These studies show that dopamine neurons receive inputs containing glutamate, which evokes fast excitatory synaptic currents mediated by NMDA and non-NMDA receptors, as well as a slow-onset long-duration synaptic current mediated by metabotropic glutamate receptors. Moreover, inputs containing GABA produce a fast IPSP mediated by $GABA_A$ receptors and a slow IPSP mediated by $GABA_B$ receptors. The use of ligands to activate or block presynaptic receptors on afferents containing glutamate and GABA may be an effective strategy for influencing dopamine neuronal activity and may ultimately be useful in the treatment of a variety of pathological human conditions in which dopamine release plays a role.

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J. M. Tepper, C. A. Paladini, and P. Celada

Center for Molecular and Behavioral Neuroscience
Rutgers, The State University of New Jersey
Newark, New Jersey 07102

GABAergic Control of the Firing Pattern of Substantia Nigra Dopaminergic Neurons

In vivo, dopaminergic neurons fire spontaneously in three different patterns: a pacemaker-like regular firing pattern, a random mode, and a bursty mode. Whereas the random pattern is the most common firing pattern in anesthetized rats, dopaminergic neurons recorded *in vitro* fire spontaneously

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almost exclusively in the pacemaker-like mode. This last fact suggests that afferent inputs play an important role in the control of the firing pattern of the nigrostriatal dopaminergic neurons. The origins of the different firing patterns are important, because increases in bursting have been correlated with increases in dopamine release.

Excitatory amino acid afferents originating in the frontal cortex, subthalamic nucleus, and pedunculopontine nucleus have been shown to play an important role in the modulation of dopaminergic neuron firing pattern (1), although these inputs may not be the only ones important for the control of the firing pattern *in vivo*. The predominant inputs to dopamine neurons in the substantia nigra are GABAergic, the best characterized of which originate from the neostriatum and globus pallidus. Data have shown that substantia nigra dopaminergic neurons also receive a significant input from the axon collaterals of substantia nigra pars reticulata projection neurons (2). Dopaminergic neurons have been shown to exhibit both $GABA_A$ and $GABA_B$ responses *in vitro* (3).

In vivo extracellular recordings from dopaminergic neurons in urethane-anesthetized rats following direct stimulation of striatum, globus pallidus, or antidromic stimulation of substantia nigra pars reticulata projection neurons revealed that all three afferents produce inhibition of dopaminergic neurons. Data from experiments using minute local pressure application of $GABA_A$ and $GABA_B$ antagonists via a picospritzer from antidromically identified nigrostriatal neurons show that the inhibition arising from each of the principal GABAergic inputs is completely and reversibly blocked by the $GABA_A$ antagonist, bicuculline (250 μM, 1 nl). This is illustrated for inhibition elicited from globus pallidus for one representative dopaminergic neuron (Fig. 1A, 2). The $GABA_B$ antagonist, saclofen (400 μM, 1 nl), not only failed to block these evoked inhibitions, but often facilitated them (Fig. 1A, 3). Thus, it appears that the monosynaptic inhibition arising from striatum, globus pallidus, and the axon collaterals of the pars reticulata GABAergic projection neuron is mediated by a $GABA_A$ receptor. Furthermore, these data also suggest that all three of these GABAergic afferents possess presynaptic inhibitory $GABA_B$ autoreceptors on their nerve terminals.

Local application of $GABA_A$ and $GABA_B$ antagonists differentially affect the firing pattern of dopaminergic neurons. The pattern of activity of substantia nigra dopaminergic neurons was quantified by computing autocorrelograms, coefficients of variation, and the percentage of spikes fired in bursts (2). Local nigral application of the $GABA_A$ antagonist, bicuculline, through the recording pipette markedly increases the proportion of neurons that fire in the bursty mode, or shifts single dopaminergic neurons firing in the pacemaker or random modes to bursty firing when applied by local pressure injection. Conversely, local application of the $GABA_B$ antagonist, saclofen, through the recording pipette (or excitotoxic lesion of globus pallidus) decreases bursty firing and produces a modest increase in the proportion of neurons that fire in a pacemaker-like mode, or shifts single dopaminergic neurons to pacemaker-like firing when applied by local pressure injection through a second pipette. This latter effect is apparently due to increased GABA release acting on postsynaptic $GABA_A$ receptors as a result of the blockade of inhibitory presynaptic $GABA_B$ autoreceptors. These data suggest that bursty firing in substantia nigra dopamin-

