LETTERS TO THE EDITOR

[Brief letters to the Editor that make specific scientific reference to papers published previously in THE JOURNAL OF GENERAL PHYSIOLOGY are invited. Receipt of such letters will be acknowledged, and those containing pertinent scientific comments and scientific criticisms will be published.]

A Single Channel or a Dual Channel Mechanism for Nerve Excitation

Dear Sir:

Narahashi and Haas (1968) and Hille (1968) have suggested that data obtained when various pharmacological agents are applied to nerve make it likely that the channels in the axon membrane that carry Na and K are separate entities in contrast with the suggestion that there is a single channel (Mullins, 1959) that modifies its selectivity first in favor of Na and later in favor of K. My purpose is to examine the assumptions underlying the analysis that has been made and to show that the evidence for two channels is not at present compelling.

The models to be considered are shown below.

Single Channel Scheme

$$\text{Impermeable } (A) \xrightarrow[m]{} \text{Na Permeable} \xrightarrow[h]{} \text{Impermeable } (B) \xrightarrow[n]{} \text{K Permeable}$$

Separate Channel Scheme

$$\begin{array}{l} \text{Na Channel Impermeable} \xrightarrow[m]{h} \text{Na Permeable} \\ \text{K Channel Impermeable} \xrightarrow[n]{} \text{K Permeable} \end{array}$$

Implicit in the single channel scheme is the assumption of a single, voltage-dependent, time constant that controls the change of the membrane from *Impermeable* (*A*) to K *Permeable*. The Na *Permeable* phase is governed by two subsidiary time constants and any appreciable lengthening of especially the *h* process will preclude the appearance of the K *Permeable* state. The two-channel scheme has a Na *Permeable* state that can be turned on by the *m* process and turned off either by promoting *h* or by a voltage change reversing *m*, while the K process, independent of the Na channel, is turned on or off by the *n* process. For either scheme it is necessary to suppose that the membrane has a dispersion of channel time constants so that there is some overlap between Na and K currents flowing through the membrane.

The classical Hodgkin-Huxley analysis treated the three processes, represented by the conductance variables *m*, *h*, and *n* as independent of each other, but the analysis

itself does not imply either a single channel or a dual channel mechanism, concerned as it is only with convenience in representing the phenomena of excitation.

One may well ask what sort of experimental demonstration would be conclusive in showing that there were two channels in the excitable membrane. The maximum values of partial ionic conductance, g_{Na} and g_K, are about the same for lobster and squid axons (Narahashi and Haas, 1968; Cole and Moore, 1960) and are each about 100 mmho/cm^2. If there were two channels, both could, in principle, be opened at the same time; the membrane conductance of the axon would then be 200 mmho/cm^2. There is not, however, any experimental data where the membrane conductance exceeds the maximum value of the conductance for a single ion, or, 100 mmho/cm^2. This is an important point because the fact that the conductance does not exceed a single ion conductance makes it possible in principle for a single channel to carry either Na, K, or a mixture of these ions.

A key observation in the two-channel argument is that tetrodotoxin (TTX) when applied to the outside of axons is able to block the Na current without influencing either the time course or the magnitude of the K current. The inference drawn from such an experiment is that TTX has blocked the Na channels and left the K channels unaffected. In the absence of specific information as to how g_{Na} is increased in response to depolarization, it is equally possible to suppose that TTX changes the channel size or channel affinity for Na without TTX entering the channel.[1] Such an arrangement could be brought about in a variety of ways that are not relevant to the present argument; it is perhaps simplest to distinguish between a channel assuming Na selective form and the ability of Na^+ to flow through the channel. The channel selectivity depends upon a membrane time constant controlling the opening and closing of the channel to Na and a final opening of the channel to K^+. Whether any ions flow through the channel could depend on the modulating influence of substances present at the channel entrance on either side of the membrane.

A second pharmacological argument used in favor of separate channels for Na and K is the effect of tetraethylammonium ion (TEA) in blocking K^+ currents while leaving unaffected the Na^+ currents of the axon (Hille, 1967; Armstrong and Binstock, 1965). This effect is related to that produced by introducing Cs^+ inside the axon, where I_K is blocked with an increase in the duration of I_{Na} (Adelman and Senft, 1966 *a*) and to the introduction of a high Na, low K inside the axon where the sodium currents are greatly prolonged (Adelman and Senft, 1966 *b*). What all these observations suggest is that the internal ionic composition of the axon affects K and Na currents in a way that is not clearly understood but is most easily summarized by saying that foreign cations compete with K^+ for transfer so that the *apparent* number of K channels is greatly reduced. While the effect of TTX is poorly reversible, the effect of TEA is reversible so that it is possible to assume that this ion enters channels in the K form and so lowers channel mobility for K that no K current flows. It is,

[1] The TTX molecule is rather large compared with what one imagines is a plausible value for channel size. The fact that TTX acts only when applied to the outside of the membrane favors the notion that it cannot pass through the membrane and may well act at a site distant from the Na channel.

of course, equally reasonable to assume that TEA acts at a site different from the K channel and merely prevents the channel from attaining the proper size or affinity so that it can carry K^+.

A final pharmacological argument (Narahashi and Haas, 1968; Hille, 1968) concerns the effects of dichlordiphenyl-trichloroethane (DDT) on the Na and K currents in frog nodal membranes and in lobster axons. The effects of DDT on these two membranes are not identical but for the present discussion the differences are not important. The principal actions of DDT are: (*a*) about a 5-fold slowing in the rate at which the Na current is turned off by inactivation, and (*b*) about a 3-fold decrease in the maximum value of g_K, the potassium conductance. There are, in addition, less important changes in the rate of turning on of I_{Na} and I_K. The result of these changes induced by DDT is that a very appreciable Na^+ current continues to flow at times as long as, for example, 5 msec following a depolarizing pulse while in a normal axon depolarized to 0 mv such a Na current would be entirely negligible. Since, at 5 msec I_K has been turned on, the fact that Na and K currents flow at the same time has been used as evidence in support of a two-channel mechanism. What has been overlooked in this analysis is the very large effect that DDT has in reducing the maximum value of g_K. As an example, at 5 msec, I_{Na} in a DDT-treated lobster axon has been inactivated to about half its initial value for a depolarization to −20 mv while I_K is not quite at its steady-state value. The corresponding conductances for Na and K are g_{Na} = 50 mmho/cm² and g_K = 30 mmho/cm² so that the sum is 80 mmho/cm², a value appreciably less than that of the maximum peak[2] g_{Na} which is 100 mmho/cm². It would appear, therefore, that the results obtained with DDT support, if anything, a single-channel model for nerve excitation in the sense that g_K is appropriately reduced as the time course of g_{Na} is prolonged.

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[2] I have used the maximum peak g_{Na} because it is an experimentally measured quantity. The Hodgkin-Huxley $\bar{g}_{Na}$ is about twice as great and implies that (on a single channel scheme) only half the Na channels reach a K-carrying state.

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