Neuron, Volume *82*

Supplemental Information

Cell Types, Network Homeostasis, and Pathological Compensation from a Biologically Plausible Ion Channel Expression Model

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Supplemental methods

Simulation details for example of integral controller (Figure 1)

In Figure 1B we numerically integrated the equations,

$$C\dot{V} = g_{\text{leak}}(E_{\text{leak}} - V) + g(E_g - V)$$

$$\tau_g \dot{g} = m - g$$

$$\tau_m \dot{m} = [\text{Ca}^{2+}] - \text{Ca}_{\text{tgt}}$$

for a single compartment with two leak conductances. Intracellular calcium was calculated from a monotonic increasing function of V:

$$[\text{Ca}^{2+}] = 109.2(\mu\text{M}) \times \exp(V/(12.5(\text{mV})))$$

Integration was performed using the ordinary differential equation solver in MATLAB with C = 1 nF, $\text{Ca}_{\text{tgt}} = 1\ \mu M$, $g_{\text{leak}} = 0.1$ µS, $\tau_m = 9.6 \times 10^5$ µM s µS^{-1}, $\tau_g = 3600\ s$, $E_{\text{leak}} = -85$ mV, $E_g = 50$ mV.

Simulation details for example of windup/mismatched targets (Figure 2)

In Figure 2 we numerically integrated the equations,

$$C\dot{V} = \sum_i g_i(E_i - V)$$

$$\tau_g \dot{g}_i = m_i - g_i$$

$$\tau_{m,i} \dot{m}_i = [\text{Ca}^{2+}] - \text{Ca}_{\text{tgt},i}$$

for a single compartment with two leak conductances. Intracellular calcium was calculated from a monotonic increasing function of V:

$$[\text{Ca}^{2+}] = \frac{20(\mu\text{M})}{1 + \exp\left[\frac{V}{10(\text{mV})}\right]}$$

In the first model we set $\text{Ca}_{\text{tgt},1} = \text{Ca}_{\text{tgt},2} = 3.5\ \mu M$. In the second model we set $\text{Ca}_{\text{tgt},1} = 3\ \mu M$ and $\text{Ca}_{\text{tgt},2} = 4\ \mu M$. All other model parameters were identical for the two cases: C = 1 nF, $\tau_{m,1} = 10$ µM s µS^{-1}, $\tau_{m,2} = -10$ µM s µS^{-1}, $\tau_g = 1$ s, $E_1 = -80$ mV, $E_2 = 10$ mV. Integration was performed using the ordinary differential equation solver in MATLAB.

Example of an integral control rule using biochemical reaction schemes (Results: equation 2 and following calculations)

Here we provide the details of a concrete example how integral control could be implemented biochemically using mass-action kinetics.

In the main text, we considered a "master regulator" T that provides a readout of Ca^{2+} error:

$$\left[\dot{T}\right] = k\left([\mathrm{Ca}^{2+}] - \mathrm{Ca}^{2+}_{\mathrm{targ}}\right).$$

Here, $[T]$ is the concentration of the regulatory enzyme/enzyme complex, k is a constant that scales the time constant of integration, $[\mathrm{Ca}^{2+}]$ is the intracellular calcium concentration, and $\mathrm{Ca}^{2+}_{\mathrm{targ}}$ is a positive constant that represents the target calcium concentration. This equation is a linear approximation of many potential underlying biochemical schemes. In particular, integral control will be provided by any system where $[\mathrm{Ca}^{2+}]$ in the above equation is replaced by a positive monotonic function of $[\mathrm{Ca}^{2+}]$. We derive what we consider to be the simplest such scheme in what follows in order to give a concrete example, but note that there are many other ways of achieving integral control (Drengstig et al., 2008; Yi et al., 2000).

The rate of change of concentration of a biochemical species can be written in terms of the difference between its production and degradation rates, which we label α and β respectively:

$$\left[\dot{T}\right] = \alpha - \beta$$

We next assume the rate of production of $[T]$ is Ca^{2+}-dependent. This can be captured by a straightforward reaction scheme in which the production rate, α, is proportional to the equilibrium fraction of a Ca^{2+}-bound factor, F. In this case, F could be a Ca^{2+}-dependent transcription factor that controls the production of T. If we consider simple first order kinetics for the Ca^{2+} binding reaction:

$$F + \mathrm{Ca}^{2+} \underset{k_b}{\overset{k_f}{\rightleftharpoons}} F\mathrm{Ca}^{2+}$$

and assume this reaction is at quasi-steady state (i.e. the binding and unbinding of Ca^{2+} is much faster than the rates for T), then α is given by a Hill equation:

$$\alpha([\mathrm{Ca}^{2+}]) = \frac{[F\mathrm{Ca}^{2+}]}{[F] + [F\mathrm{Ca}^{2+}]} = \frac{[\mathrm{Ca}^{2+}]}{K_d + [\mathrm{Ca}^{2+}]},$$

where $K_d = k_b/k_f$ (the dissociation constant). Throughout this analysis, we assume that the binding of Ca^{2+} to F (or any other molecule) does not alter the intracellular calcium concentration. This is valid when $[\mathrm{Ca}^{2+}] \gg [F]$, or when F is localized to a subcellular compartment whose $[\mathrm{Ca}^{2+}]$ is buffered by cytosolic Ca^{2+}.

