Does the Na channel conduct ions through a water-filled pore or a condensed-state pathway?

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SUMMARY

Many investigators assert that the ion-conducting pathway of the Na channel is a water-filled pore. This assertion must be reevaluated to clear the way for more productive approaches to channel gating. The hypothesis of an aqueous pore leaves the questions of voltage-dependent gating and ion selectivity unexplained because a column of water can neither serve as a switch nor provide the necessary selectivity. The price of believing in an aqueous pore therefore is a futile search for separate ad hoc mechanisms for gating and selectivity. The fallacy is to assume that only water is available to carry ions rapidly, ignoring the role of the glycoprotein, which can form an *elastomeric* phase with water. The elastomer is a state of matter, neither liquid nor solid, in which the molecules of a liquid are threaded together with cross-linked polymer chains; it supports fast ion motion (Owen, 1989).

An alternative hypothesis for channel gating, based on condensed-state materials science, already exists (Leuchtag, 1988, 1991*a*). The ferroelectric-superionic transition hypothesis (FESITH) postulates that the Na channel exists in a metastable ordered (closed) state at resting potential and, on threshold depolarization, undergoes a reversible order-disorder phase transition to a less-ordered, ion-conducting (open) state. The ordered state is ferroelectric; the disordered state is a fast ion conductor selective for Li^+ and Na^+. The basis of the voltage dependence is elevation of transition temperature with electric field, well established in ferroelectrics. FESITH is consistent with single-channel transitions, gating currents, heat and cold block, and other phenomena observed at channel or membrane level. An implication of FESITH, the Curie-Weiss law, has been shown to explain existing data on membrane capacitance versus temperature in squid axon (Leuchtag, 1991*c*). Only on the basis of a clear understanding of function can we expect new structural data on the Na-channel glycoprotein to generate realistic structure-function models at the molecular level.

QUESTIONS AND ANSWERS

The following dialogue is intended to clarify the differences between frequently accepted views (questions) and the views proposed by the author (answers):

(*a*) *Does the high transfer rate of sodium ions through a single Na channel demonstrate that the permeation pathway is a water-filled transmembrane pore?* (see Hille, 1984; Eisenman and Dani, 1987):

Not really. Aqueous solutions certainly have high ionic conductivities, but they do not represent the only way ions could pass through the channel at high transfer rates. Other condensed-state materials, called superionic conductors or fast ionic conductors are known to have ionic conductivities comparable to aqueous solutions, over $1\ S\ m^{-1}$ (Laskar and Chandra, 1989). What's more, these materials are highly selective for certain ions or groups of ions, whereas bulk water, as in a transmembrane pore, is not.

(*b*) *Because we know that gramicidin molecules and gap junctions are pores, can't we conclude that the Na channel and related channels are pores as well?*

That does not logically follow. Na, Ca, K and other excitable ion channels exhibit similar behavior; comparison of their primary structures indicates close evolutionary relationships between them. But neither gramicidin nor gap junctions exhibit close structural relationships to the excitable channels. Functionally, the only property they have in common with excitable channels is that they conduct ions; but they do not exhibit the selectivity, gating and other special properties that characterize the excitable channels. Therefore we cannot conclude that ion conduction in Na channels is like that in gramicidin or gap junctions.

(*c*) *What does the aqueous-pore hypothesis explain? What does it leave unexplained?*

The aqueous-pore hypothesis explains the ability of ions to pass through the membrane rapidly. Other phenomena that are left unexplained and so require separate explanations include: voltage-controlled gating, selectivity, inactivation, channel hysteresis, heat and cold block, voltage-dependent membrane swelling and temperature-induced currents. Thus, explanations of channel behavior by proponents of the aqueous-pore hypothesis re-

0006-3495/92/04/22/03 $2.00

quire special mechanisms, usually described in terms of mechanical devices, such as hinged or sliding gates, selectivity filters, and tethered balls.

(*d*) *What's wrong with describing gating and other mechanisms in terms of models based on mechanical devices?*

These models apply mechanical concepts valid at the centimeter scale to a channel at the angstrom scale. It must be remembered that atoms do not contact one another (as some models suggest) but influence each other by their electric fields. Matter at the atomic scale must be described by quantum mechanics. Because the electronic wavefunctions of neighboring atoms overlap at the angstrom scale, classical mechanics cannot validly describe the behavior of a channel. Thus, these models are invalid, and must be replaced by models that reflect the quantum nature of matter.

(*e*) *Are there any superionic conductors that selectively conduct sodium ions?*

β-alumina, $NaA1_{11}O_{17}$, is the best superionic conductor of Na^+ ions at moderate temperatures; it also conducts AG^+, Li^+ and K^+. It is a crystal with a planar triangular lattice, which grows with an excess of cations in the conducting plane. About one third of the unit cells are occupied by two alkali cations instead of one. The low activation energy for hopping of ions from doubly to singly occupied sites is believed to be the reason for the high ionic conductivity of the material (Kennedy, 1976; Mahan, 1976).

