The domains in the Na channel have specific functions

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The classical voltage-dependent sodium channel is responsible for the upstroke of the action potential. Its pore forming subunit is a single polypeptide that has four homologous domains. Although each one of these domains is similar to a single subunit of the voltage gated potassium channel, the structures differ among each other. For example, the number of basic residues in S4 segments, responsible for voltage-sensing, is different in each domain. We have probed the function of each domain in the overall operation of the channel using fluorescent tags because they can detect local conformational changes. The results indicate that the S4 segments of domains I, II and III have faster kinetics than the S4 segment of domain IV. The kinetics and voltage dependence as reported by the fluorescent probe in the first three domains agree well with that of the fast component of the gating currents. On the other hand, the kinetics of the fluorescence probe attached in S4 of domain IV matches the kinetics of the slow component of the gating current. In addition, the turn-on of the fluorescence of domain IV exhibits a lag that is not observed in the fluorescence of the first three domains, indicating that the movement of S4-DIV does not start until one of the other three S4's has moved. The kinetics of S4-DIV (or the slow component of the gating current) is too slow to account for the activation of the conductance but is faster that the time course of inactivation. In fact, the ionic current develops before the fluorescence change in S4-DIV. Taken together, these results indicate that the first three domains are responsible for the activation of the conductance and suggest that S4-DIV does not participate in channel opening. Other correlations of the fluorescence of S4-DIV with the kinetics and steady-state properties of inactivation indicate that the function of S4-DIV is related to the voltage dependence of inactivation that is ultimately produced by the IFM motif that blocks ion conduction.

The local detection of conformation by the fluorescent probe can also be used to test possible interactions between domains during the operation of the channel. Thus, by introducing a mutation in one domain that affects the voltage dependence of the movement of that domain, one can ask whether that mutation has an effect in the movement of another domain. We have found that in fact all domains interact with each other with positive cooperativity. It can be demonstrated that positive cooperativity among voltage sensors increases the overall kinetics of channel opening. These results provide at least a partial explanation as to why sodium channels are so much faster than potassium channels, a requirement to generate the action potential.

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