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Analysis of Neuron Models with Dynamically Regulated Conductances

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We analyze neuron models in which the maximal conductances of membrane currents are slowly varying dynamic variables regulated by the intracellular calcium concentration. These models allow us to study possible activity-dependent effects arising from processes that maintain and modify membrane channels in real neurons. Regulated model neurons maintain a constant average level of activity over a wide range of conditions by appropriately adjusting their conductances. The intracellular calcium concentration acts as a feedback element linking maximal conductances to electrical activity. The resulting plasticity of intrinsic characteristics has important implications for network behavior. We first study a simple two-conductance model, then introduce techniques that allow us to analyze dynamic regulation with an arbitrary number of conductances, and finally illustrate this method by studying a seven-conductance model. We conclude with an analysis of spontaneous differentiation of identical model neurons in a two-cell network.

1 Introduction

Mathematical models based on the Hodgkin–Huxley approach (Hodgkin and Huxley 1952) describe active neuronal conductances quite accurately over time scales ranging from milliseconds to several seconds. Model neurons constructed from these descriptions (see for example Koch and Segev 1989) exhibit a wide variety of behaviors similar to those found in real neurons including tonic spiking, plateau potentials and periodic bursting. However, neuronal conductances can change over longer time scales through additional processes not modeled by the Hodgkin–Huxley equations. These include modification of channel structure and/or density through biochemical pathways involving protein phosphorylation (Kaczmarek 1987; Chad and Eckert 1986; Kaczmarek and Levitan 1987) and gene expression (Morgan and Curran 1991; Sheng and Greenberg 1990; Smeyne *et al.* 1992). These processes can be activity dependent. For

example, when rat myenteric neurons are chronically depolarized they show decreased calcium currents (Franklin *et al.* 1992). Electrical activity can induce the expression of immediate-early genes like *fos* over a period of about 15 min (Morgan and Curran 1991; Sheng and Greenberg 1990; Smeyne *et al.* 1992) and expression of the immediate-early gene *ras* has been associated with an increased potassium conductance (Hemmick *et al.* 1992). From these studies it is clear that the biochemical processes that affect membrane conductances act on many different time scales. Relatively fast effects, such as the voltage and calcium dependence of channel conductances, are included in the usual Hodgkin–Huxley descriptions. However, activity-dependent modifications of membrane currents due to slower and less direct processes are not.

Unfortunately, there is not enough information available at the present time to build a detailed model of the biochemical processes producing slow modification or, as we will call it, regulation of membrane conductances. However, we feel that it is not too early to try to assess what impact such a process might have on the behavior of neurons and neural networks. To do this we have constructed a simple phenomenological model with slowly varying, dynamically regulated conductances and studied its behavior using computer simulation (LeMasson *et al.* 1992). The model reveals several interesting features:

- Starting from a wide variety of initial conductances, the model neurons can automatically develop the currents needed to produce a particular pattern of electrical activity.

- Slow regulatory processes can significantly enhance the stability of the model neuron to environmental perturbations such as changes in the extracellular ion concentrations.

- The intrinsic properties of model neurons are modified by sustained external currents or synaptic inputs.

- In simple networks, model neurons can spontaneously differentiate developing different intrinsic properties and playing different roles in the network.

These features have obvious implications for the development and plasticity of neuronal circuits. Our previous work (LeMasson *et al.* 1992) relied solely on computer simulation involving a fairly complex neuronal model. In this paper we devise a general procedure for analyzing the process of dynamic regulation. We will examine the properties listed above in detail both for a simple neuron model and for the more complex model considered previously.

2 A Model of Dynamic Regulation

We consider a single compartment, conductance-based neuron model with the membrane potential V determined by the basic equation

$$C\frac{dV}{dt} = -\sum_i I_i \tag{2.1}$$

C is the membrane capacitance and I_i are the membrane currents, which are written in the form (Hodgkin and Huxley 1952; see Koch and Segev 1989)

$$I_i = \bar{g}_i m_i^{p_i} h_i^{q_i} (V - E_i) \tag{2.2}$$

where E_i is the equilibrium potential corresponding to the particular ion producing the ith current, p_i and q_i are integers, and $\bar{g}_i$ is the maximal conductance for the current i. The dynamic variables m_i and h_i are determined by first-order, differential equations linear in m_i and h_i but with nonlinear voltage-dependent coefficients,

$$\frac{dm_i}{dt} = \alpha_{m_i}(V)(1 - m_i) - \beta_{m_i}(V)m_i \tag{2.3}$$

and

$$\frac{dh_i}{dt} = \alpha_{h_i}(V)(1 - h_i) - \beta_{h_i}(V)h_i \tag{2.4}$$

These equations describe the voltage-dependent characteristics of the conductance. Calcium dependent properties can be included by allowing α and β to depend on the intracellular calcium concentration as well as on the voltage.

In conventional Hodgkin–Huxley type models, the maximal conductances $\bar{g}_i$ are fixed constants. However, these are likely candidates for the slow modulation that we refer to as dynamic regulation. This is because the maximal conductance of a given current is the product of the conductance of an individual membrane channel times the density of channels in the membrane. Any slow process that alters the conductance properties of the channel or adds or removes channels from the membrane will affect $\bar{g}_i$. These slow, regulatory processes can be included in the model by making the maximal conductances dynamic variables instead of fixed parameters (LeMasson *et al.* 1992). Regulatory mechanisms could also, in principle, modify the kinetics of channel activation and inactivation, but we will not consider this possibility here.

