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Cocaine Withdrawal and Neuro-Adaptations in Ion Channel Function

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Abstract

Chronic exposure to psychostimulants induces neuro-adaptations in ion channel function of dopamine (DA)-innervated cells localized within the medial prefrontal cortex (mPFC) and nucleus accumbens (NAc). Although neuroplasticity in ion channel function is initially found in drug-sensitized animals, it has recently been believed to underlie the withdrawal effects of cocaine, including craving that leads to relapse in human addicts. Recent studies have also revealed remarkable differences in altered ion channel activities between mPFC pyramidal neurons and medium spiny NAc neurons in cocaine-withdrawn animals. In response to psychostimulant or certain "excitatory" stimuli, increased intrinsic excitability is found in mPFC pyramidal neurons, whereas decreased excitability is observed in medium spiny NAc cells in drug-withdrawn animals compared to drug-free control animals. These changes in ion channel function are modulated by interrupted DA/Ca^{2+} signaling with decreased DA D2 receptor function but increased D1 receptor signaling. More importantly, they are correlated to behavioral changes in cocaine-withdrawn human addicts and sensitized animals. Based on growing evidence, researchers have proposed that cocaine-induced neuro-adaptations in ion channel activity and DA/Ca^{2+} signaling in mPFC pyramidal neurons and medium spiny NAc cells may be the fundamental cellular mechanism underlying the cocaine withdrawal effects observed in human addicts.

Index Entries: Cocaine; withdrawal; neuroplasticity; excitability; ion channel; electrophysiology; prefrontal cortex; nucleus accumbens.

Introduction

The prefrontal cortex (PFC) is a major association area connected to all areas of the neocortex and to various allocortical, limbic, and subcortical brain regions. It is also a terminal region of the mesocorticolimbic dopamine (DA) system that innervates the NAc and the

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ventral tegmental area (VTA). The NAc is a limbic structure that receives glutamatergic and dopaminergic inputs from the amygdala, hippocampus, medial dorsal thalamus, medial PFC (mPFC), and VTA. Functioning as an "interface" in the mesocorticolimbic DA system, the NAc participates in the regulation of motivation-driven behaviors. In humans, both the PFC and NAc are implicated in the control of cognitive tasks (e.g., attention, perception, thinking, learning, working memory, and executive functioning) and underlie several neurological disorders, including cocaine addiction *(1–4)*. In rodents, the PFC and NAc are necessary for the induction of behavioral sensitization (e.g., increased locomotion and stereotypy) and increased self-administration ("incubation" of cocaine craving), two established animal models for the study of drug addiction *(5–9)*. Lesions of the mPFC abolish psychostimulant-induced neuro-adaptations in the mesocorticolimbic DA system (also known as the reward pathway) and prevent cocaine sensitization *(6,10,11)*. Deficiency of the DA D1-class receptor in the NAc also results in the abolishment of cocaine-induced behavioral sensitization (e.g., increased locomotor activity and stereotypy; ref. *12*). Because behavioral sensitization and incubation of cocaine craving only occur in withdrawn animals in response to drug/cue-associated stimuli, these findings indicate that the glutamatergic output originating from the mPFC to the NAc plays a critical role in the development of cocaine withdrawal symptoms, including craving and relapse.

Chronic exposure to cocaine induces numerous changes in the activity of the mesocorticolimbic DA system. Previous clinical studies have indicated that the basal neuronal activity (reflected by glucose utilization) in the orbitofrontal cortex (OFC; including the mPFC) of cocaine-withdrawn human addicts is markedly decreased *(13,14)*. However, this reduction in functional energy metabolism is rapidly reversed and remarkably increased in the OFC of these addicts in response to drug reinstatement (an additional challenge dose of cocaine-like drug). A similar increase in functional metabolism to drug reinforcement is found in the mPFC of cocaine-withdrawn nonhuman primates after chronic self-administration of the drug *(15,16)*. These findings reveal that the neuronal activity is reduced in the mPFC during drug withdrawal but is increased with additional drug-associated stimuli in cocaine-withdrawn subjects.

