Reconstructing Phasic Vasopressin Cells

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Introduction

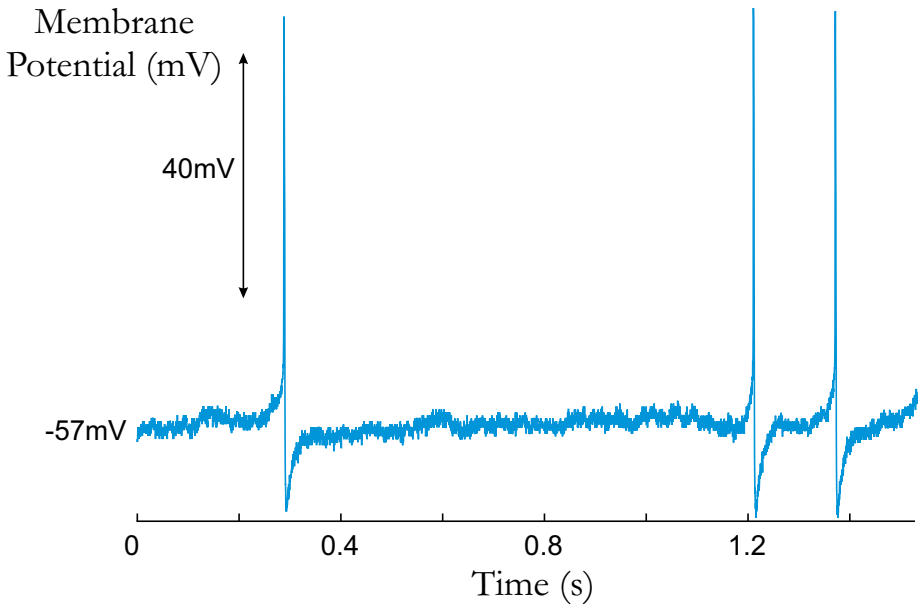
Arginine vasopressin (AVP) magnocellular neurosecretory cells (MNCs) are neurons that project from the hypothalamus to the pituitary. Their axons synapse directly onto capillaries and they secrete AVP directly into the blood. AVP controls osmolality and blood pressure and is released in response to dehydration and haemorrhage.

We present a mathematical model of an AVP cell and show how it reproduces

- controlled experimental protocols
- single action potentials and after-potentials
- the initiation of bursts
- burst termination
- the post-burst depolarized after-potential and slow depolarization

Action Potentials

Under normal conditions AVP cells fire action potentials (AP's) slowly and irregularly at < 10Hz.

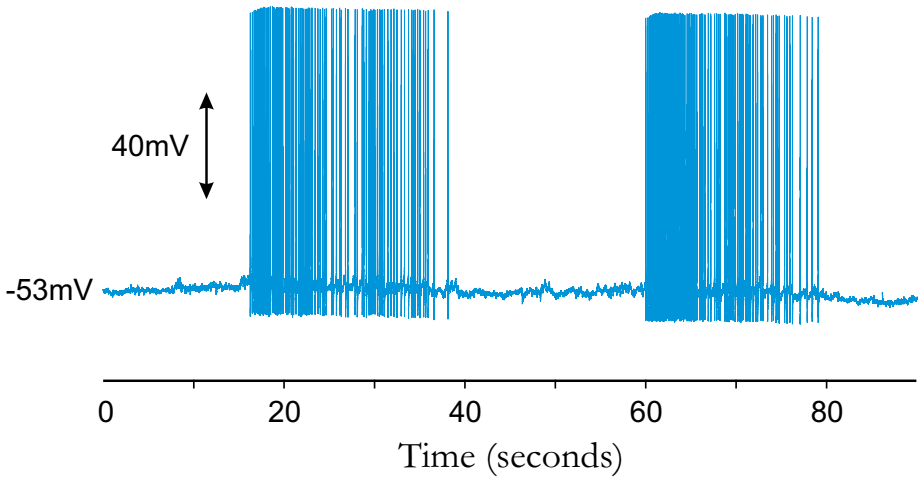


Spotaneously active, irregularly spiking cell *in vitro*. Sharp electrode recording from SON hypothalamic explant, courtesy of R. Teruyama, Memphis, TN.

Phasic Activity

When vasopressin release is strongly stimulated (*e.g.* by dehydration), AVP cells begin to fire phasically and emit long, repeating bursts of spikes that are separated by equally lengthy silences.

Whole cell patch clamp recording of an SON phasic neuron from hypothalamic slice *in vitro*, **courtesy of Chunyan Li, University of Tennessee, Memphis**



Mathematical Model

We model the electrical activity as a Hodgkin-Huxley type system with a simple calcium dynamics

$$\begin{aligned}\frac{\mathrm{d}V}{\mathrm{d}t} &= -\frac{I(t)}{C} \\ &= -\frac{1}{C}(I_{Na} + I_K + I_A + I_C + I_{AHP} + I_{Ca} + I_{leak})\end{aligned}$$