To construct a model with dynamic regulation, we need to describe a mechanism by which the activity of a neuron can affect the maximal conductances of its membrane currents. Numerous possibilities exist including modified rates of channel gene expression, structural modifications of the channels either before or after insertion into the membrane,

and changes in the rates of insertion or degradation of channels. These (and many other) processes often depend on the intracellular calcium concentration (Kennedy 1989; Rasmussen and Barrett 1984; Sheng and Greenberg 1990; Murphy *et al.* 1991). For example, activity-dependent expression of immediate early genes has been linked to an elevation in calcium levels due to influx through voltage-dependent calcium channels (Murphy *et al.* 1991) and calcium is implicated in many other examples of slow, activity-dependent modulation (Kennedy 1989; Rasmussen and Barrett 1984; Sheng and Greenberg 1990). In addition, the intracellular calcium concentration is highly correlated with the electrical activity of the neuron (Ross 1989; LeMasson *et al.* 1992). For these reasons, we use the intracellular calcium concentration as the feedback element linking activity to maximal conductance strengths (LeMasson *et al.* 1992).

Since the maximal conductances $\overline{g}_i$ depend on both the number and properties of the membrane channels, their values will be affected by the processes outlined above. If these processes are regulated by calcium the values of the maximal conductances will also depend on the intracellular calcium concentration. We will assume that the kinetics is first-order and that both the equilibrium values of the maximal conductances and the rate at which they approach the equilibrium value may be calcium dependent. As a result, the behavior of the maximal conductances $\overline{g}_i$ is described by the equations

$$\tau_i([\mathrm{Ca}])\frac{d\overline{g}_i}{dt} = F_i([\mathrm{Ca}]) - \overline{g}_i \tag{2.5}$$

where [Ca] is the intracellular calcium concentration. At fixed intracellular calcium concentration [Ca], the maximal conductance $\overline{g}_i$ will approach the asymptotic value F_i[Ca] over a time of order τ_i[Ca]. If the calcium concentration changes the maximal conductances will also change their values. This regulation is a slow process occurring over a time ranging from several minutes to hours. This time scale distinguishes the calcium regulation of equation 2.5 from the more familiar and rapid calcium dependence of currents like the calcium-dependent potassium current.

The full neuron model with dynamic regulation of conductances is described by equations 2.1–2.5 and an equation for the intracellular calcium concentration [Ca]. For the model to work, it is crucial that one of the membrane currents $I_i = I_{\mathrm{Ca}}$ be a voltage-dependent calcium current because this is what links the intracellular calcium concentration to activity. We will assume that entry through voltage-dependent calcium channels is the only source of intracellular calcium and will not consider release from intracellular stores. Calcium is removed by processes that result in an exponential decay of [Ca]. Thus, [Ca] is described by the equation

$$\frac{d[\mathrm{Ca}]}{dt} = -k(AI_{\mathrm{Ca}} - [\mathrm{Ca}]) \tag{2.6}$$

The constant A depends on the ratio of surface area to volume for the cell. We typically use a value between 1/(100 msec) and 1/sec for the constant k controlling the rate of calcium buffering.

To complete the model we must specify the functions $\tau_i([\mathrm{Ca}])$ and $F_i([\mathrm{Ca}])$ appearing in equation 2.5. As in our previous work (LeMasson *et al.* 1992) we are guided in the choice of these functions by considerations of simplicity and stability. We are primarily interested in the equilibrium behavior of the regulated model. Because of this, we can simplify equation 2.5 by setting all the time constants equal and making them calcium-independent,

$$\tau_i([\mathrm{Ca}]) = \tau \tag{2.7}$$

where τ is a constant independent of [Ca]. This simplification has no effect on the equilibrium behavior of the model. In our simulations, we have taken the time constant τ to vary from 1 to 50 sec. We expect real regulatory processes to be considerably slower than this. However, the only condition on the model is that τ be much longer than the time scales associated with the membrane currents so we have accelerated the regulatory process to speed up our simulations.

The functions F_i determine how the asymptotic values of the maximal conductances depend on the calcium concentration. We assume that the regulation mechanism can vary the maximal conductances $\bar{g}_i$ over a range $0 < \bar{g}_i < G_i$ where G_i is the largest value that $\bar{g}_i$ can possibly take. In addition, a given maximal conductance can either increase or decrease as a function of the intracellular calcium concentration. These considerations lead us to consider just two possible forms (up to an overall constant) for the F_i, either a rising or a falling sigmoidal function,

$$F_i([\mathrm{Ca}]) = G_i\sigma\left(\pm\frac{C_T - [\mathrm{Ca}]}{\Delta}\right) \tag{2.8}$$

where G_i, C_T, and Δ are constants and σ is the standard sigmoidal function

$$\sigma(x) = \frac{1}{1+\exp(-x)} \tag{2.9}$$

In equation 2.8, the parameter G_i sets the scale for the particular maximal conductance $\bar{g}_i$. C_T determines the concentration at which the asymptotic value of $\bar{g}_i$ is $G_i/2$ and Δ sets the slope of the sigmoid. The choice of the plus or minus in equation 2.8 determines whether $\bar{g}_i$ will fall or rise as a function of [Ca].