Cocaine-induced changes in the PFC neuronal activity are associated with apparent neuro-adaptive alterations in the intrinsic excitability of mPFC pyramidal neurons and medium spiny NAc neurons. Recent investigations have demonstrated that noncontingent repetitive cocaine administration increases evoked Na^+ and Ca^{2+} action potentials in mPFC pyramidal neurons in drug-withdrawn animals in response to excitatory stimuli *(17–19)*. Importantly, these changes persist for at least 3 wk, revealing a neuroplasticity in ion channel activity. Conversely, an enduring decrease in the frequency of evoked sodium spikes is found in medium spiny NAc neurons of cocaine-withdrawn rats compared to drug-naive control animals *(20–23)*. Although the decreased firing is recorded in vitro with brain slice preparations, this remarkable difference in the intrinsic excitability between NAc spiny cells and mPFC pyramidal neurons indicates that the cellular mechanisms underlying psychostimulant-induced modulation of ion channel activity are distinct in the two cell populations.

Although the exact mechanisms underlying cocaine-induced neuro-adaptations in the mPFC and NAc of drug-withdrawn animals remain unknown, recent studies have revealed that psychostimulant-induced neuroplasticity in ion channel activity is modulated by altered DA/Ca^{2+} signaling. Here, our work provides a perspective on the literature that integrates the findings with respect to functional and conformational changes in membrane ion channels and DA/Ca^{2+} signaling into a hypothesized working model (Fig. 1). In this model, decreased basal neuronal activity in the reward pathway during cocaine withdrawal and increased mPFC