A sodium conductor of similarly high conductivity is Nasicon (Na SuperIonic CONductor, $Na_3Zr_2Si_2PO_{12}$). The structural units in Nasicon are zirconium octahedra and phosphorus/silicon tetrahedra, with open spaces interconnected in a three-dimensional lattice of intersecting conduction channels (Alpen and Bell, 1979; Collin and Boilot, 1989; Kohler et al., 1983).

(*f*) *Does a superionic conductor have to be a crystal?*

No, fast ion transport has been reported in glasses (including Na conductors), polymers and heterogeneous solids. Examples of fast ion conducting polymers are poly(ethylene oxide), poly(vinylidene fluoride) and Nafion (Laskar and Chandra, 1989).

(*g*) *If the Na channel transports ions by superionic conduction, does that imply that water is not involved?*

No; on the contrary, fast ion conduction in organic polymers most frequently involves a liquidlike mechanism of ion transport, in which the translational motion of the ions depends on the local motion of the surrounding host structure. Polymers are rarely single-phase solids; rather, they are usually composites of many phases, which may be crystalline, glassy, elastomeric or liquid.

(*h*) *What is an elastomeric phase, and how can it account for superionic conduction?*

An elastomer is the host phase for ion conduction in a polymer electrolyte. In an elastomeric phase, local motion is liquidlike, but long-range displacements are forbidden, just as they are in a solid. Like a solid, an elastomer has low vapor pressure, a well-defined shape and negligible long-range self-diffusion of the structure. These properties come about by the threading together of molecules of a liquid with long and occasionally cross-linked chains of covalent bonds. In the Na channel, these would be water molecules with polypeptides.

An elastomer is a state of matter that is neither liquid nor solid, but which requires a new approach to the understanding of its conductive properties. A fundamental difference between liquid and elastomeric ion conduction is this: In a liquid, the solvent molecules associated with the ion can remain with it throughout its trajectory, i.e., all the way through the channel, whereas, in an elastomer, dissociation must occur (Owen, 1989).

(*i*) *How can ion gating in the Na channel be explained by a condensed-state approach?*

This has been explained by the assumption that the Na channel possesses the property of ferroelectricity. Ferroelectricity is a common property of materials, occurring in crystals, liquid crystals, and polymers. Ferroelectric materials have ordered dipoles, producing a spontaneous polarization that remains after an applied electric field has been removed, and that can be reversed by an opposing field. Ferroelectrics therefore exhibit hysteresis. Ferroelectric order can be removed by raising the temperature to a transition temperature, the Curie point (Lines and Glass, 1977; Mitsui et al., 1976).

Gating in the Na channel has been explained by the ferroelectric-superionic transition hypothesis (FESITH) as a phase transition. The closed state seen at the high electric field of the resting potential is a ferroelectric state. The open state reached by reducing the electric field in a threshold depolarization is a paraelectric (nonpolar), superionically conducting state (Leuchtag, 1987 *a*, *b*, 1988, 1989, 1990 *a*, *b*, *c*).

A phase transition from closed to open can be achieved either by (1) raising the temperature above the Curie point (heat block), or (2) lowering the Curie point below the existing temperature.

The Curie point of ferroelectrics is field dependent. Decreasing the electric field (as in a threshold depolarization) lowers the Curie point to bring about a phase transition that opens the channel.

(*j*) *What is the FESITH explanation for Na-channel block by neurotoxins such as TTX?*

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The TTX molecule binds on the external surface of the Na channel. As it binds, it changes the local electric field configuration in a special way—it freezes, or clamps, the ferroelectric domain. Once clamped in this way, the channel cannot be induced to undergo a phase transition to the paraelectric-superionic state by depolarization at physiological temperatures. Domain clamping has been observed in triglycine sulfate and other ferroelectrics (Lines and Glass, 1984, 116).

(k) Is the link between ferroelectricity and excitable membranes a new idea? Has it been further elaborated by others?

The idea that ferroelectricity may be related to the generation of electric impulses in nerves and muscles was proposed by Arthur von Hippel in 1969, and again later by Eisenman, Edmonds and others. A ferroelectric mechanism was even suggested for memory in the brain (Leuchtag, 1988 and references therein). Recently, Shirane et al. (1990) adapted their self-organized dynamic theory to a ferroelectric membrane, obtaining time-dependent functions of membrane excitation.

(l) Is there any evidence to support the ferroelectric-superionic transition hypothesis?

Indeed there is. Hysteresis, the defining property of ferroelectrics, has been reported in excitable membranes and in Na channels. Heat block and cold block, required by the model, have long been known to exist. Pyroelectricity, which always accompanies ferroelectricity, was observed as temperature-jump currents in frog nodes. Piezoelectricity, another concomitant of ferroelectricity, was observed as membrane swelling. The surface charge of the channel is quantitatively consistent with the spontaneous polarization of ferroelectrics (for details and references see Leuchtag, 1987*a*, 1991*a*).

(m) Have you fitted data to FESITH predictions?

Yes. The FESITH requires that the Na channel obey the Curie-Weiss law of ferroelectrics, which relates dielectric permittivity ("constant") to temperature. Data on temperature dependence of squid-axon membrane capacitance obtained by Palti and Adelman (1969) have been fitted accurately to the Curie-Weiss law (Leuchtag, 1991*c*).

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