_1) for binding of either Na^+ or K^+ by 18-crown-6 are similar, ~2.2×10^8 and 4.4×10^8 M^{-1}. Selectivity for K^+ over Na^+ is determined by the respective cation release rates (k_{-1}) which are, respectively, 3.4×10^7 s^{-1} and 3.7×10^6 s^{-1}.[44] Thus, Na^+ is simply released faster. This means that channel transport selectivity will be *opposite* to that of complexation if the binding relationships are the same.

Since it is the alkali metal cations that are of greatest interest in the channel context, we observed Na^+ transport directly in phospholipid vesicles using dynamic ^{23}Na NMR. In this approach, pioneered by

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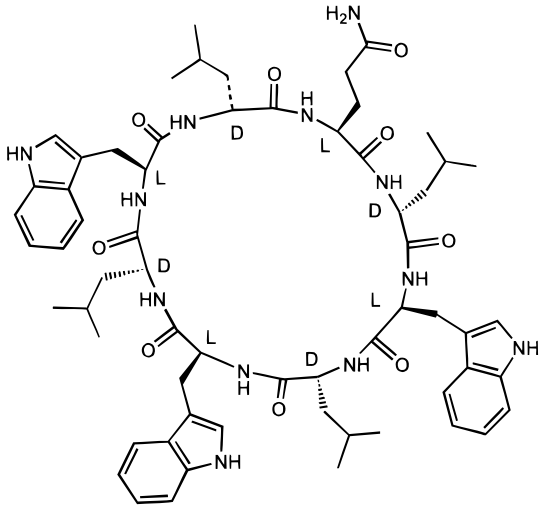
Riddell and co-workers[45] and applied to the gramicidin system by Hinton and co-workers,[46] phospholipid vesicles are prepared in the presence of NaCl. Addition of a Dy^{3+} shift reagent to the aqueous medium shifts the external Na^+ signal with respect to internal Na^+. When a channel is present in the membrane, the line width varies in proportion to cation flux. A plot of [channel] *vs* line width gives a rate that is, in all cases, normalized to the rate observed in the same system for gramicidin. A family of compounds has been prepared and investigated. Because the structures are complicated, we have devised an abbreviation system[47] to represent them. Thus, the compound of Figure 9 is C_{12}<N18N>C_{12}<N18N>C_{12}<N18N>C_{12}. This compound has a sodium cation transport rate of 28 compared to gramicidin's (normalized) 100.

The current state of knowledge concerning these compounds[48] is as follows. First, it appears that the central ring is important, but it is not required that the cation pass through it. This is known from studies in which the central ring was varied from 4,13-diaza-18-crown-6 (<N18N>) to 4,10-diaza-18-crown-6 (<N15N>) to $O(CH_2CH_2O)_3$. Transport rates were identical in the first two cases and reduced only by ~50% in the latter. Second, removing a side chain (C_{12}<N18N>C_{12}<N18N>C_{12}<N18N>C_{12} → H<N18N>C_{12}<N18N>C_{12}<N18N>H) did not affect the rate, but when the distal rings were converted from C_{12}<N18N> to C_{12}<N18> (diaza-18-crown-6 to aza-18-crown-6), all transport ability was lost. Third, changing the distal chains from *N*-dodecyl to *N*-benzyl caused a 40% increase in cation flux from 28 to 39. Fourth, exchanging the dodecyl side arms for cholestanyl residues cost the model system all activity. Our explanation for this surprising phenomenon, admittedly based upon very limited evidence, is that the essentially flat α-surface of the steroid interacts strongly in the van der Waals sense with the opposite (intramolecular) dodecyl chain. This essentially closes the channels, making passage of any ion impossible. Of course, cation flux may only be slowed beyond our ability to detect it; certain other channel models equilibrate far slower than ours.

Some of these channel model compounds can function as carriers in a bulk $CHCl_3$ membrane (*vs* valinomycin), but there is no correlation with activity in a bilayer *vs* gramicidin. The cation appears to pass through the distal macrocycles because substituting benzyl by nitro or methoxy makes the cation transport, respectively, slower and faster. This three-point line for the Hammett equation shows a correlation coefficient of 0.96.[49]

Identical ^{23}Na NMR experiments done in the presence of either an ionic or a nonionic detergent substituted for the channel model failed, showing that a simple detergent effect could not account for the cation transport activity. Additional "control" studies were accomplished by using segments of the channel model system. The following fragments of the successful model system were also shown to be inactive: <18N>C_{12}<N18N>C_{12}<N18>, C_{12}<N18N>C_{12}<N18N>C_{12}, $HOOC(CH_2)_{11}$<N18N>$(CH_2)_{11}COOH$, and C_{12}<N18N>C_{12}. A combination of experimental measurements and calculations has resulted in octanol/water partition coefficients ("*P*" values) which confirm that membrane solubility is favored over water by $>10^{10}$ in all cases. The differences in cation transport activity are thus not due to differential membrane solubility. We currently believe that the tris(macrocycle) uses each distal macrocycle as a head group to anchor the amphiphilic molecule at the membrane boundaries.[50]



cyclo[-(Trp-D-Leu)$_3$Gln-D-Leu]

Figure 10. A cyclopeptide-based nanotube channel.