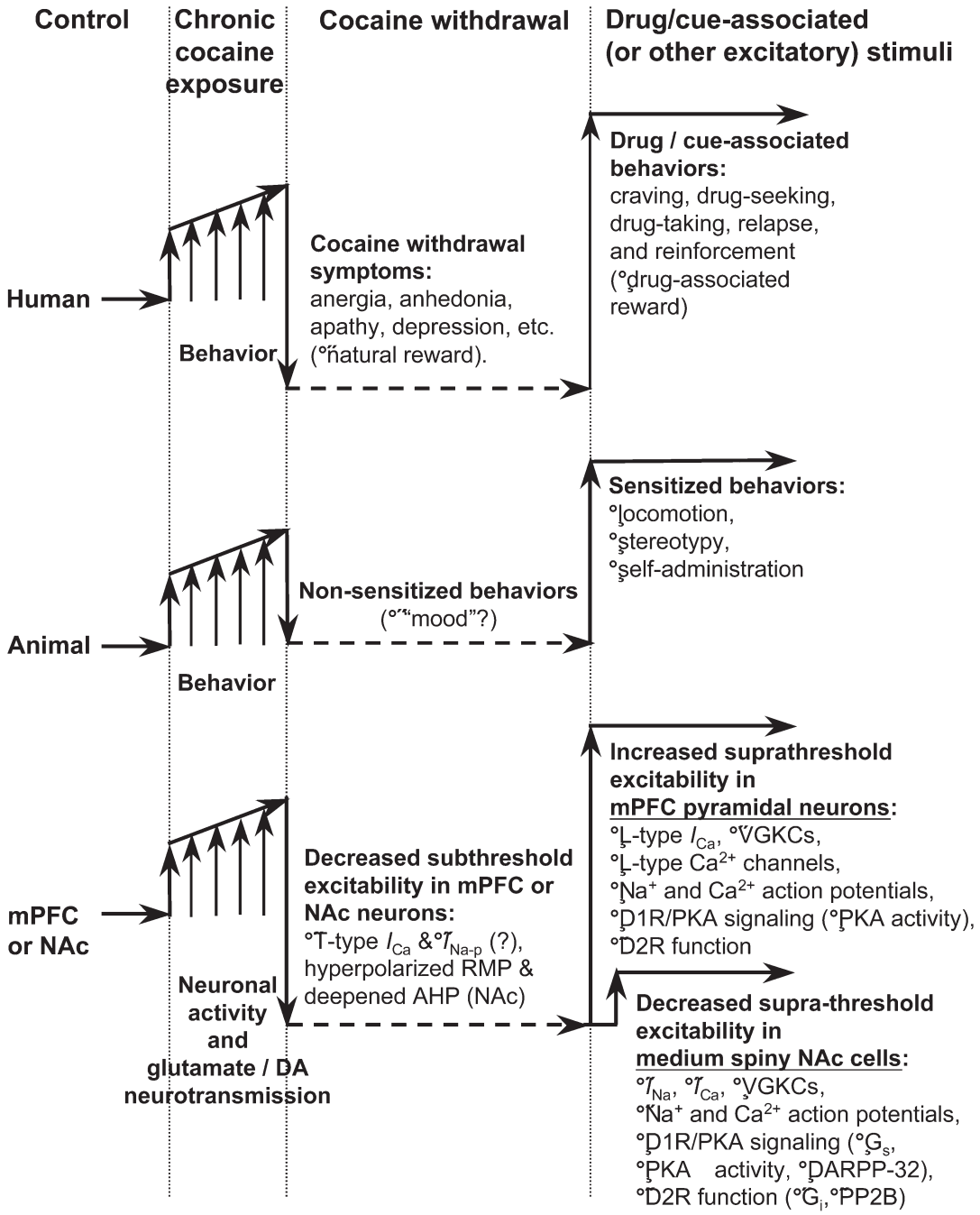


Fig. 1. Chronic cocaine-induced neuroadaptations in behaviors, neuronal activity and ion channel function of mPFC pyramidal neurons and medium spiny NAc cells either with or without drug-associated or other excitatory stimuli. Cocaine-induced neuroplasticity is found in subcellular, neuronal, system, and behavioral levels in cocaine-withdrawn animals. The maladaptations in ion channel function and DA/Ca^{2+} signaling could cause significant changes in the intrinsic excitability, basal and evoked neuronal activity, and neurotransmission output from the mPFC and NAc. Eventually they would lead to alterations in motivated behaviors, including drug craving and relapse. Based on these findings, it is proposed that neuro-adaptations in mPFC pyramidal neurons and medium spiny NAc cells are fundamental to, and critical for, the mechanisms underlying the withdrawal effects of cocaine in human addicts.

excitatory drive to the NAc with drug-associated stimuli are considered to underlie various withdrawal effects of cocaine.

DA Modulation of Ion Channel Activity in the Reward Pathway

The DA innervation of the mPFC and NAc modulates various higher order behavioral and cognitive processes *(24,25)*. DA modulation is executed in part via a regulation of the activity of the glutamatergic pyramidal neurons localized in the mPFC and of the GABAergic medium spiny neurons in the NAc *(26,27)*. Activity of these cells depends on their intrinsic excitability, which is primarily controlled and dynamically regulated by various voltage-gated Na^+, Ca^{2+}, and K^+ channels (a class of transmembrane ion channels activated by membrane depolarization near the channel). However, although membrane depolarization is essential and required for the activation of these ion channels, channel function in cortical and striatal neurons is also modulated by DA *(28–34)*, Ca^{2+}, and glutamate signaling, as well as by other intracellular pathways *(27,35,36)*.

D1R Modulation of Ion Channel Activity

Interacting with different signaling pathways, DA modulates the activity of voltage-gated ion channels by stimulating the DA D1-like receptor (D1R, including $D_{1,5}$ subtypes) and D2-like receptor (D2R, including $D_{2,3,4}$ subtypes) in the mesocorticolimbic DA system *(26,27)*. Previous findings have indicated that stimulation of D1Rs usually facilitates the intrinsic excitability of PFC pyramidal neurons, resulting in an increase in evoked firing *(28,35–38)*. This excitatory effect of D1R stimulation on evoked action potentials is apparently associated with increased persistent Na^+ currents ($I_{Na\text{-}per}$; refs. *28* and *30*, but *see also ref. 39*), enhanced high-voltage-activated (HVA-) L-type Ca^{2+} currents (I_{Ca}; refs *40* and *41*), and decreased voltage-gated K^+ currents (VGKCs or I_K; refs. *31,42,* and *43*). Moreover, it is also associated with an increased surface expression of α-mino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA) receptors *(44)* and excitatory postsynaptic *N*-methyl-D-aspartic acid (NMDA) currents *(45–47)*, along with the generation of "upstate"-like membrane potentials *(48,49)*. Conversely to voltage-gated ion channels, AMPA and NMDA receptors are ligand-gated transmembrane ion channels (also referred to as ionotropic receptors), which are opened in response to binding of glutamate (or AMPA and NMDA receptor agonists, respectively).

D1R stimulation also enhances the function of L-type Ca^{2+} channels of the medium spiny striatal neurons by increasing the activity of the cyclic adenosine monophosphate (cAMP)/protein kinase A (PKA) cascade *(50,51)*, which facilitates phosphorylation of the L-channel, thereby increasing Ca^{2+} influx and the excitability of these cells in response to membrane depolarization. Most of these ion channel activities modulated by D1Rs are implicated in an increased suprathreshold excitability, which would facilitate neuronal responsiveness to various excitatory stimuli and, therefore, increase either spontaneous or evoked firing.