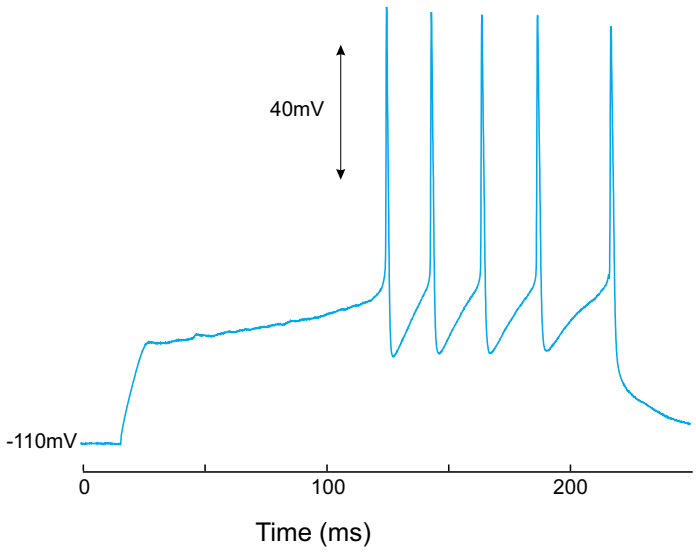
$$\frac{\mathrm{d}[\mathrm{Ca}^{2+}]_i}{\mathrm{d}t} = \alpha I_{Ca}(t) - \gamma\left([\mathrm{Ca}^{2+}]_i - [\mathrm{Ca}^{2+}]_{rest}\right)$$

and we have fitted parameters to reproduce the single action potentials and evoked activity that can be observed experimentally in MNC's.

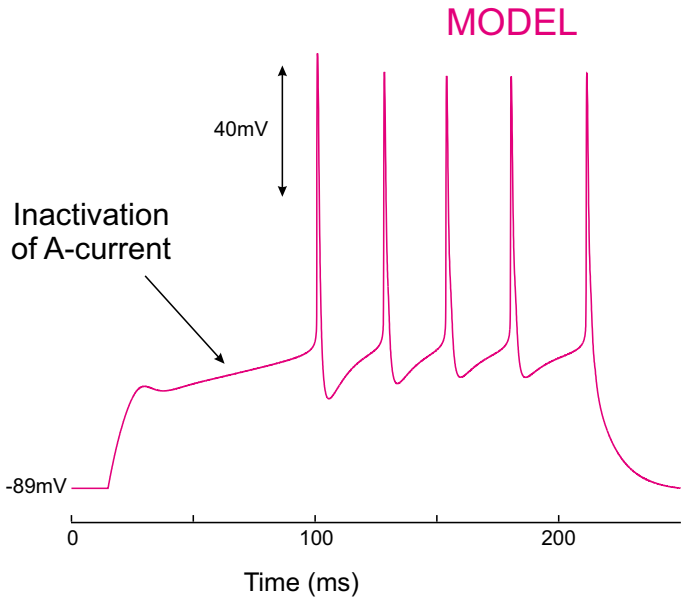
Evoked activity

When held at a hyperpolarised potential and given depolarizing pulses, MNC's display a latency to the first spike caused by the inactivation of an A-type current. Evoked trains of action potentials also undergo spike frequency adaptation which derives from the activation of a calcium dependent AHP current.

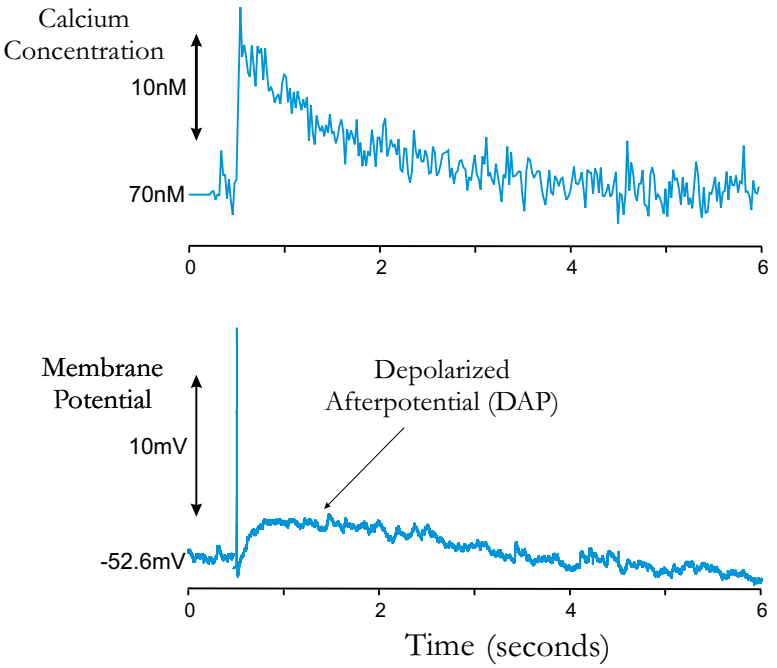
Whole cell recording from from SON neuron in hypothalamic explant. Cell first held at hyperpolarized potential to remove inactivation of A-current, then given 200ms depolarizing pulse. Note shoulder and subsequent slower excitation (following the initial steep rise), reflecting activation and inactivation of A-current. Courtesy of T. Shevchenko, Memphis, TN.