The slow regulatory processes we are modeling must not destabilize the activity of the neuron. To assure stability of the neuron, the choice of the plus or minus sign in equation 2.8 must be made correctly. Suppose that a specific set of maximal conductances has been established producing a certain level of electrical activity. If the neuron becomes more active

than this level, calcium entering through voltage-activated channels will raise the intracellular calcium concentration. Under these conditions, outward currents should increase in strength and inward currents decrease so that the activity of the neuron will be reduced back to the original level. Conversely, if the activity level drops, the calcium concentration will also fall. In this case, the inward currents should increase in strength and the outward currents should decrease. In other words, the feedback from activity to maximal conductances should be negative. To assure this we use the plus sign in equation 2.8 for inward currents and the minus sign for outward currents. With this sign convention increased calcium results in an increase of the outward and a decrease of the inward currents while decreased calcium has the opposite effect. With the choices we have made, the evolution of the maximal conductances is given by

$$\tau \frac{d\bar{g}_i}{dt} = G_i \sigma \left(\pm \frac{C_T - [Ca]}{\Delta} \right) - \bar{g}_i \tag{2.10}$$

where the variable sign is plus for inward currents and minus for outward currents. Because the intracellular calcium concentration depends on the maximal conductances, these are highly nonlinear equations.

The parameter C_T in equation 2.10 plays the role of a target calcium concentration. If [Ca] is well below C_T, activity will increase due to the enhancement of inward and depression of outward currents. This will bring [Ca] up closer to the target value C_T. If [Ca] is well above C_T, there will be an opposite effect on the currents and [Ca] will drop toward C_T. Since the electrical activity of the neuron is highly correlated with the intracellular calcium concentration, stabilization of the intracellular calcium concentration results in a stabilization of the electrical activity of the neuron. As we will see, there is a direct connection between the target calcium concentration C_T and the activity level maintained by the model neuron.

Even without the dynamic regulation we have added, conductance-based neuronal models tend to be quite complex. However, the model specified above can be analyzed in considerable detail because of the large difference between the rates of the slow regulatory processes described by equations 2.10 and the faster processes of equations 2.1–2.4 and 2.6.

3 A Two-Conductance Model

The simplest model we will use to study dynamic regulation of conductances is the Morris–Lecar model (Morris and Lecar 1981), which has one inward and one outward active current. The inward current is a calcium current given (using the parameters we have chosen) by

$$I_{Ca} = \bar{g}_{Ca} \left[\sigma \left(\frac{V+1}{7.5} \right) + 0.1 \right] (V - E_{Ca}) \tag{3.1}$$

and the outward current is a potassium current,

$$I_K = \bar{g}_K n(V - E_K) \tag{3.2}$$

with n given by

$$\left\{\frac{3}{\cosh[(V-10)/29]}\right\}\frac{dn}{dt} = \sigma\left(\frac{V-10}{7.25}\right) - n \tag{3.3}$$

In addition, there is a passive leakage current

$$I_L = 0.5(V + 50) \tag{3.4}$$

and we will sometimes add an external current as well. In these equations, V is measured in millivolts and time in milliseconds. Under control conditions, we take $E_{Ca} = 100$ mV and $E_K = -70$ mV although we will vary these parameters to simulate changes in the extracellular ion concentrations. We have added a persistent component (the 0.1 in equation 3.1) to the calcium current, which is not present in the original model (Morris and Lecar 1981). This is useful in the regulated model because calcium provides the feedback signal for the regulation process. Without a persistent component, loss of the calcium current would mean a loss of this signal. We take $C = 1\ \mu F/cm^2$, $G_{Ca} = 3$ mS/cm^2, and $G_K = 6$ mS/cm^2. The behavior of this model neuron for the control values of the parameters is shown in Figure 1A.

In the two-conductance model, the maximal conductances $\bar{g}_{Ca}$ and $\bar{g}_K$ are regulated by equations like 2.10, specifically

$$\tau\frac{d\bar{g}_{Ca}}{dt} = G_{Ca}\sigma\left(\frac{C_T - [Ca]}{\Delta}\right) - \bar{g}_{Ca} \tag{3.5}$$

and

$$\tau\frac{d\bar{g}_K}{dt} = G_K\sigma\left(\frac{[Ca] - C_T}{\Delta}\right) - \bar{g}_K \tag{3.6}$$

We wish to analyze the dynamics of these two maximal conductances. Dividing the first equation by G_{Ca} and the second by G_K we find that the quantities $\bar{g}_{Ca}/G_{Ca}$ and $\bar{g}_K/G_K$ obey very similar equations. By adding the resulting two equations and using the identity $\sigma(x) + \sigma(-x) = 1$ we find that the quantity y defined by

