Channel adaptation (1) involves complex behavior of the sarcoplasmic reticulum (SR) Ca\textsuperscript{2+} release channel (ryanodine receptor, RyR). The adaptive behavior is different from conventional ion channel behavior in that adapting channels transiently activate (open) in response to repeated increments of agonist (Ca\textsuperscript{2+}). No published kinetic model known to us could adequately account for this adaptation. We propose a model that readily accounts for the complexities associated with adaptation.

The model is based on the established tetrameric RyR channel structure. Biochemical studies indicate that at least three high affinity Ca\textsuperscript{2+}-binding sites exist per monomer (2). We postulate that one kind of binding site (the O-domain) tends to open the channel when activated by Ca\textsuperscript{2+} and another (the A-domain) tends to close (adapt) it. Thus, the tetramer would then have four O-domains and four A-domains.

The model uses the following rules to describe the behavior of the channel. The channel opens when the number of occupied O-domains (O) on the tetramer exceeds or equals the number of active A-domains (A), that is, $O \geq A$. However when $A > O$, or $O = 0$, the channel remains closed. In order for the model to fit the published experimental data, each O-domain must be cooperative ($n = 2$) and have lower affinity and faster Ca\textsuperscript{2+}-binding kinetics than each A-domain (Fig. 1, methods). At high [Ca\textsuperscript{2+}], when both O- and A-domains are occupied, the data are best fit if the channel opens 75\% of the time when $O = A$.

This model predicts that the steady-state probability of a channel being open ($P_o$) varies as a monotonically increasing function of [Ca\textsuperscript{2+}] (Fig.1A) and that fast Ca\textsuperscript{2+} steps trigger transient bursts of channel activity well above the steady-state level (Fig. 1, A and B). The predicted concentration- and time-dependent occupancy of O- and A-domains on a tetramer during two step increases in [Ca\textsuperscript{2+}] are shown (Fig. 1C). Our model accurately accounts for the experimental data which defines RyR adaptation (1). Published models (3) cannot reproduce the observed second transient response of the “apparently inactivated” channel. This is expected in light of theoretical thermodynamic analysis of RyR behavior, which indicates that one would need a large number of Ca\textsuperscript{2+}-binding sites to explain adaptation (4).

Our adaptation model resolves apparently contradictory results and suggests future areas of experimentation. The model explains how elevated [Ca\textsuperscript{2+}] can both apparently “inactivate” peak SR Ca\textsuperscript{2+} release in situ and increases steady-state $P_o$ of individual RyR channels. It also enables the SR Ca\textsuperscript{2+} release channels to respond to [Ca\textsuperscript{2+}]\textsuperscript{+}, a classical observation of Fabiato (3) in skinned muscle. Adaptation in vitro, however, appears too slow to regulate SR Ca\textsuperscript{2+} release in cells (1, 5). Thus, the model predicts that (as yet) unknown endogenous factors may accelerate the rate of adaptation (for example, modulate binding kinetics of A-domain) in cells.

Finally, this model may apply to other intracellular Ca\textsuperscript{2+} release channels as it could also provide the basis for the incremental, or “quantal,” Ca\textsuperscript{2+} release from inositol triphosphate (IP\textsubscript{3})-sensitive Ca\textsuperscript{2+} stores (6) [An IP\textsubscript{3} receptor channel tetramer with O- and A-domains incrementally occupied by IP\textsubscript{3} may explain the quantal character of the IP\textsubscript{3}-induced channel opening as well as the incremental channel “inactivation” observed by Hajnoczy and Thomas (6)].

Thus, we propose that the complex adaptive channel behavior arises from the assembly of simple interactive monomers. Although each monomer by itself is not able to produce the complex behavior, the interplay among the monomers exponentially extends the functional flexibility of the assembled unit.

### TECHNICAL COMMENTS

**Models of Ca\textsuperscript{2+} Release Channel Adaptation**


20. Control of cochlear amplification by extracellular voltages has been mentioned in passing by a number of authors (or, for example, Davis [Am J. Otolaryngol. 2, 153 (1981)] and (15).


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**Fig. 1.** Numerical simulation of exact adaptation with successive stimuli. Upper trace shows current, lower trace shows ligand concentration. Steps are 3 s in duration. L = 0 from 0 to 1.5 s, and it then increases in exponential steps, that is, 0.0316, 0.1, 0.316, 1, ... 100 mM. Rate constants are $k_1 = 1$ s$^{-1}$, $k_{-1} = 100$ s$^{-1}$, $k_2 = 10$ s$^{-1}$, $k_{-2} = 0.1$ s$^{-1}$, $k_3 = 1000$ mM$^{-1}$ s$^{-1}$, $k_{-3} = 12000$ s$^{-1}$, $k_4 = 2000$ mM$^{-1}$ s$^{-1}$, $k_{-4} = 2.4$ s$^{-1}$. Spectral expansion of the transition matrix was used in this simulation.

The ryanodine receptor channel in bilayers displays unusual kinetics (1). When Ca is elevated, the channels open and then appear to inactivate. However, in contrast to traditional inactivation such as observed in the acetylcholine receptor, when Ca is further increased the channels reopen and again appear to inactivate. Thus the channel acts as a differentiator, responding primarily to changes in concentration. To avoid confusion with the term inactivation, this reactivatable kind of behavior is termed adaptation.

In adaptive behavior, there is a peak in the response associated with every increase in ligand concentration, and the steady-state response may be independent of the ligand concentration (Fig. 1). Adaptive behavior has been analyzed in the context of bacterial chemoreception (2, 3), and the results are readily transferred to channels. The reaction can be described by a simple Markov scheme in thermal equilibrium.

Following the notation of Segel et al. (3), we consider the reaction (Fig. 2) where $L$ represents the ligand, states X and D are open, states R and Y are shut, and the rate constants are as indicated. Segel et al. (3) assume for simplicity that the binding reactions, $R+X$ and $D+Y$ are fast compared to the allosteric reactions, $R=D$ and $X=Y$. However, the characteristic adaptive behavior is not dependent upon this assumption.

There are three main conditions to be satisfied by parameters of the model, (i) that the system adapts, (ii) that the peak activity in response to a step input is significantly greater than the steady-state activity, and (iii) that the system be in thermal equilibrium, that is, satisfy detailed balance. In the most extreme case, called exact adaptation (Fig. 1), the steady-state activity is independent of ligand concentration. We have made the additional assumption that the channel has only two conductances, open and closed. One can show that,

1) If the channel is mostly closed in the absence of ligand, then R and Y must be shut and D and X open.

2) For exact adaption, $K_1 K_2 = 1$, where $K_1 = k_{-2}/k_2$ and $K_2 = k_{-3}/k_3$. For weaker adaptation, that is where the steady-state activity increases with ligand concentration, $K_1 K_2 > 1$, and where it decreases with concentration, $K_1 K_2 < 1$.

3) For microscopic reversibility, $K_1 K_D = K_2 K_R$, where $K_R = k_{-1}/k_1$ and $K_D = k_{-4}/k_4$. The peak current following a step

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**References and Notes**

4. M. D. Stern, personal communication.
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