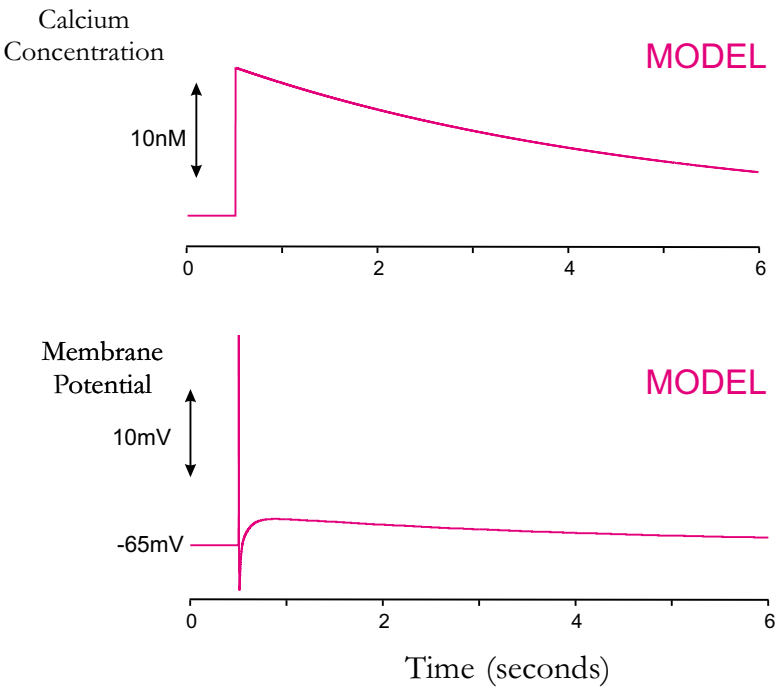
Model of above protocol. In what follows, experimental recordings will be shown in blue and modelling results will appear in red (unless otherwise stated) .



A depolarized after-potential (DAP) of duration $\sim$ 5s and amplitude $\sim$ 3mV follows each spike. The DAP depends strongly upon calcium and its time course matches $[Ca^{2+}]_i$ decay. Summation of successive DAP's is thought to underly phasic activity when these cells are strongly stimulated.



DAP and associated calcium transient recorded from MNC soma, average of 3 spikes. Electrical activity: whole cell recording from SON hypothalamic explant; Calcium concentration imaged with FURA-2. Spike amplitude has been trimmed for clarity.



Modelled DAP and associated calcium transient, spike amplitude has been trimmed for clarity.

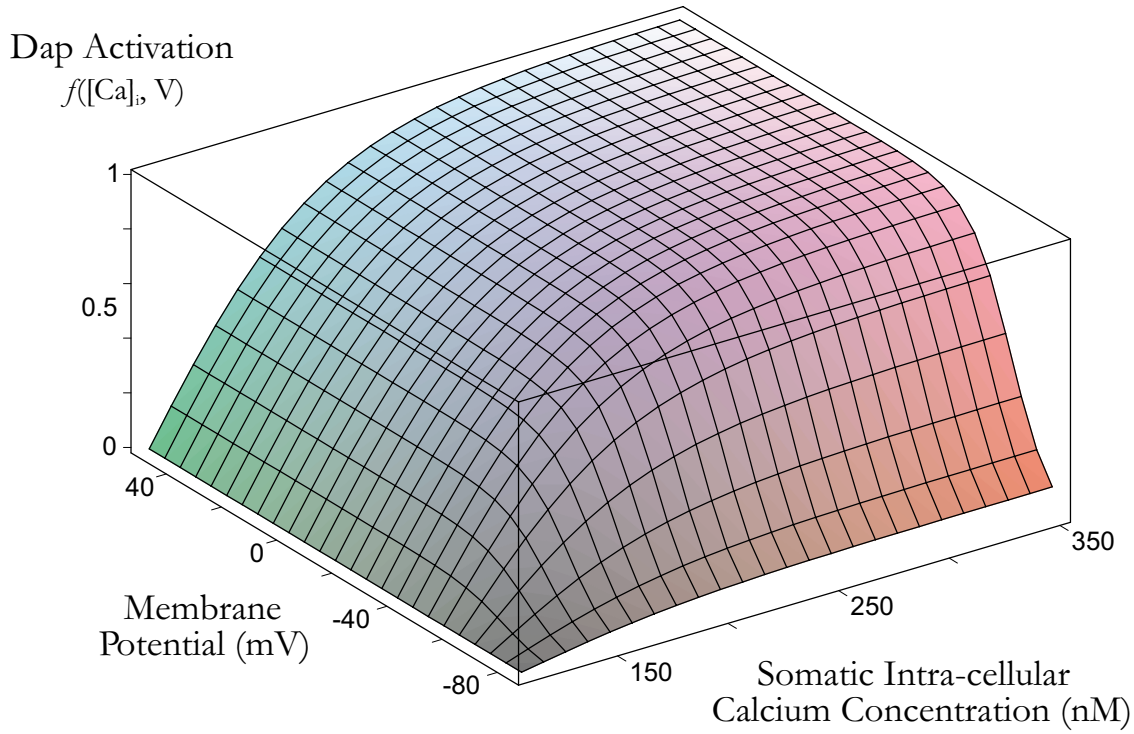
The DAP is thought [Li & Hatton, 1997] to derive from the calcium-mediated inactivation of a resting K^+ channel (*e.g.* TASK-1). We write

$$I_{leak} = I_{Na,leak} + I_{K,leak}$$

We propose that $I_{K,leak}$ has an inactivation function $f([Ca^{2+}]_i, V)$, which can be thought of as an activation function for the DAP ,