$$y = \frac{\bar{g}_{Ca}}{G_{Ca}} + \frac{\bar{g}_K}{G_K} \tag{3.7}$$

obeys the trivial equation

$$\tau\frac{dy}{dt} = 1 - y \tag{3.8}$$

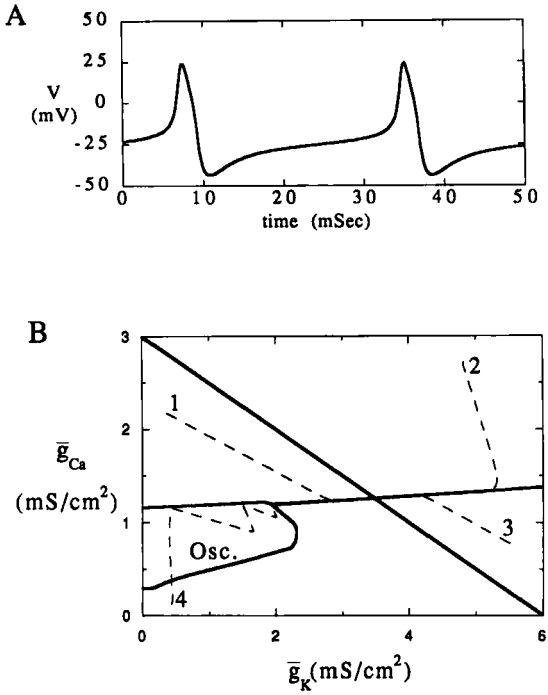


Figure 1: (A) Membrane potential versus time at the quasi-steady-state point for the two-conductance model. (B) Maximal conductance "phase-plane" for the two-conductance model. Straight lines are nullclines of the slow, regulatory dynamics. Region marked Osc. is where oscillations of the regulatory system occur. The quasi-steady-state is where the two nullclines cross. Dashed paths marked 1–4 show routes to the steady-state point from four different starting conditions. For convenience (and without loss of generality) we have chosen the units of [Ca] so that the coefficient A in equation 2.6 is one. In these units $C_T = 20$ and $\Delta = 5$. In addition, we take $k = 1/(100$ msec). These parameters are used for Figures 2-4 as well (except that C_T is varied in Fig. 3).

Likewise taking the difference of these two equations and defining

$$z = \frac{\bar{g}_{Ca}}{G_{Ca}} - \frac{\bar{g}_K}{G_K} \tag{3.9}$$

we find that

$$\tau \frac{dz}{dt} = \tanh\left(\frac{C_T - [Ca]}{2\Delta}\right) - z \tag{3.10}$$

Using equations 3.8 and 3.10, we can completely analyze the behavior of the model in the "phase-plane" of maximal conductances $\bar{g}_{Ca}$ and $\bar{g}_K$. First, there is a nullcline $y = 1$ or equivalently

$$\frac{\bar{g}_{Ca}}{G_{Ca}} + \frac{\bar{g}_K}{G_K} = 1 \tag{3.11}$$

from equation 3.8 and this is approached exponentially with time constant τ. The behavior of the z variable is more complex. Under some conditions, z will approach a quasi-equilibrium state. An equilibrium solution of equation 3.10 would occur when $z = \tanh[(C_T - [Ca])/2\Delta]$. However, if this value of z results in oscillatory behavior of the model neuron the calcium concentration [Ca] will oscillate as well. Thus, this value of z will not truly be fixed. We can circumvent this complication because we are assuming that the time scale τ governing the motion of z is much greater than the time scale of the membrane potential oscillations. Although z will oscillate around the quasi-equilibrium value, if τ is large these oscillations will be very small. The quasi-equilibrium value of z is just the average value of the hyperbolic tangent

$$z = \left\langle \tanh\left(\frac{C_T - [Ca]}{2\Delta}\right)\right\rangle \tag{3.12}$$

where the brackets denote a time average over many membrane potential oscillation cycles. Equation 3.12 defines an approximate nullcline for the dynamics of the z variable for the maximal conductances.

In Figure 1B, the solid lines indicate the nullclines 3.11 and 3.12 for the regulatory dynamics. The diagonal line with negative slope is the y nullcline $g_{Ca}/G_{Ca} + g_K/G_K = 1$ while the more horizontal line is the z nullcline. In the center of the figure, where the two nullclines cross, is the quasi-steady-state point of the full system which results in the behavior seen in Figure 1A. This point is stable and its domain of attraction is the entire plane. There is a region of the plane (at the lower left of Fig. 1B) where z does not approach quasi-steady-state behavior at fixed y but instead goes into oscillations with a period of order τ. In this area there is, of course, no z nullcline. Instead, we have drawn the upper and lower bounds of the region over which the oscillations in z take place. Regions like this provide an interesting mechanism for generating rhythms with very long periods such as circadian rhythms. These slow oscillations arise from the regulatory process interacting dynamically with the more conventional mechanisms producing the much faster membrane potential oscillations.

The dynamically regulated model can spontaneously construct its conductances starting from any initial values of $\bar{g}_{Ca}$ and $\bar{g}_K$. The dashed curves in Figure 1B show the approach to steady-state behavior from four different sets of initial conductances. There are no obstructions to the recovery of the quasi-steady-state values from any initial position in the plane.

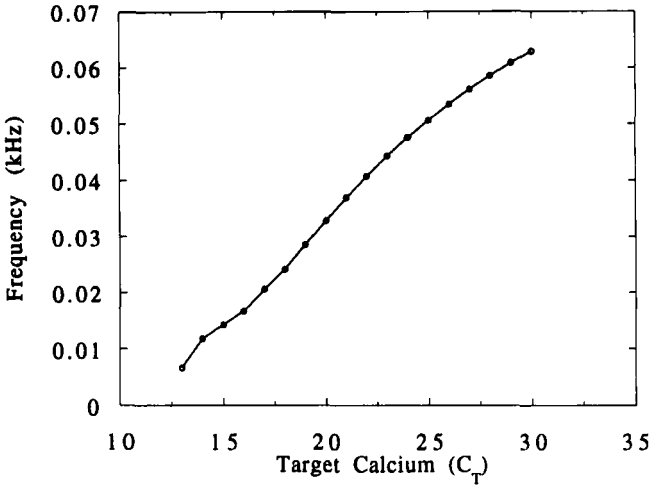


Figure 2: The range of steady-state oscillation frequencies that can be obtained using different values of the target calcium concentration C_T. Units of calcium concentration are as in Figure 1.