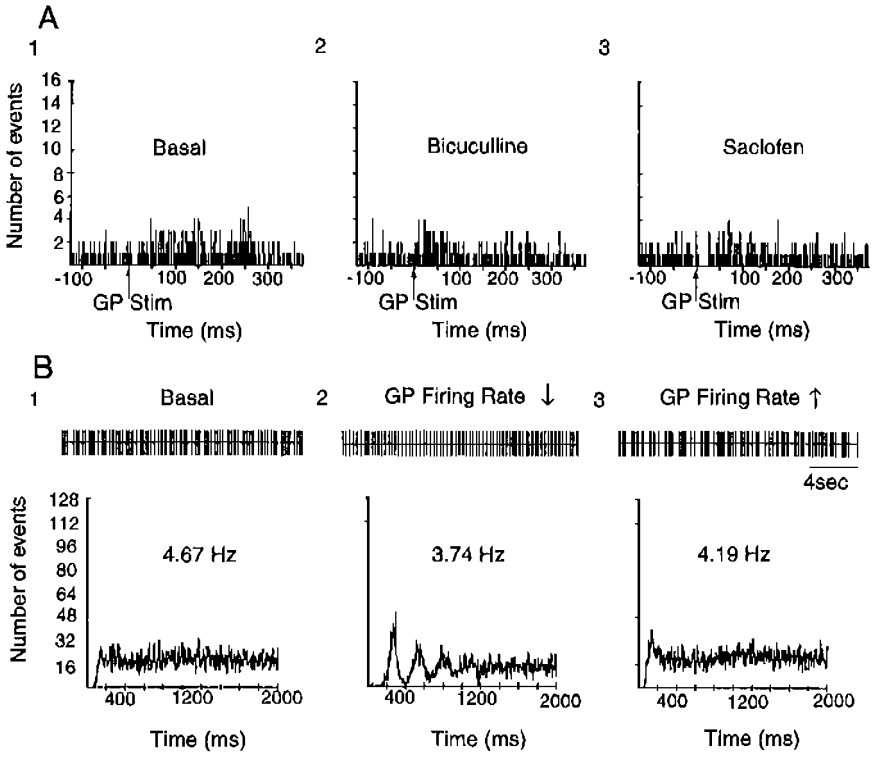

FIGURE 1 (*A*) Globus pallidus (GP) stimulation (1 mA, 0.5 ms, marked by arrow at time = 0) produces inhibition of an antidromically identified nigrostriatal dopaminergic neuron (*1*). Local application of bicuculline to the same neuron blocked the inhibition completely (*2*) while subsequent local application of saclofen (after recovery from bicuculline) increases the strength and duration of the inhibition (*3*). (*B*) Spike trains and autocorrelograms showing changes in the firing pattern of a single dopaminergic neuron as globus pallidus neurons are inhibited (*2*) and excited (*3*) by local infusion of muscimol and bicuculline, respectively, into globus pallidus. Inhibition of globus pallidus by muscimol leads to a decrease in spontaneous firing rate and a shift to pacemaker firing. Excitation of globus pallidus neurons by subsequent infusion of bicuculline leads to an increase in dopaminergic neuron firing rate and a shift to bursty firing.

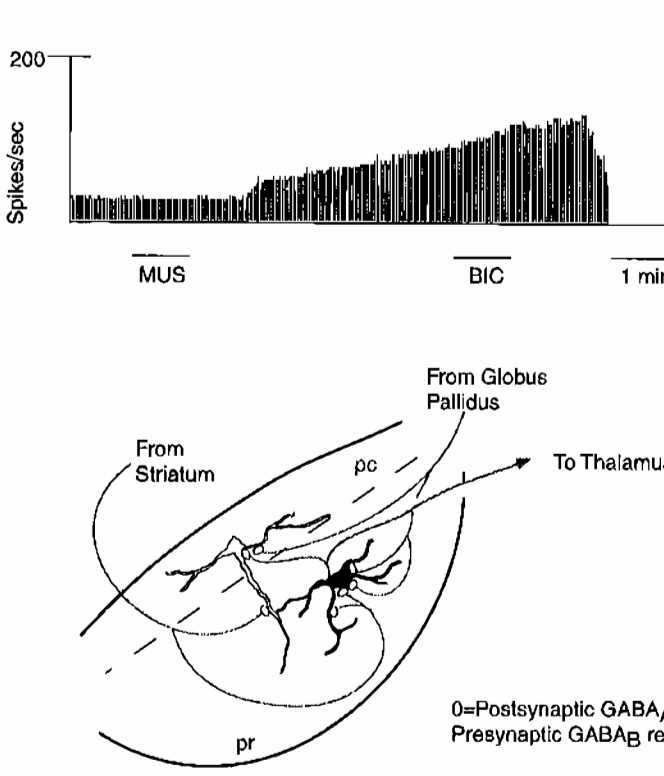

FIGURE 1 Continued. (*C*) Infusion of muscimol (MUS) into globus pallidus produces a dramatic increase in the spontaneous activity of an unidentified pars reticulata GABAergic neuron to over 100 spikes/sec. Subsequent infusion of bicuculline (BIC) reverses the excitation and produces complete cessation of firing. (*D*) A proposed model of interaction of the GABAergic inputs into the dopamine neurons in substantia nigra pars compacta.

