Dendritic spike initiation in globus pallidus neurons results in an enhanced influence of excitatory inputs not predicted by somatic dynamic current clamping.

Jesse E Hanson and Dieter Jaeger[1]
Dept. of Biology, Emory University, Atlanta, Georgia 30322, USA
[1]Corresponding author, email address: djaeger@emory.edu

Oral presentation preferred.

Abstract

The presence of dendritic action potentials triggered by synaptic input in globus pallidus (GP) indicates a complex integration of synaptic inputs. To examine synaptic integration in GP neurons we created a realistic compartmental model. We found that the input-output function of the model neuron as determined by applying synaptic inputs to the soma, simulating a dynamic current clamp experiment, did not predict the input-output function observed with distributed dendritic synapses. This indicates that in neurons where action potentials can be initiated at multiple locations in the dendritic tree, dynamic current clamping may miss important coding features of the neuron under investigation.

Summary

Less than 10% of synapses onto Globus Pallidus (GP) neurons are excitatory, and many of these excitatory inputs contact distal dendritic sites [1]. The power of the excitatory inputs is potentially enhanced, however, by the presence of sodium channels throughout the dendrites of the neurons, which we found to be clustered at locations postsynaptic to excitatory synapses [2]. The excitability of GP dendrites allows distal synapses that might otherwise have no significant effect on the somatic membrane potential to trigger action potentials in the dendrites (Figure 1). To examine the effect of dendritic spike initiation on synaptic integration we constructed a multi-compartment model of a GP neuron based on a Neurolucida reconstruction using the GENESIS simulation software. Based on the GP neuron literature, the model neuron contained transient and persistent sodium channels, Kv2 and Kv3 family delayed rectifier and Kv3 and Kv4 family A-type potassium channels, high-voltage-activated calcium channels, BK, SK, and slow after-hyperpolarization calcium-dependent potassium channels and hyperpolarization-activated mixed cation channels [3-8]. The densities of each channel were chosen so that the model replicated

basic properties of experimental data (Fig. 2A). In keeping with the known anatomy, the voltage-gated channels were distributed in both the soma and dendrites and the sodium channels had elevated levels at sites of synaptic input. Also based on the anatomy, excitatory synapses were outnumbered by inhibition by 10:1, and both inputs were distributed across the dendritic tree. (Figure 2B) [1].