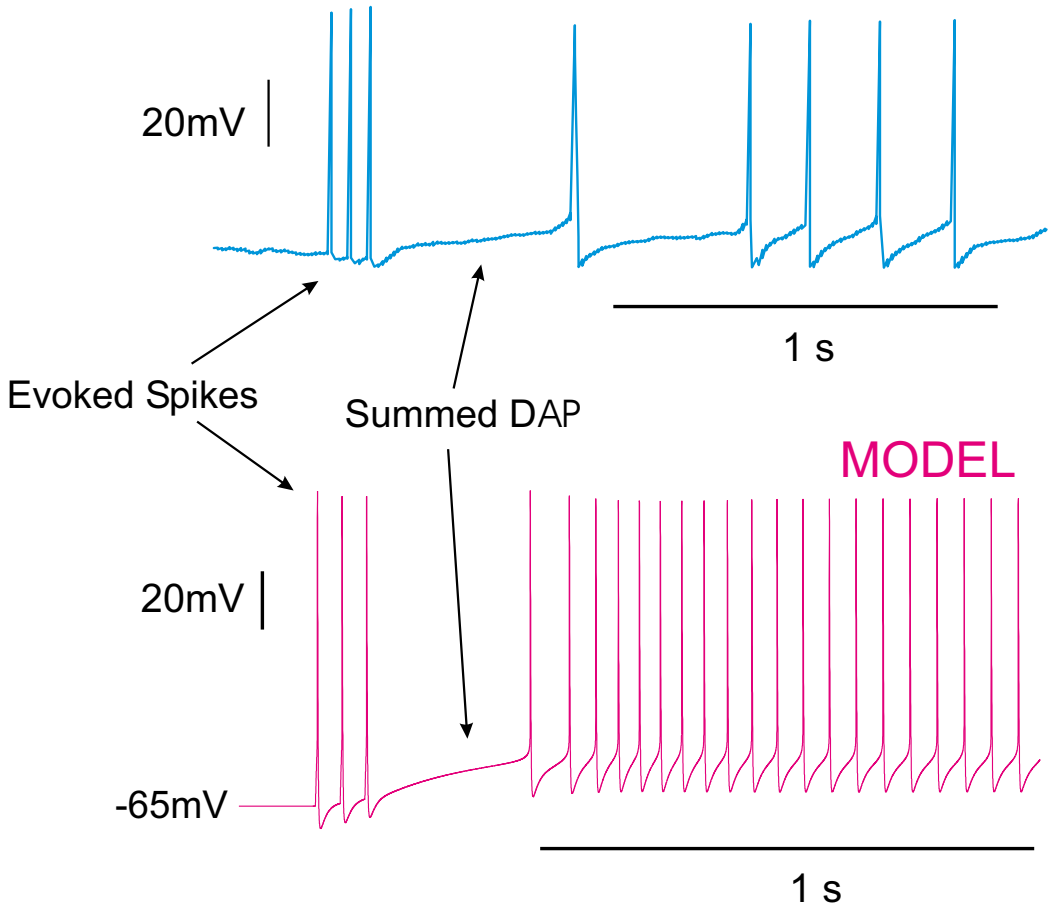
$$I_{K,leak} = G_{K,leak}(1 - f)[V - E_K]$$

We assume that $f([Ca^{2+}]_i, V)$ has the form:



so that calcium influx from each spike increases f, which inhibits $I_{K,leak}$ and so depolarizes the cell during the time that $[Ca^{2+}]_i$ decays to rest.

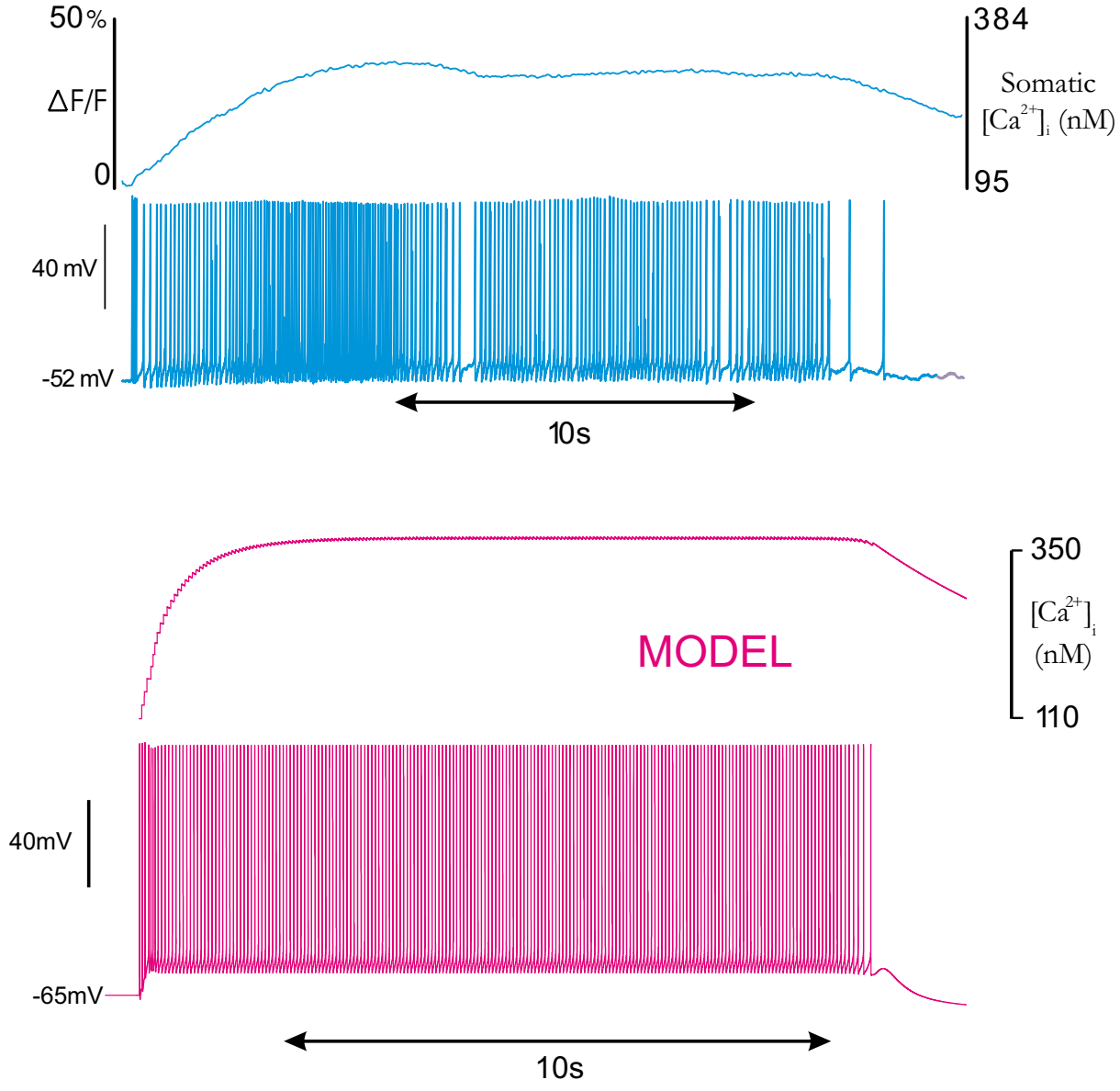
Bursts are initiated by the summation of several DAP's into a plateau potential which depolarizes the cell into a regularly firing regime.