In the usual, unregulated, conductance-based models, the values of the maximal conductance parameters determine the behavior of the model neuron. In the regulated model, the maximal conductances are dynamic variables and, instead, the behavior of the model is governed by the parameters C_T and Δ that control the quasi-steady-state values of the maximal conductances. Of these, C_T is by far the more important parameter. By adjusting the value of this target calcium concentration, we can determine what sort of behavior the neuron will exhibit. In contrast to conventional models, once this value is chosen the desired behavior will be exhibited over a variety of external conditions. In Figure 2, we see that a wide range of oscillation frequencies can be obtained in the regulated, two-conductance model by choosing different values for the target calcium concentration C_T without changing any other parameters of the model.

The stabilizing effects of dynamic regulation are illustrated in Figure 3. When dynamic regulation is not included in the model, the firing frequency is extremely sensitive to the values of E_{Ca} and E_K and firing only occurs over a limited range of these parameters. With dynamic regulation, stable firing at roughly the same frequency can be maintained over a wide range of E_{Ca} and E_K. Since these parameters are affected by the extracellular ionic concentrations, this reflects the ability of a dy-

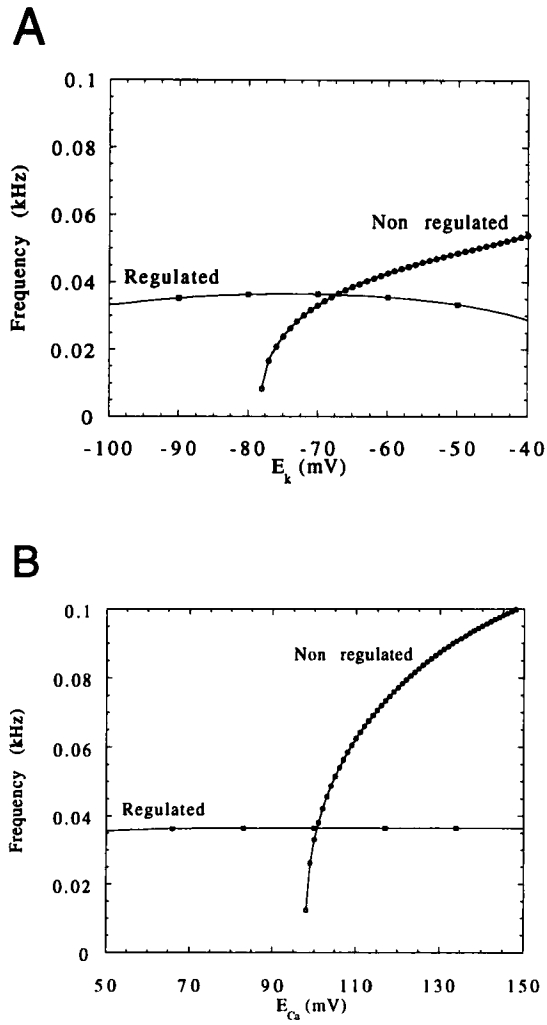


Figure 3: The dependence of oscillation frequency on the equilibrium potentials for (A) potassium and (B) calcium in the regulated and unregulated two-conductance models. Dynamic regulation stabilizes the frequency against changes in E_K and E_{Ca}. For the unregulated case, we fix the maximal conductances at the control values for the unregulated model.

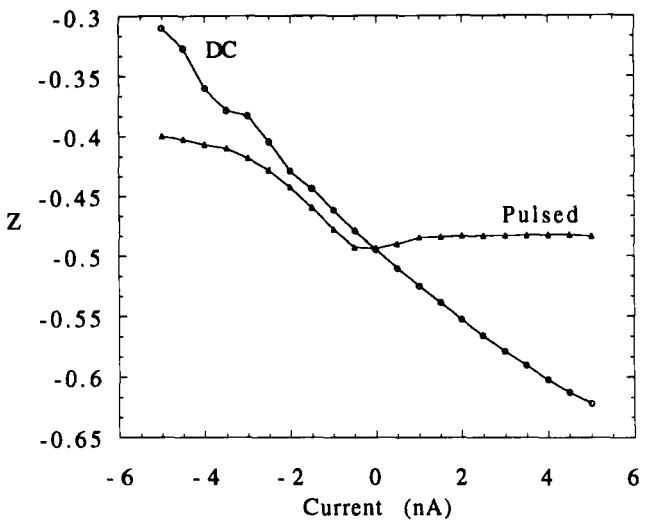


Figure 4: The quasi-steady-state value of z as a function of the amplitude of an injected current. Both DC and pulsed injection cause shifts in the value of z that modify the balance between inward and outward currents and change the intrinsic properties of the model neuron. Pulses last for 250 msec and are repeated every 500 msec.

namically regulated neuron to adjust to varying external conditions. The model maintains its firing frequency by shifting its maximal conductances in response to changes of these parameters. This is done through shifts in the value of z, which change the balance of inward and outward currents.