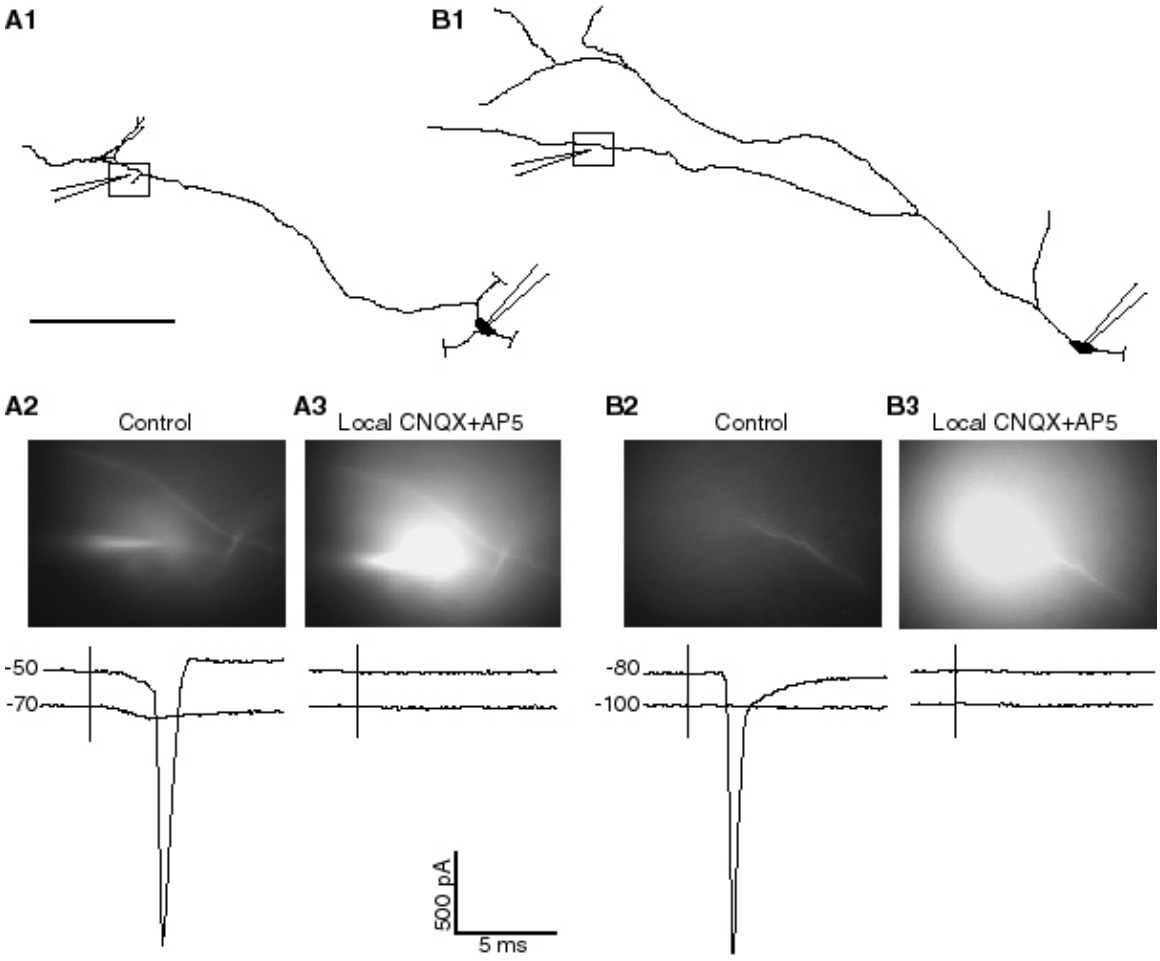

Figure 1. *Action potentials can be triggered in the dendrites of GP neurons. Stimulation electrodes were placed near dendrites that were visualized using an intracellular solution containing Alexa Fluor 568.* ***A1***, ***B1***. *The morphology of the recorded neurons. The placement of somatic recording electrodes and stimulation electrodes are illustrated. Main dendritic branches that were not stimulated are truncated.* ***A2***, ***B2***. *Images of stimulation electrodes and dendrites (boxed area from A1 and B1) obtained with fluorescent imaging during recording. Voltage clamp recordings of stimulation responses are shown. The stimulation site in A showed sub-threshold EPSCs at hyperpolarized potentials in the somatic voltage clamp, while the more distal stimulation site in B showed no sub-threshold EPSCs. In both cases, spike currents are observed*

even though the soma is clamped below spike threshold, indicating dendritic initiation. ***A3***, ***B3***. *Glutamate receptor antagonists CNQX and AP-5 were pressure applied from the stimulation electrode along with rhodamine to allow visualization. Antagonist application eliminated the stimulation responses completely indicating that action potentials were triggered by synaptic activation. (Figure adapted from [2])*