D1R stimulation may also decrease the PFC and NAc excitability by suppressing voltage-sensitive I_{Na} *(20,39)*, evoking Ca^{2+}-activated K^+ currents with increased Ca^{2+} influx from L-type Ca^{2+} channels *(52)*, diminishing non-L-type HVA-Ca^{2+} potentials and currents *(21,28)*, increasing activity of GABAergic interneurons *(42)*, and enhancing $GABA_A$ receptor-mediated inhibitory postsynaptic currents (IPSCs) *(53)*. Additionally, activation of D1Rs can also increase or decrease the inwardly rectifying K^+ currents via different signaling pathways (*31,32,54*). Although these findings regarding D1R regulation of excitability appear controversial, they actually unmask the existence of a dynamic D1R modulation of ion channel activity through multiple signaling pathways. Therefore, the final effect of D1R modulation on the excitability and activity of a neuron depends on the integrated balance of the func-

tion of all membrane ion channels mediated by D1Rs.

D2R Modulation of Ion Channel Activity

Conversely to the effects of D1R stimulation on neuronal discharge (firing), activation of the D2R usually suppresses evoked action potentials in PFC pyramidal neurons as well as in medium spiny NAc cells of rats *(35,55–58)*. This inhibitory effect of D2R regulation on Na^+ spike firing most likely is mediated partly by the activation of A-type K^+ channels through a signaling pathway in which D2R-coupled neuronal Ca^{2+}-sensor proteins are functionally and conformationally involved (refs. *59–64*; Fig. 2A). D2R stimulation also decreases NMDA currents and attenuates the facilitating effects of NMDA/AMPA on evoked Na^+ spikes *(35,65)*, thereby diminishing the excitatory mPFC output to the NAc *(33)*. These inhibitory effects of D2Rs on the regulation of ion channel activity and evoked firing should be mainly attributed not only to its coupling with neuronal Ca^{2+}-sensor proteins and A-type K^+ channels that increases A-type K^+ outflow (Fig. 2A) but also to D2R-facilitated dephosphorylation of Ca^{2+} channels by calcineurin (CaN; protein phosphatase 2B or PP-2B) that decreases Ca^{2+} influx (our unpublished observation). Together, these findings show that D2R modulation could decrease the intrinsic excitability in DA-targeted neurons with interaction of DA, glutamate, and Ca^{2+} signaling.

Nevertheless, the D2R also regulates the excitability of mPFC pyramidal neurons and NAc spiny cells in certain positive ways. For example, we have determined that D2R stimulation induces a significant increase in whole-cell voltage-sensitive Na^+ currents (VSSCs or I_{Na}) in the majority of freshly dissociated medium spiny NAc neurons *(66)*. This D2R action has been attributed to decreased PKA activity and enhanced dephosphorylation of the Na^+ channel by activation of Ca^{2+}/CaM-dependent calcineurin following increased cytosolic-free Ca^{2+} levels (Fig. 3A). Additionally, activation of D_4Rs also diminishes IPSCs by facilitating dephosphorylation of $GABA_A$ receptors *(53,54,67)*. Obviously, these discrepancies observed in D2R-regulated responses complicate the interpretation of D2R modulation of neuronal excitability and have confused our understanding of the modulatory effects of D2R (and DA) on the activity of mPFC and NAc neurons.

Similarly to D1R modulation of intrinsic excitability and neuronal activity, these "controversial" findings in D2R modulation should not simply be concluded as "opposite" results. Because the D2R regulates the activity of various types of ion channels in a dynamic manner, the overall excitatory or inhibitory response of DA-innervated neurons to D2R stimulation depends on the integration of the responses of all types of membrane ion channel at any time-point, which may differ from each other in different types of neurons or the same type of cells but different individual. Furthermore, many D2R-modulated neuronal responses depend on cytosolic-free Ca^{2+} levels and on various DA/Ca^{2+} signaling-related proteins. It is well-established that stimulation of D2Rs leads to a drastic increase in intracellular Ca^{2+} release in the medium spiny neurons of the ventral and dorsal striatum (NAc and caudate-putamen CPu, respectively) *(68)*. This increase in Ca^{2+} mobilization parallels the activation of protein phospholipase C *(50,51)* and the inhibition of cAMP/PKA activity (refs. *23* and *66*; Fig. 2A). These changes in DA/Ca^{2+} signaling also functionally modulate ion channel activity in medium spiny neurons, thereby altering the intrinsic excitability and tonic activity of these cells. Although a thorough understanding of the exact mechanism underlying D1R and D2R modulation of voltage-sensitive sodium (VSSCs or I_{Na}), calcium (VSCCs or I_{Ca}), and potassium currents (VGKCs or I_K) requires further investigation, previous findings have demonstrated that a dynamic and integrated interaction among DA, glutamate, and Ca^{2+} signaling is essential in regulating the intrinsic excitability, basal neuronal activity, and information output from the mesocorticolimbic DA system.