Dynamically regulated neurons also exhibit activity-dependent shifts in their intrinsic characteristics. As we have seen, the regulatory mechanism tends to stabilize the activity of the neuron by shifting the values of the maximal conductances to maintain the level of activity that results in an average intracellular calcium concentration near the target value C_T. The introduction of external or synaptic inputs will likewise cause slow shifts in the values of the maximal conductances as the regulatory mechanism tries to maintain the same level of calcium and activity that existed in the absence of inputs. As a result, prolonged inputs cause changes in the intrinsic characteristics of the neuron. This is shown in Figure 4 where we investigate the effect of external current on a regulated model neuron. The external current causes a shift in the value of z which changes the intrinsic electrical properties of the neuron by modifying the

balance between inward and outward currents according to equation 3.9. The quasi-steady-state value of z depends not only on the amplitude of the applied current but also on its time course. As shown in Figure 4, DC current injection has a different effect than pulses of current and we have found that the shift in z is also sensitive to the frequency and duty cycle of the pulses, in particular, the relation of the pulse frequency to the natural frequency of the model. These shifts occur over a slow time scale. Thus, the regulated model neuron will respond normally to brief pulses of current. However, prolonged current injection or synaptic input will change intrinsic properties.

4 General Analysis

The type of analysis we performed for the two-conductance model in the last section can be extended to models with arbitrarily large numbers of conductances. The key observation is that when equation 2.10 is divided by G_i, all of the ratios $\bar{g}_i/G_i$ satisfy the same equation except for the plus and minus sign difference for inward and outward currents. This implies that the difference $\bar{g}_i/G_i - \bar{g}_j/G_j$ between any two outward or any two inward currents will go exponentially to zero with the time constant τ. Furthermore, the identity $\sigma(x) + \sigma(-x) = 1$ we used before implies that the sum $\bar{g}_i/G_i + \bar{g}_j/G_j$, where i is an outward current and j is an inward current, goes exponentially to one with the same time constant. As a result, we can write an explicit solution for all of the maximal conductances satisfying equation 2.10 expressed in terms of just one dynamic variable z,

$$g_i(t) = \frac{G_i}{2}[1 \pm z(t)] + c_i e^{-t/\tau} \tag{4.1}$$

where the plus/minus sign is for inward/outward currents and c_i are constants that determine the initial values of the maximal conductances $g_i(0)$. The remaining dynamic variable z obeys the same equation as before,

$$\tau \frac{dz}{dt} = \tanh\left(\frac{C_T - [Ca]}{2\Delta}\right) - z \tag{4.2}$$

Thus we have reduced the analysis of dynamic regulation in a model with any number of currents to the study of this single equation interacting with the rest of the model through the z dependence of [Ca].

As in the two-conductance case, there are two general types of behavior. First, the system can settle down to a quasi-steady-state as far as the slow dynamics is concerned. Again, although the membrane potential and calcium concentration may fluctuate (due to action potentials for example), there are no fluctuations over the time scale associated with dynamic regulation. These faster fluctuations have little effect on the slowly

varying maximal conductances. Alternately, the slow system may never settle down and oscillations or even chaotic behavior characterized by the slow time scale typical of regulatory processes may appear. Again, these can provide a model of circadian or other slow rhythms.

5 A Seven-Conductance Model

We have studied dynamic regulation in a more complex and realistic model, a variant of the model of Buccholtz *et al.* (1992) describing the LP neuron in the stomatogastric ganglion of the crab. This model has seven active conductances corresponding to Hodgkin–Huxley sodium and potassium currents, a slow and a fast A current, a calcium-dependent potassium current, a calcium current, and a mixed-ion current I_H. In addition, there is a passive leakage current. We allow all seven maximal conductances for the active currents to be modified by the calcium-dependent regulation scheme as described by equations 2.10.

Depending on the value of the target calcium concentration C_T, the regulated LP model can exhibit silent, tonic firing, bursting, or locked-up (permanently depolarized) behavior. Although the model has seven dynamic maximal conductance variables, we can analyze the regulatory dynamics quite simply by using the z variable defined in the last section. After the exponential terms in equation 4.1 get small, the maximal conductances will take values

$$\bar{g}_i = \frac{G_i}{2}(1 \pm z) \tag{5.1}$$

with z determined by equation 4.2. To study the behavior of z in this model, we plot dz/dt given by the right side of equation 4.2 as a function of z in Figure 5. We also note the type of activity displayed by the model neuron for different values of z. For this figure, we have chosen the target calcium concentration C_T so that the neuron exhibits bursting behavior once the z parameter has relaxed to the point where $dz/dt = 0$. The quasi-steady-state is given by the zero crossing in the center of the figure and it exhibits bursting behavior. In the bursting range, Figure 5 shows a double line because we have plotted both the maximum and minimum values of dz/dt. At a given z value (the quasi-steady-state value for example) dz/dt will oscillate rapidly between the two lines shown due to the bursting behavior. These oscillations are not the same as those shown in Figure 1. The oscillations in Figure 1 are slow and are caused by the regulatory mechanism itself, while the oscillations here are just the result of the normal bursting activity of the neuron.