Burst initiation by the summation of DAP's into a plateau potential, following 3 evoked spikes. *Top*: **Whole cell recording from SON hypothalamic explant;** *Bottom*: **Model results.**

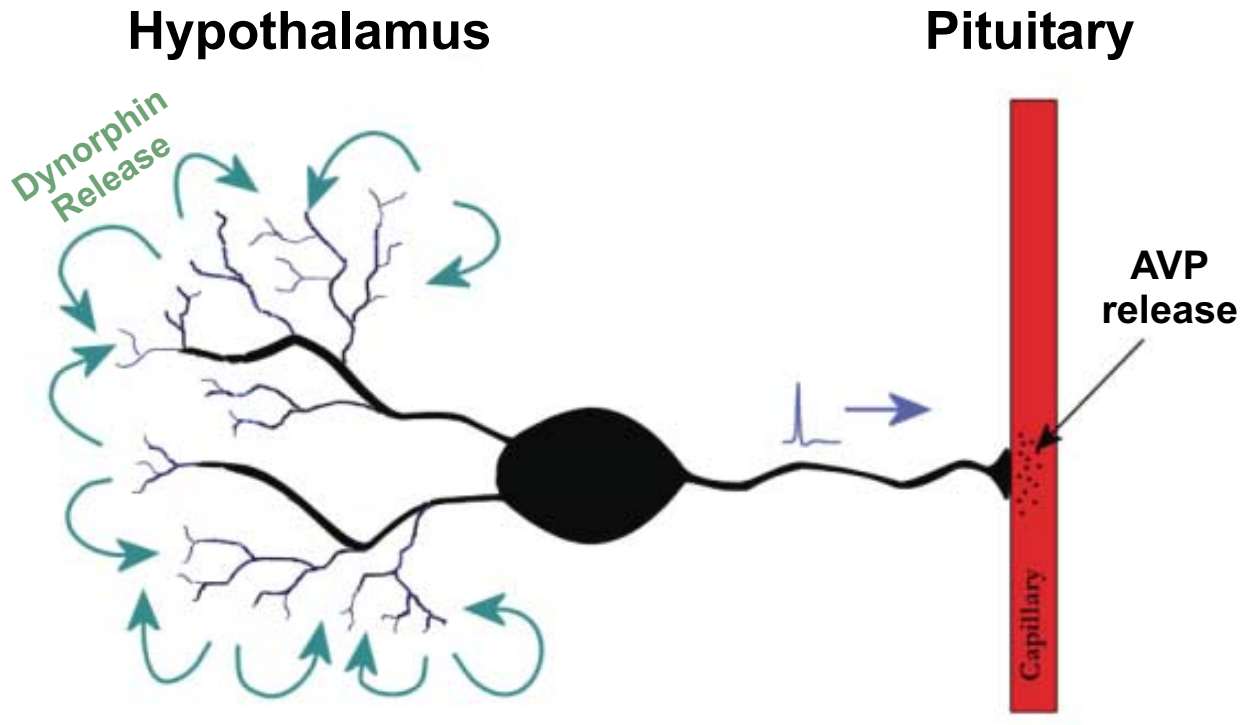
The plateau is sustained by the influx of calcium during each spike, which maintains the inhibition of $I_{K,leak}$.

Calcium remains elevated throughout the burst, and reaches a plateau within the first few seconds of activity. Burst termination is not associated with an increase in calcium, and hence is unlikely to be solely mediated by a calcium dependent K^+ AHP current.



