Gating currents
Machinery behind the molecule

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Na channels equip cells to generate and propagate electrical signals. These signals transmit information, trigger contraction, regulate spontaneous beating, and initiate other activities required for our existence. For example, the Na channel plays a role in the process of fertilization. The Na channel works because it can sense membrane voltage. But how does voltage sensitivity work? In this issue, Crouzy and Sigworth have made a significant step toward answering that question. The problem they solve involves the electrical fluctuations in Na channel gating currents. Before understanding the implications of this work, we need to state some basic facts and retrace some earlier work.

Channels open with certain probabilities; ions then flow through the channels, this current causes the membrane voltage to change, and the voltage in turn induces the opening probabilities of the channels to assume new values. This cycle explains most of membrane excitability, i.e., the ability of cells to produce action potentials. For the mechanism to work, however, something in the channel has to sense the voltage. Presumably, the sensor is electric charge embedded in the protein: when the voltage changes, the charge moves, causing a conformational change in the protein that leads to channel opening. We call this charge the gating charge and its movement the gating current.

Hodgkin and Huxley (1952) predicted that gating currents ought to exist, and Armstrong and Bezanilla (1974) eventually measured them. The explanation for the long delay between theory and experiment comes from the difficulty of measurement: gating currents have small amplitudes and extremely fast kinetics. Even now, gating currents are far too small to see as individual events, and all gating currents come about from the addition of currents from thousands of channels. Some parallels exist here with ionic currents. Before we were able to measure single channel currents, we estimated their size from an analysis of noise (the minute fluctuations in the total current that comes from many channels). Now, in a remarkable replay of this story, Conti and Stühmer (1989), and Crouzy and Sigworth, have shown us how to estimate the properties of individual Na channel gating currents in exactly the same way. However, the noise-estimation method for obtaining elementary events is much easier for ionic currents, because ionic currents involve the movement of thousands of charges over tens of milliseconds, whereas gating currents involve the displacement of only a few charges for an immeasurably

short time. Thus, ionic currents come in temporal blocks that we can measure one at a time or deduce from noise analysis, but gating currents come as tiny blips that we cannot see. For the present, at least, noise analysis remains the closest method that we have to looking at them directly.

To examine this further, imagine one Na channel in a patch, and suppose that we could measure both its ionic current and its gating current. The gating currents would appear as a series of blips just before each channel opening. Interestingly, they could also occur at other transitions between the closed or inactivated states when no ions flow. Now eliminate the ionic current and picture only the random blips: the same model that would predict channel kinetics would also predict the pattern of arrival of the blips. Next, picture thousands of channels with all of the blips adding up randomly to create the macroscopic gating current; this current will have a fluctuating component (the gating-current noise) that deviates from the mean current (which Armstrong and Bezanilla measured). The problem that Crouzy and Sigworth have solved concerns the theory of gating-current noise. In their paper, they show how to go from virtually any Markov model of ion channel kinetics to an experimental expectation for gating-current noise. And, in a neat parallel with channel noise theory, they compare that expectation with an actual measurement to tell us something about the elementary events.

Conti and Stühmer had made that theoretical leap in their experimental paper, but Crouzy and Sigworth have generalized it. Sigworth had already developed a method for the analysis of nonstationary noise, which he used to study Na channels. This strategy takes into account the transient nature of the Na current. Measuring the deviation from the mean at each moment during the transient results in the variance of the current as a function of the mean: plotting these two against each other provides us with a useful graph. From such a graph comes the number of channels and the size of the current through any one of the channels. So simple and powerful, this analysis has all but eclipsed correlation functions and spectral densities. The complete correlation function, especially for nonstationary noise, is far more complicated. In their expanded application of this theory to the random arrival of individual gating currents, Crouzy and Sigworth have given us the general formulas and the mean-variance reduction. Then they use the mean-variance plot to compare two different models of Na channel kinetics.

Now arises an intriguing difference between channels and gates: channel noise decreases at high frequencies, but gating noise increases. The clue to this behavior lies in something already mentioned; channel currents are blocks, but gating currents are blips. Because gating currents last only very short times, they contain information at extremely high frequencies. This feature of gating-current noise causes the bandwidth of the measurements to figure centrally in the theory. Analysis shows that conclusions about models get stronger at frequencies above the present 8 kHz. The exact opposite is true for channel-current noise. Crouzy and Sigworth end their paper with a warning to users: noise analysis (especially the mean-variance plots) may not be a good way to distinguish models. We could, however, look at this apparent drawback in a more positive light: the estimates obtained for the channel number and size from the mean-variance plots depend only weakly on the specific details of channel models and have validity beyond them.

Nevertheless, we would have to admit that, as a model tester, nonstationary gating-current noise analysis has some way to go. However, as a glimpse into a world that we cannot watch directly, the paper already succeeds, and we get a bonus. The theory applies not only to ion channel gating, but also to other situations of interest to membrane biophysicists. For example, the theory pertains to open-channel noise (the fluctuations that occur in the ionic current as charges move through the open pore). Moreover, Crouzy and Sigworth's approach may work for the analysis of transient currents observed in transporters and pumps (Läuger, 1991). The original attempts to measure noise from carriers yielded very little information. One reason for this failure may have been the emphasis on the low frequency noise associated with the slow kinetics of active transport. However, from the viewpoint of the gating theory presented in this issue, we might look forward to a rebirth of transporter noise analysis, but now with a focus in the high frequency range. One last remark about channels and pumps: in channels, the ions that move through the pores far outnumber the charged gates that dislocate; in pumps, however, the ions that shuttle across the membrane are comparable in number to the fixed charges that move in the transporter protein. This feature of transporters might lead to novel theories for transporter noise and to new experiments that could give us a biophysical peek into that molecular domain.

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