ergic neurons is suppressed when $GABA_A$ inhibition is increased and is facilitated when $GABA_A$ input is decreased.

To determine the physiological relevance of these pharmacological phenomena with respect to the control of firing pattern of dopaminergic neurons *in vivo*, and to begin to identify the pathways involved, we examined the activity of substantia nigra dopaminergic neurons *in vivo* while manipulating globus pallidus neuron firing rates. Globus pallidus neuron firing rates were decreased by infusing muscimol (800 μM, 200 nl) or increased by infusing bicuculline (1000 μM 200 nl) into the globus pallidus through a 32-g cannula (4). Muscimol-induced inhibition of globus pallidus neurons, verified by extracellular recording, shifted dopaminergic neurons to the pacemaker firing pattern, accompanied by a slight but significant decrease in firing rate (86% of baseline), as shown for a typical dopaminergic neuron in Figure 1B, 2. Conversely, bicuculline-induced excitation of globus pallidus neurons shifted dopaminergic neurons to the bursty firing mode, with a concomitant increase in firing rate

(129% of baseline), as shown in Figure 1B, 3). Because the changes in firing rates of the dopaminergic neurons were opposite of what would be expected based on a monosynaptic GABAergic input from globus pallidus, the effects of manipulating the globus pallidus firing rate on the activity of nondopaminergic pars reticulata neurons were examined. These data revealed that nondopaminergic pars reticulata neurons were far more sensitive to the pharmacological manipulation of globus pallidus cells than were the dopaminergic neurons, and they responded in the opposite manner. That is, inhibition of globus pallidus activity produced a dramatic increase in the firing rate of the nondopaminergic pars reticulata neurons, while globus pallidus excitation decreased the nondopaminergic pars reticulata neuron firing rate (Fig. 1C). Thus, the changes in activity of substantia nigra dopaminergic neurons following manipulation of the globus pallidus neuron firing rate appear to be mediated, at least in part, indirectly through pars reticulata neurons. Although not conclusively identified, these nondopaminergic pars reticulata neurons exhibited spontaneous activity similar to that of identified GABAergic projection neurons (tonic firing at rates from 15 to 40/sec in a regular pattern), and some of these neurons were antidromically activated at short latency (0.8–3.0 ms) from ipsilateral thalamus.

These data suggest that GABAergic inputs can affect the activity of dopaminergic neurons *in vitro* in several ways. In addition to the well-known monosynaptic pathways originating in striatum and globus pallidus, there is a potent modulation of the firing pattern of dopaminergic neurons from globus pallidus that is mediated disynaptically through GABAergic neurons of the substantia nigra pars reticulata. The pars reticulata GABAergic projection neurons appear to exert a tonic inhibitory influence on burst firing. When they are inhibited by activation of globus pallidus neurons (or perhaps also is a result of increases in the activity of the direct striatonigral pathway), the dopaminergic neuron is released from tonic $GABA_A$ inhibition and burst firing ensues, perhaps mediated by a glutamatergic input from cortex, subthalamic nucleus, and/or pedunculopontine nucleus (1). Conversely, when the pars reticulata neurons are disinhibited, they act to depress bursty firing on dopaminergic neurons through activation $GABA_A$ receptors.

The physiological role of the postsynaptic $GABA_B$ receptor on dopaminergic neurons and the source of the $GABA_B$ input *in vivo* remain to be determined. These receptors do not appear to be stimulated *in vivo* by input from striatum, globus pallidus, or the axon collaterals of the pars reticulata projection neurons. However, these data do not rule out the possibility of an heretofore unidentified source of GABAergic input to dopaminergic neurons that may access the $GABA_B$ receptors. Spontaneous $GABA_B$-like synaptic responses have been observed in substantia nigra dopaminergic neurons *in vitro* (5), suggesting the possibility that there may be an intrinsic source of $GABA_B$ inputs to substantia nigra dopaminergic neurons.

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