In our previous work on this model (LeMasson *et al.* 1992) we observed an interesting phenomenon when two regulated neurons were electrically coupled. The techniques we have developed here allow us to explore this phenomenon more completely. The two-neuron circuit is shown in Figure 6. We start with two dynamically regulated model

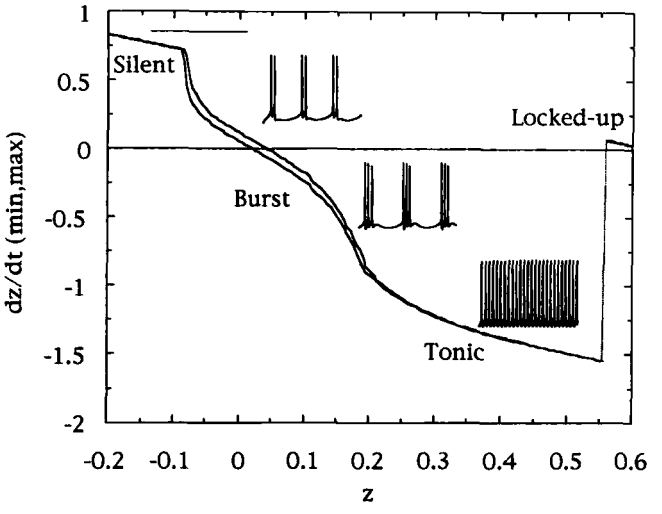
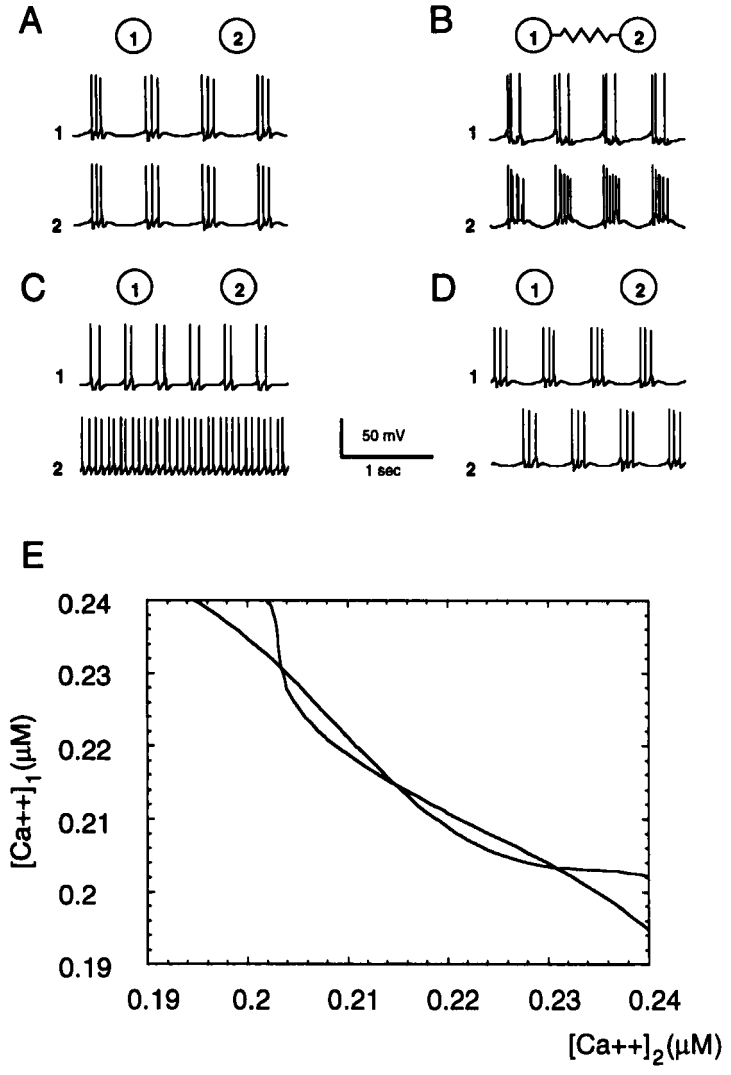


Figure 5: Plot of dz/dt versus z for the seven conductance model. Both minimum and maximum values of dz/dt at a given z value have been plotted. Two lines appear in the bursting region due to fluctuations in the calcium level during bursting activity. Distinct behaviors obtained for different values of z are indicated by the inserts. Locked-up refers to a permanently depolarized state. The quasi-equilibrium value of z produces bursting behavior as indicated by the zero crossing of dz/dt. Model parameters used for Figures 5 and 6 are as in LeMasson *et al.* (1992).

neurons described by identical sets of equations with the same parameter values. The identical activity of the two model neurons when they are uncoupled is shown in Figure 6A. The two neurons are then coupled through an electrical synapse (synaptic current proportional to the voltage difference between the two neurons) that is likewise completely symmetrical. Figure 6B shows the steady-state activity of the coupled network. The two neurons burst in unison. To examine the intrinsic properties of the two neurons individually, we uncouple them once again and show in Figure 6C their activity immediately after they are decoupled. Despite the fact that two model neurons are governed by identical sets of equations, the coupling between them has causes one neuron to display intrinsic bursting activity while the other fires tonically in isolation. The symmetric, two-cell network has spontaneously differentiated into a circuit involving a pacemaker and a follower neuron. If the two neurons are left uncoupled, the regulation process will eventually return them to their initial identical states as seen in Figure 6D.