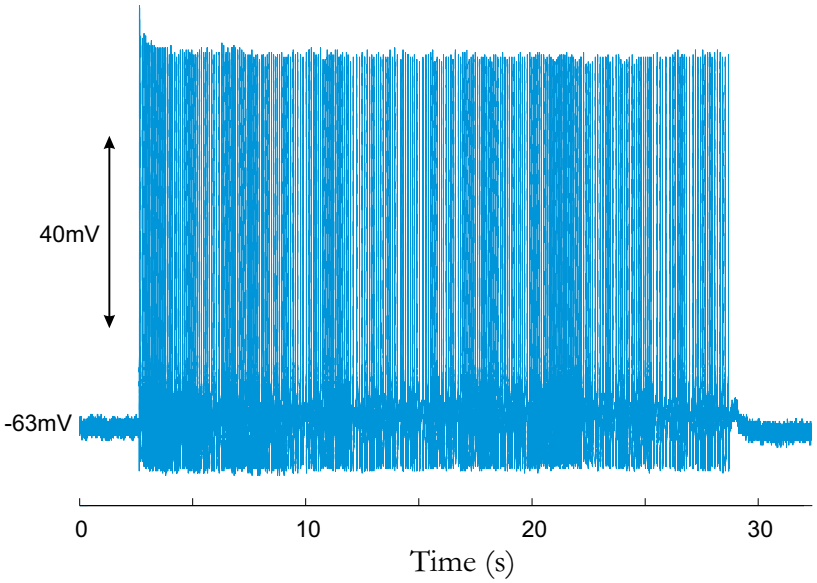
Top: **Electrical burst and profile of associated calcium concentration. Note that calcium remains elevated throughout the burst, but briefly decays when electrical activity is temporarily interrupted during the burst. Electrical activity: whole cell recording from SON hypothalamic explant; Calcium concentration imaged with FURA-2.** *Bottom*: **Model of burst and associated calcium concentration.**

The burst termination mechanism is unknown. However, AVP cells also co-secrete an inhibitory opioid, dynorphin, from their dendrites. Dynorphin acts as an autocrine inhibitor of the DAP [Brown *et al.*, 1998] and so can terminate phasic activity.

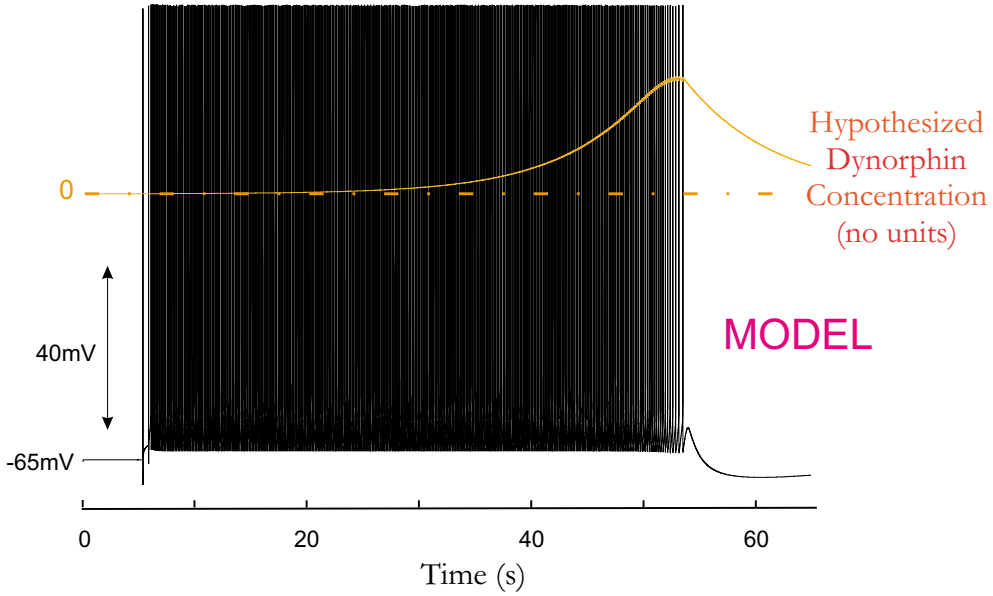


We propose that dynorphin accumulates extracellulary during the burst and provides a negative feedback that eventually terminates the burst.

Sharp electrode recording from hypothalamic explant. Courtesy of R. Teruyama, Memphis, TN.