As we stated in the main text, the fixed target is achieved in this model when the rate of degradation of T is zeroth-order. This can occur in a variety of ways (Drengstig et al., 2008). We will consider a scheme

involving a saturated degradation mechanism. Suppose T is degraded by another enzyme E according to Michaelis-Menten kinetics:

$$T + E \underset{k_2}{\overset{k_1}{\rightleftharpoons}} TE \xrightarrow{k_3} \emptyset + E.$$

This degradation scheme gives the following expression for β:

$$\beta = \frac{k_3\,[T][E]_{tot}}{K_m + [T]}\ .$$

Here, $[E]_{tot} = [E] + [TE]$, and is the total concentration of enzyme E; K_m is the Michaelis-Menten constant, $K_m = (k_3 + k_2)/k_1$. The above equation becomes zeroth-order in T, i.e. saturated when $K_m \ll [T]$. Then

$$\beta \approx k_3\,[E]_{tot}.$$

We now have the following differential equation for $[T]$:

$$\left[\dot{T}\right] = \alpha([\mathrm{Ca}^{2+}]) - k_3\,[E]_{tot}\ .$$

which is the required form for the integral control rule. In this equation the target Ca^{2+} level is given by $\mathrm{Ca}^{2+}_{\mathrm{targ}} = \alpha^{-1}(k_{cat}\,[E]_{tot}) = K_d k_3/(1 - k_3 E_{tot})$. Importantly, the error accumulated is of opposite sign on either side of $\mathrm{Ca}^{2+}_{\mathrm{targ}}$ because $\alpha([\mathrm{Ca}^{2+}])$ is monotonic.

Aggregating multiple steps in a biochemical scheme

We note here that in general there can be multiple steps between the activation of an ion channel gene and the resulting change in membrane conductance. In the model (equation 1, main text) we simplify this as two steps – 1) translation and 2) ion channel expression in the membrane:

$$\dot{m} = \alpha_m\,T - \beta_m m$$

$$\dot{g} = \alpha_g m - \beta_g g$$

Here the mRNA synthesis term $\alpha_m.T$ is the linear approximation of the quasi-steady state for the master regulator, T. These two steps correspond to rates of change of experimentally measurable variables (mRNA and membrane conductance). In general, we can consider a multi-step mass-action chain of arbitrary length:

$$\begin{aligned}
\dot{x}_1 &= \alpha_1\,T - \beta_1 x_1\\
\dot{x}_2 &= \alpha_2 x_1 - \beta_2 x_2\\
&\ldots\\
\dot{g} &= \alpha_N x_{N-1} - \beta_N g
\end{aligned}$$

Assuming the forward/backward reaction rates (α_i, β_i) at each step are fixed (or can be treated as

constant over a long time), then the asymptotic rate of change of g can be written in terms of an aggregate of all of the rate constants in the chain:

$$\dot{g} = \frac{\alpha_1 T \cdot \prod_{i=1}^{N} \frac{\alpha_i}{\beta_i} - g}{\sum_{i=1}^{N} \left[\prod_{j=i}^{N} \frac{\alpha_j}{\alpha_i \beta_j} \right]}.$$

Thus the contribution of the additional steps to long-term (quasi steady-state) behavior is to change the overall response rate of the system.

Parameter search method for CPG network (Figure 5)

Bursting neurons were identified from a random search of maximal conductances in a non regulating model. The conductances were drawn from a uniform distribution: gNa, gKd, gA, and gKCa were selected between 2.5 and 47.5 µS, while gCaT, gCaS, and gH were selected between 0.05 and 0.95 µS). From this, bursting neurons were chosen by analyzing inter-spike intervals and slow-wave amplitudes of membrane potential fluctuations. From these, three neurons were hand tuned to produce a triphasic rhythm by altering maximal conductances heuristically. The maximal conductances of the three candidate neurons were then scaled and converted to regulation rates by normalizing to the largest maximal conductance and scaling this to a timeconstant = 100 ms (τ_i in equation 3, main text). These rates were then randomly searched from a log-normal distribution in each regulation parameter with standard deviation of 50%. Leak reversal potentials, maximal conductances and calcium targets were searched at the same time from a normal distribution with 50% standard deviation around the candidate value. Each network was randomly initialized (see table S1 for initial condition range) and checked for triphasic activity at steady state. 15,000 random networks were searched in total. From these, the mean and covariance matrices of the distribution of parameters that produced triphasic networks was calculated and used to refine the search. The refined search drew parameters from a multidimensional normal distribution with the estimated covariance and mean; 113,000 sample networks were searched. From this, the parameter set that most reliably developed a triphasic rhythm from random initial conditions (see table S1) and recovered from the perturbation to PD (additional leak conductance = 0.02 µS, reversal potential = -80 mV) was chosen.

References from main text

Drengstig, T., Ueda, H.R., and Ruoff, P. (2008). Predicting perfect adaptation motifs in reaction kinetic networks. J Phys Chem B *112*, 16752-16758.
Yi, T.M., Huang, Y., Simon, M.I., and Doyle, J. (2000). Robust perfect adaptation in bacterial chemotaxis through integral feedback control. Proc Natl Acad Sci U S A *97*, 4649-4653.