To study this system, we monitor the maximal conductances of the two neurons and see how the coupling between them affects their behavior by performing the following numerical experiment. We hold z_1, the z value for one of the two neurons, fixed but allow z_2 to evolve according to equation 4.2 until it reaches its quasi-equilibrium value. This value will



depend on the fixed value z_1 we have chosen for the first neuron because the two neurons are coupled and this coupling effects the behavior of z_2 through effects like those shown in Figure 4. We then record the time-averaged intracellular calcium concentrations of the two neurons, $[Ca]_1$ and $[Ca]_2$. By repeating this process for many different holding values z_1 we obtain the curves shown in Figure 6E. Actually, only one of these curves corresponds to the procedure just outlined while the other is its reflection obtained by interchanging the roles of neuron 1 and neuron 2. One curve thus shows the quasi-equilibrium calcium concentrations of neuron 2 when neuron 1 is held fixed and the other the quasi-equilibrium concentrations of neuron 1 when neuron 2 is held fixed.

The values of z_1 and z_2 determine the maximal conductances of the two neurons through the relation 5.1 and this in turn will control their intracellular calcium concentrations. Because z and [Ca] are related, we can use either the value of z or the value of the intracellular calcium concentration to characterize the balance of inward and outward maximal conductances. Up to now, we have used z because it is directly related to the maximal conductances through equation 5.1. However, to illustrate the two-neuron network we use the time-average of the calcium concentration in the two neurons rather than their z values because the fluctuations caused by the bursting activity of the two neurons are smaller for the time-average calcium concentration making the plot clearer. Otherwise, the two approaches are completely equivalent.

The quasi-steady-state configurations of the fully regulated, interacting, two-neuron circuit are given by the points where the two curves in Figure 6E cross. The interesting feature of this particular network is that the lines cross in three places. The middle of these three crossings is the symmetric equilibrium point where the calcium concentrations, the z values, and the maximal conductances of the two neurons are identical. However, as is typical in cases of spontaneous symmetry breaking, this point is unstable for this particular network. The other two crossings are stable equilibrium points and they have the novel feature that the intrinsic conductances of the two neurons are different. One neuron exhibits a higher calcium concentration than the other so, according to equation 4.2, its z value will be lower than that of the other neuron. As a

Figure 6: *Facing page.* (A) The behavior of two identical model neurons before they are coupled. (B) Electrical coupling between the neurons results in a bursting two-cell network. (C) Decoupling the two neurons reveals their intrinsic properties and indicates that one is acting as a pacemaker and the other as a tonically firing follower. (D) Long after the two neurons are decoupled, the regulation mechanism has returned them to their original identical states. (E) A plot of the time-averaged calcium concentration of one neuron when the other neuron's regulation dynamics is held fixed. The three crossing points are equilibrium points. The central, symmetry crossing is unstable while the two outer crossing are stable quasi-steady-states with nonsymmetric properties.

result, one of the neurons will have smaller inward and larger outward conductances than the other neuron as given by equation 5.1. This is what causes the spontaneous differentiation of intrinsic properties seen in Figure 6C.

The symmetry-breaking phenomenon that we have discussed requires electrical coupling between the two neurons that lies in a specific range. The coupling must be strong enough so that the two neurons have an impact on each other, but not so strong that their activity is forced to be identical.

6 Discussion

We have used a single second messenger, the intracellular calcium concentration, to act as the negative feedback element linking the maximal conductances of a model neuron to its electrical activity. If similar mechanisms exist in real neurons they may be controlled by multiple second messengers. In addition, we have taken a particularly simple form of the regulatory equations by choosing a single sigmoidal curve (and its flipped version) for all of the conductances. What is surprising about these simplifications is that they nevertheless allow the full range of behaviors of the model neuron to be explored as seen in Figure 5. The parameterization of equation 5.1 may thus be useful even in cases where dynamic regulation is not being studied. Any scheme based on a single second messenger will similarly probe a single line in the multidimensional space of maximal conductance values characterizing a particular model. The simple form of the functions F_i we used means this line is given by the simple equation 5.1, more general forms of the F_i would result in more complex curves. Nevertheless, it should be possible to find a variable like z, even with nonidentical forms for the F_i, that parameterizes path length along this general curve. As a result, we expect that the behavior of the model in the more general case will be qualitatively similar to the simple case we have analyzed. This argument also applies to models in which some of the maximal conductances are not regulated at all.

We have thus far studied dynamic regulation as a global phenomenon in single compartment models. A local form of dynamic regulation could have important consequences in a multicompartment model of a neuron. In such a model, the density of channels in various parts of the neuron would be correlated with the time-average calcium concentration in that region. This provides a mechanism for controlling the distribution of conductances over the surface of a neuron (for a different approach to this problem see Bell 1991) and for correlating the local channel density with structural and geometrical characteristics affecting calcium buffering and diffusion (preliminary work done in collaboration with M. Siegel).

The dynamic regulation scheme was motivated by a need to build more robust neuronal models, and Figure 3 clearly shows that this goal

has been achieved. The fact that the dynamically regulated model also exhibits shifts in intrinsic characteristics due to interactions with other neurons is an interesting and unavoidable consequence of this robustness. If maximal conductances depend on activity, neurons in networks will be affected by each other and will adapt accordingly. Our two-neuron model resulted in an oscillating circuit with a pacemaker and a follower neuron. This differentiation was caused solely by the interaction of the two neurons. Either neuron could have developed into the pacemaker with the other becoming the follower. As in this simple example, it should be possible for identical dynamically regulated model neurons to self-assemble into more complex networks in which they play well-defined but different functional roles.

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