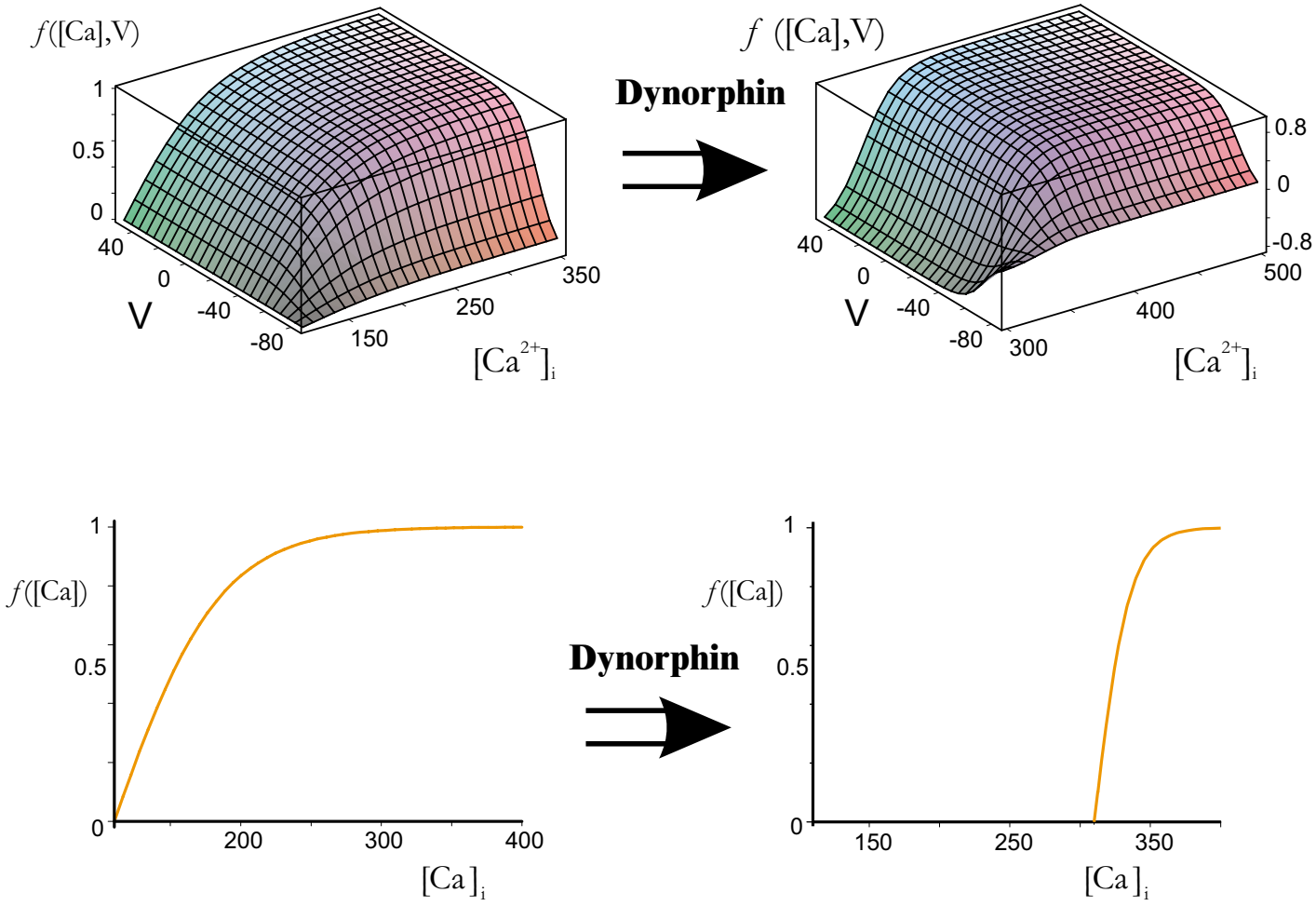


Modelled burst plotted against profile of extracellular dynorphin concentration.



To terminate the burst, and to prevent it from restarting, the DAP must be inhibited in such a way that it is brought (and kept) below its calcium threshold for activation.

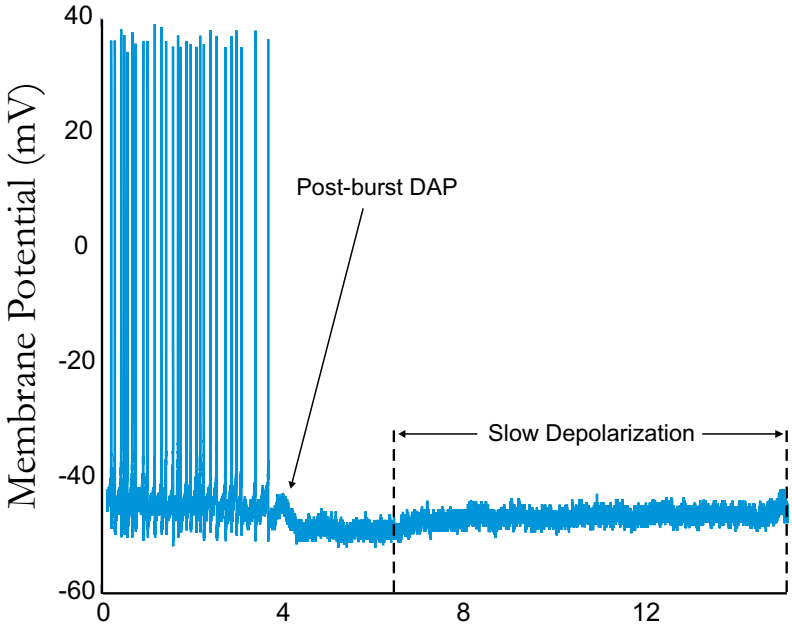
However, since Ca^{2+} remains at a constant elevated plateau throughout the burst, this can only happen if the function f is shifted by dynorphin to higher calcium concentrations.



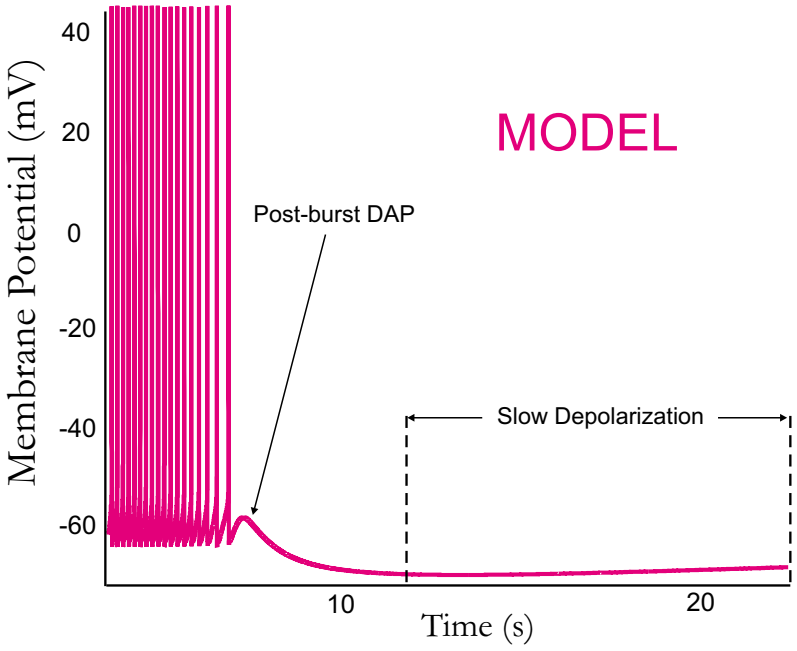
Top: **The proposed action of dynorphin on the DAP activation function, note that f now goes negative for certain elevated calcium concentrations, this is the mechanism underlying the slow depolarization.** *Bottom*: **Cross section of upper figure showing how dependency on calcium is altered by dynorphin.**

If the termination of a burst is caused by raising the threshold for DAP activation and the consequent support of a plateau, then a very brief DAP will occur at the end of the burst, and this is observed in real recordings.