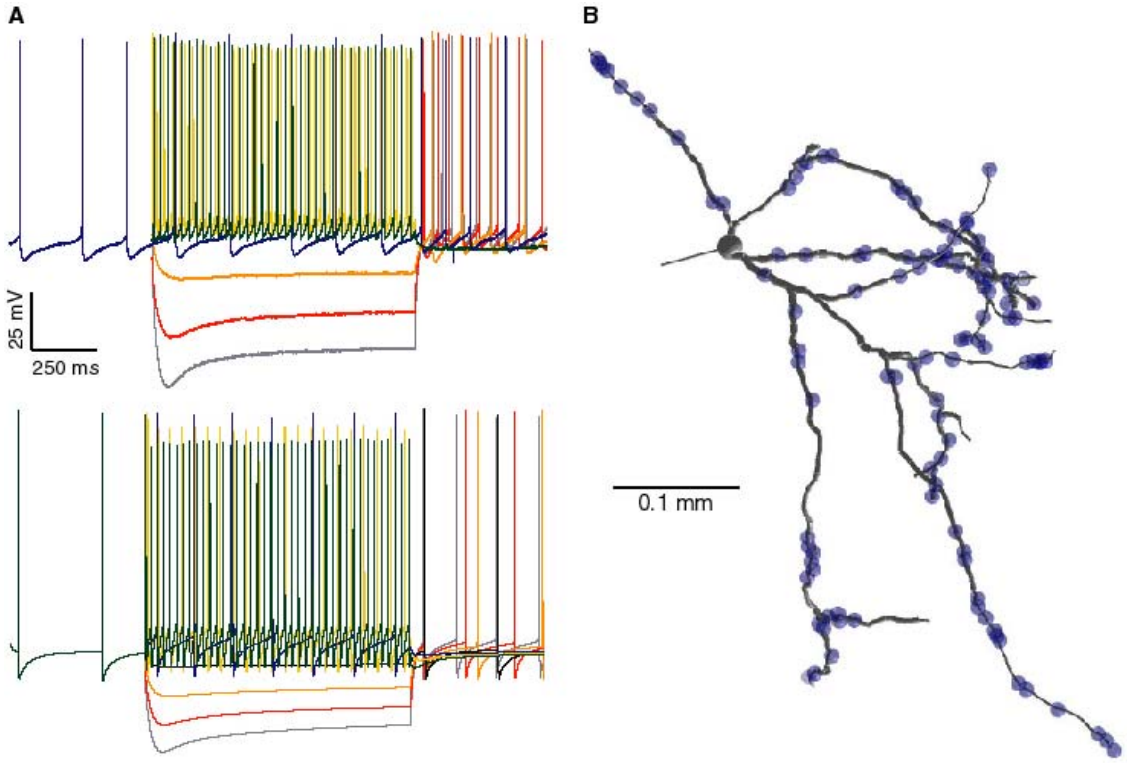

Figure 2. *The model GP neuron.* ***A****. Response of a real GP neuron (top) and the model GP neuron (bottom) to current injection steps of -.35 to +.25 nA. Features of GP membrane properties matched by the model included spontaneous pacemaker activity, the ability to sustain high frequency firing, a hyperpolarization 'sag', post-inhibitory rebound, and post-depolarizing burst cessation of spiking [7].* ***B****. The morphology of the model neuron. Sites of excitatory synaptic input are indicated with blue spheres. Sites of inhibitory input are not illustrated but outnumber the excitatory inputs by a factor of 10 and completely cover the neuron.*

To examine synaptic integration in the model, excitatory and inhibitory synaptic inputs were activated at rates that might be seen *in vivo* and the membrane potential was recorded simultaneously from every compartment in the model (Figure 3A). Based on these simulations, it is evident that spike initiation occurs at dendritic locations and that these spikes may or may not

propagate to the soma and cause output firing (Figure 3, inset). This indicates the importance of dendritic contributions to synaptic integration in the model neuron.

