Time Delay in Structural Shifts: Modeling Multiple States

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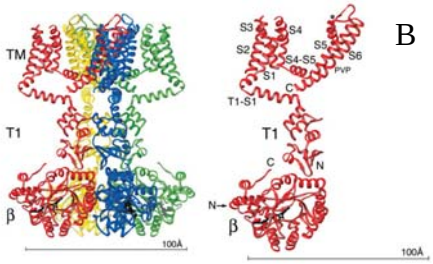
Introduction:

The reaction rates of most biological processes are linked to the concentrations or pressures of reactants and constant parameters. Additionally, some systems must “remember" a state for long periods of time. For example, two repressor proteins might control one another's synthesis by negative regulation. Qualitative reasoning suggests that such a system will have two stable states, one in which the "first element is on" and the second “off ", and another in which these states are reversed [1]. However, state mediation may take place via any number of intermediary states. Although direct observation is potentially difficult experimentally, these intermediate states can more accurately model entire systems where only equilibria were before considered. Consider the two reactions: (1) $[H]+[A]\underset{k2}{\overset{k1}{\leftrightarrow}}[HA]$ and (2) $[H]+[A]\underset{k2}{\overset{k1}{\leftrightarrow}}[\overline{HA}]\underset{k4}{\overset{k3}{\leftrightarrow}}[HA]$, where $\overline{HA}$ represents the intermediary step between the reactants and products with its own reaction rates in either direction. By carefully choosing k1-k4, we can describe a system with identical equilibra as in (1) but with different mediating landscapes and time scales. We see this approach as providing more variability than models which only predict equilibria.

Applications:

Voltage-dependent ion channels of biological membranes are formed by pore-like single proteins that extend through the cell membrane [2]. When dwelling in open

conformations, (specific) ions are allowed to pass through the membrane and participate in important cellular processes (neural excitability, etc.). In closed (resting/blocked) conformations ion flow is prevented. Current models of these so-called gating dynamics emphasize the residence times at structural conformations and the temp./voltage-dependent ODE's that can describe the equilibria. Conceptually simplifying the channel to a "switch" is useful, since electrically, the channel is open during ion flow, and closed when no current is present, but this description of channel dynamics subtly assumes that once a voltage/temp threshold is surpassed, opening and closing take place instantly, whereas *in vivo* this important step lasts ~ 1 ms [3]. We hope to introduce a new conformational state that accurately considers opening/closing delay and provides agreement with past voltage clamp studies *and* the voltage-sensor/pore coupling elucidated by the crystal structure [4].

A 

B

Figure 1

Each ion channel (fig. 1A) is composed of four sub-units (fig. 1B). Helices S1-S4 constitute the voltage sensing apparatus and are mechanically linked to S5 and S6 (the inner helices), which perform the actual opening and closing [5].

Charged amino acids, termed gating charges, allow the sensors to react to changes in voltage across the cell membrane. The displacement of the voltage sensors (and S4 in particular) has been the subject of much debate [4], but their temporal impact on recovery time-scales will be of primary importance here. In addition to being open or closed, channels can be "inactivated" (blocked) by the N-terminal domain of the β subunit,

which binds to the pore in potentially either open or closed states [3, 6]. Conversely, "deactivation", the unbinding of the N-terminal domain, takes place either before or after the channel closes on the return to a closed, unblocked state, termed recovery, which can take different routes with distinct physiological meanings [6].

Qualitative descriptions of gating dynamics tend to predict gating currents far too simple [2] or suffer from a "proliferation of parameters" that contain as many as 14 structurally unidentified closed substates [2]. We hope to determine a new kinetic scheme with rate coefficients that accurately model channel equilibria *and* time delays caused by structural constraints.

References:

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