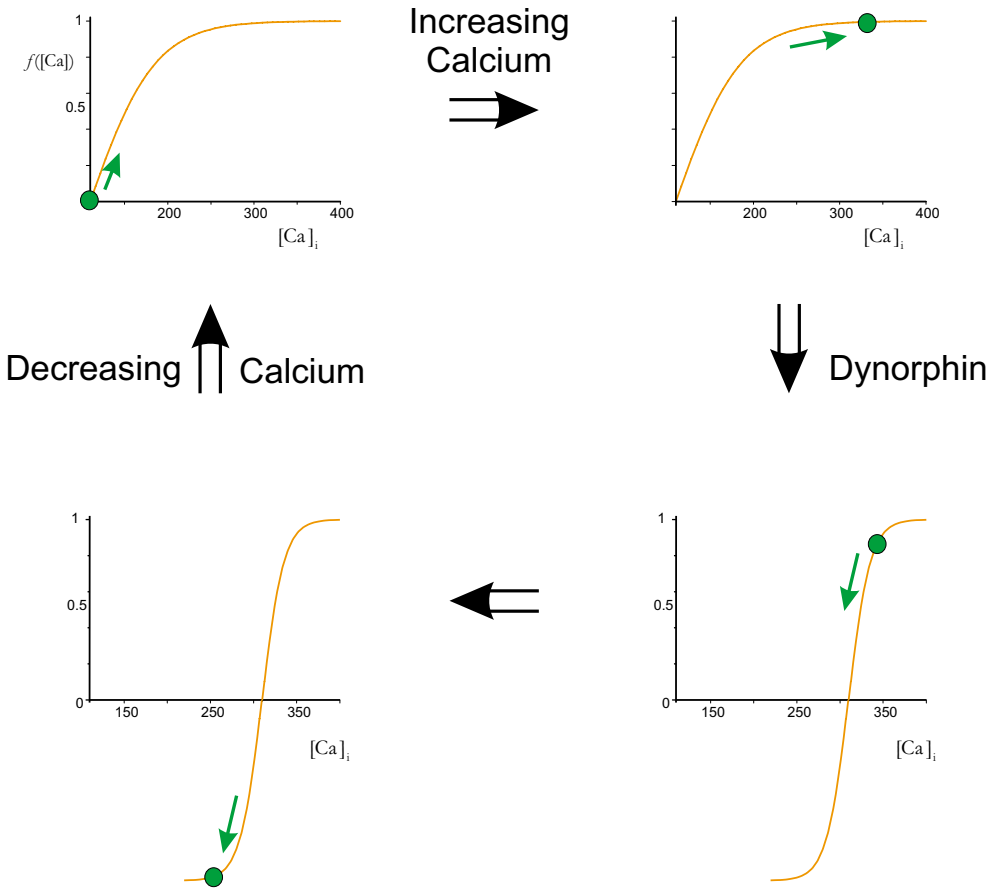
Recorded burst termination, note frequency adaptation during the final few spikes; the brief persistence and then rapid decay of the plateau as a post-burst DAP; and the slow depolarization following the burst. Sharp electrode recording from hypothalamic explant (enlargement of previous figure). Courtesy of R. Teruyama, Memphis, TN.



Model of burst termination, enlargement of burst shown previously.



The silent period between bursts derives from the unbinding and clearance of dynorphin. Silences are typically $\sim$ 20s which implies that the dynorphin time constant is much greater than that of $[Ca^{2+}]_i$ decay ($\sim$ 5s). Thus dynorphin decays more slowly than Ca^{2+} and so the DAP activation function, $f([Ca^{2+}]_i, V)$, remains shifted to the right while calcium decreases. There is then a short time when f is briefly negative and the resting K^+ current is more strongly activated than at its equilibrium state. Subsequently both Ca^{2+} and dynorphin return to their resting values, f returns to its baseline and $I_{K,leak}$ returns to its resting level. The recovery of $I_{K,leak}$ appears as a *slow depolarization* of the membrane potential, and is observed in recordings.



Schematic showing the evolution of the DAP activation function as the burst progresses and the mechanism underlying the post-burst DAP and slow depolarization. The calcium concentration at each stage is depicted by the filled green ball.

Conclusions

- Our AVP cell model incorporates the known neurophysiological properties of these cells and reproduces their firing activity.
- We have shown how phasic bursts can be initiated and supported by a calcium dependent reduction in a resting K^+ current.
- We propose that the autocrine messenger dynorphin plays a role in burst termination and we show how the interaction of dynorphin and the inhibition of a K^+ leak supports bursting.
- If dynorphin acts as an autocrine messenger for burst termination then we propose that it does so by shifting the DAP activation curve, $f(\ldots)$, to higher calcium concentrations.
- Our model further predicts the mechanism underlying the brief post-burst depolarized afterpotential (DAP) and the slow depolarization between bursts.

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References

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Li, Z.H., & Hatton, G.I. 1997. Reduced outward K^+ conductances generate depolarizing after-potentials in rat supraoptic nucleus neurones. *Journal of Physiology (London)*, **505**(1), 95–106.