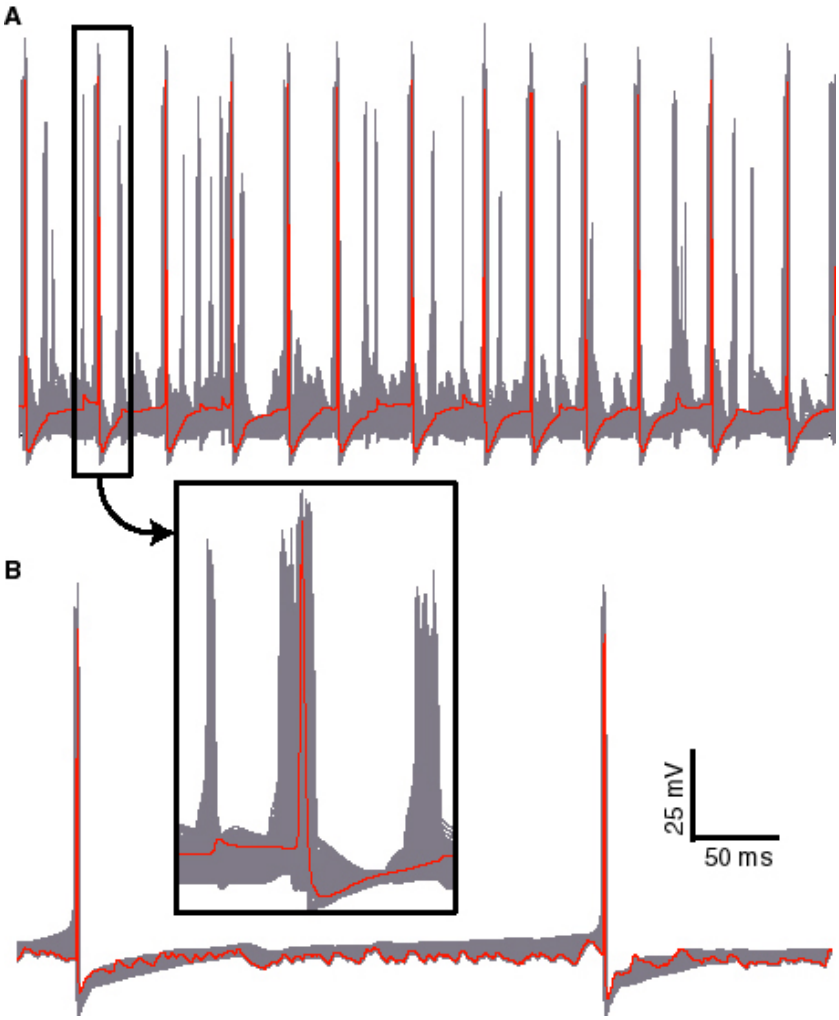

Figure 3. *Response of the model to 30 Hz excitation and 10 Hz inhibition. Membrane potentials from all compartments are shown (gray) with the somatic membrane potential in red.* ***A****. With the synapses distributed across the dendritic tree, action potentials are triggered at a high frequency. Action potentials triggered in the dendrites may either fail before they reach the soma or propagate to the soma and then back-propagate into the other dendrites (inset).* ***B****. Simulated dynamic current clamping experiment. In this simulation, the identical excitatory and inhibitory synaptic inputs were all applied to the soma of the model. Under these conditions the much more numerous inhibition results in a total synaptic drive that is much weaker than in the distributed case. A manuscript describing the basic model behavior is in preparation.*

A powerful technique used to examine synaptic integration *in vitro* is dynamic current clamping [9]. This technique can be used to apply simulated synaptic conductances from an entire presynaptic network onto a real GP neuron *in vitro* while the resulting output of the neuron is measured [10]. A limitation of the technique is that all of the synaptic inputs must be applied to the soma through the recording electrode. In light of the present findings of dendritic contributions to processing in GP neurons, this limitation may result in significant errors in determining the input-output function of these neurons. Therefore, we simulated a dynamic current clamp experiment in the model by applying all of the synaptic inputs directly to the soma of the model (Figure 3B). The simulated dynamic clamping experiment indicated that the same balance of inhibition and excitation that allowed rapid spiking when distributed throughout the dendrites resulted in a much lower spike rate when applied to the soma. To further examine this effect we examined distributed vs. somatic application of synaptic input at different rates of excitatory and inhibitory input (Figure 4). This analysis showed that especially in the presence of inhibition, that dynamic current clamping does not accurately reflect mean steady state input-output relations of GP neurons. Thus while dynamic clamping is a useful tool for studying synaptic integration in general, in situations with spike initiation occurring in the dendritic tree this technique has significant limitations.