Self-Assembling "Nanotubes"

The formation of transmembrane ion channels from self-assembling peptide nanotubes was reported by Ghadiri and co-workers.[51] The system design was based upon cyclic peptides. The eight-residue, cyclic peptide has the structure cyclo[(Trp-D-Leu)$_3$Gln-D-Leu-] (Figure 10). Alternating the stereochemistry places the amino acid side chains roughly along the equator of the cycle and directed away from the center. This obviously favors interactions with lipid chains if the system is membrane-bound. The diameter of the internal orifice is ~7.5 Å.

Hydrogen bond formation presumably drives channel formation as does the increase in the lipid chain entropy arising from side chain−lipid interactions. The stacking of these cyclopeptides can be envisioned as being similar to *β*-sheet formation. The fact that this tubular compound stack forms a large pore is reflected in its lack of cation selectivity: the conductance of NaCl is 55 pS, and that of KCl is 65 pS.

Cation Selectivity

The goal of most of the work described here has been first and foremost to see if channel function can be mimicked by synthetic organic model systems. The difficulty of this quest makes the number of successes

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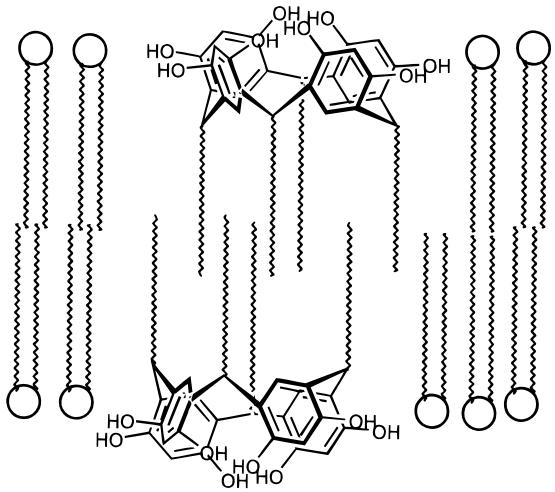

Figure 11. A resorcinarene half-channel element.

limited, but the present Account shows that there are successes. Collateral goals of understanding mechanism and achieving selectivity are of obvious consequence. An attempt to achieve selectivity was made by Kobuke and co-workers[52] who used a resorcinarene derivative to mimic the postulated "desolvating collar" of some natural channels. The notion was that the rigid tetraphenylene structure would present a cation entering the membrane with a physical barrier that selects cations on the basis of size. Space-filling molecular models support this expectation. The channel model is shown in Figure 11. Like Tabushi's cyclodextrin, this compound was expected to serve as a "half-channel" element.

After incorporation of the channel former into planar soybean lecithin bilayers, the compound exhibited stable and constant conductances at various voltages, and exhibited transitions between open and closed states. The compound discriminates potassium from sodium, with a permeability (selectivity) ratio (P_{K^+}/P_{Na^+}) of almost 3. It was also found that the channel was blocked by the larger Rb^+ cation.

Conclusions

The development of synthetic models for cation function poses a complex problem. The model systems must be designed in consideration not only of structure but also of function. We have referred to this principle as "property directed synthesis".[53] Once the design is decided, the compound or compounds must be synthesized. After preparation, alkali and alkaline earth cation flux induced by the model structures must be assessed in phospholipid bilayers if the effort is to be biologically relevant. Taken together, this requires a range of techniques and breadth of resources. The relatively small number of groups that have undertaken the model channel challenge reflects these difficulties. The potential payoff is considerable, however, because transmembrane channels are currently among the most important and widely studied biological structures. Channels are ubiquitous and, unfortunately, still poorly understood.

The present Account shows that diverse approaches to model channels may succeed although flux rates and cation selectivities vary considerably among model systems. The efforts made by several groups have only demonstrated the potential of synthetic models as an aid to understanding channel function and selectivity. It seems reasonable to think that, in the coming years, information resulting from the chemical programs will be of crucial importance to the vigorous biological community currently studying diverse channels and their various functions.

AR9601353

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