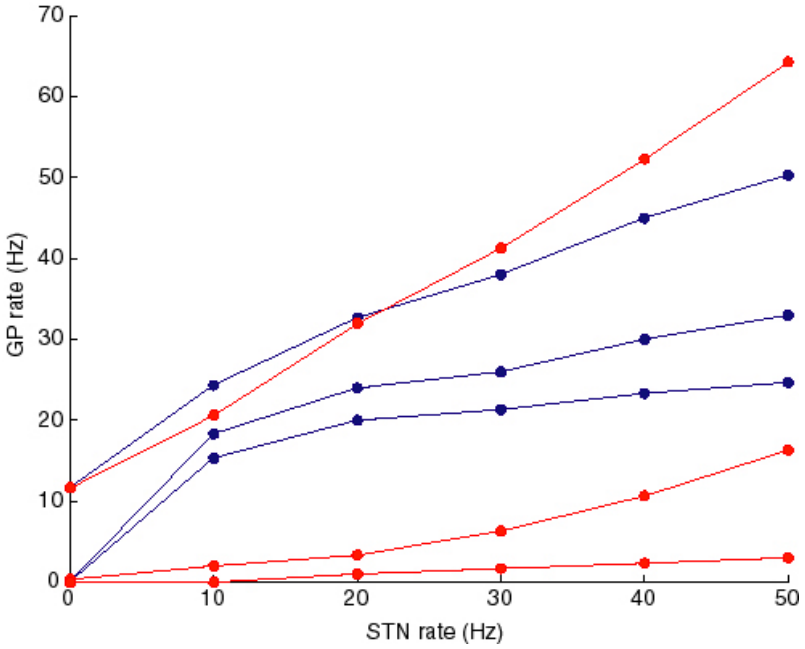

Figure 4. *Spike rate of the model GP neuron as a function of different excitatory input spike rates. Blue lines indicate distributed synapese during 0, 10 or 20 Hz inhibition (top to bottom). Red lines indicate the identicle excitatory and inhibitory inputs all applied to the soma.*

References

1. Shink, E. and Y. Smith, *Differential synaptic innervation of neurons in the internal and external segments of the globus pallidus by the GABA- and glutamate-containing terminals in the squirrel monkey.* Journal of Comparative Neurology, 1995. **358**(1): p. 119-141.
2. Hanson, J.E., Y. Smith, and D. Jaeger, *Sodium channels and dendritic spike initiation at excitatory synapses in globus pallidus neurons.* submitted, 2003.
3. Chan, C.S., K. Burnell, and D.J. Surmeier. *Persistent sodium current and hyperpolarization-activated cation current drive tonic firing in globus palliuds neurons*. in *Society for Neuroscience*. 2002. Washington, D.C.: #164.5 Online.
4. Tkatch, T., G. Baranauskas, and D.J. Surmeier, *Kv4.2 mRNA abundance and A-type K(+) current amplitude are linearly related in basal ganglia and basal forebrain neurons.* J Neurosci, 2000. **20**(2): p. 579-88.
5. Baranauskas, G., T. Tkatch, and D.J. Surmeier, *Delayed rectifier currents in rat globus pallidus neurons are attributable to Kv2.1 and Kv3.1/3.2 K(+) channels.* J Neurosci, 1999. **19**(15): p. 6394-404.
6. Hernandez-Pineda, R., et al., *Kv3.1-Kv3.2 channels underlie a high-voltage-activating component of the delayed rectifier K+ current in projecting neurons from the globus pallidus.* J Neurophysiol, 1999. **82**(3): p. 1512-28.
7. Cooper, A.J. and I.M. Stanford, *Electrophysiological and morphological characteristics of three subtypes of rat globus pallidus neurone in vitro.* J Physiol, 2000. **527 Pt 2**: p. 291-304.
8. Surmeier, D.J., N. Seno, and S.T. Kitai, *Acutely isolated neurons of the rat globus pallidus exhibit four types of high-voltage-activated Ca2+ current.* Journal of Neurophysiology, 1994. **71**(3): p. 1272-80.
9. Sharp, A.A., et al., *Dynamic clamp: computer-generated conductances in real neurons.* J Neurophysiol, 1993. **69**(3): p. 992-5.
10. Hanson, J.E. and D. Jaeger, *Short-term plasticity shapes the response to simulated normal and Parkinsonian input patterns in the globus pallidus.* J Neurosci, 2002. **22**(12): p. 5164-5172.