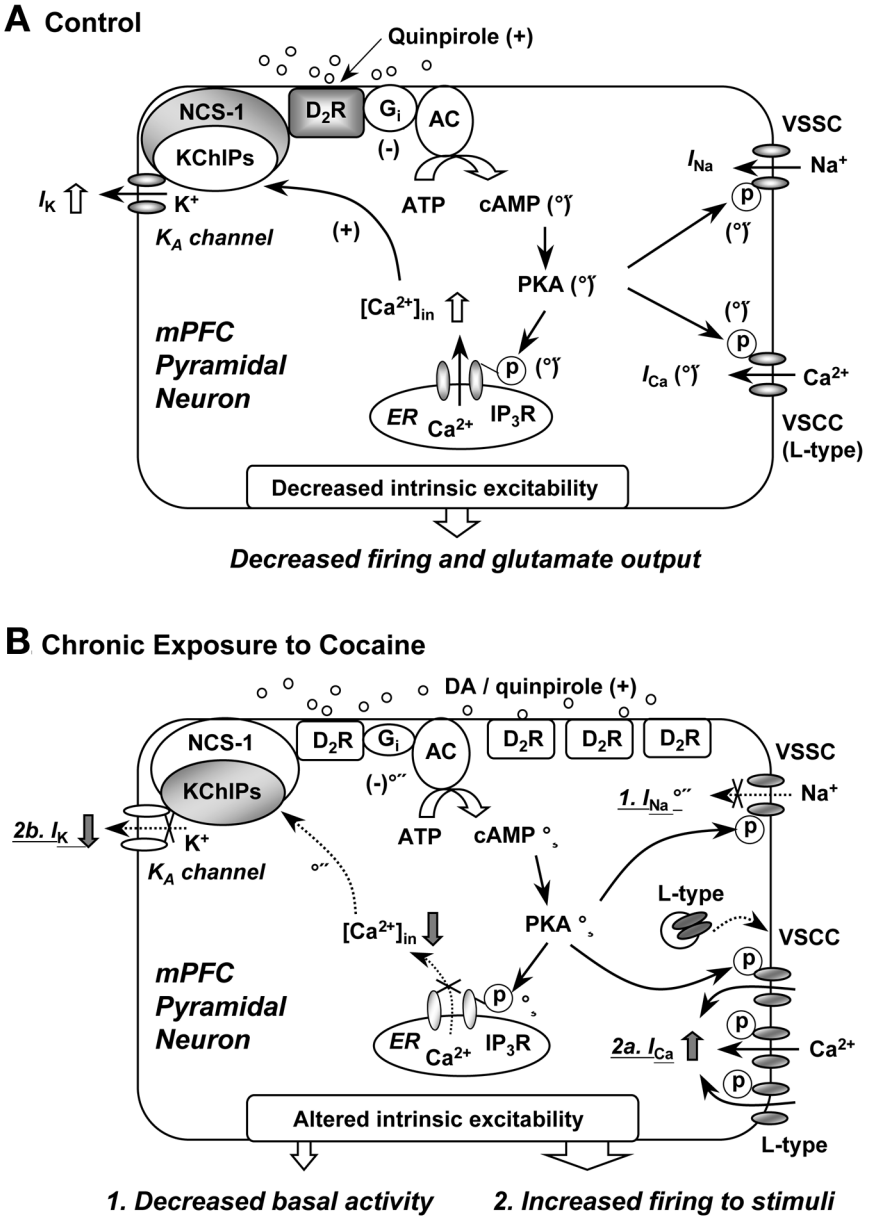


Fig. 2. Chronic exposure to cocaine alters D2R-modulation of membrane ion channel activity by interrupting the D2R/Ca^{2+}/NCS1-KChIPs/I_A pathway in mPFC pyramidal cells. **(A),** Stimulation of the D2R may activate A-type K^+ channels and increase I_K, thereby suppressing firing of mPFC pyramidal cells in drug naïve rats. This D2R modulation is regulated by inhibiting PKA, disinhibiting IP_3Rs, increasing Ca^{2+} release, and activating the neuronal Ca^{2+} sensor (NCS) proteins. D2R-mediated inhibition of PKA activity also results in an increase in Na^+ but a decrease in Ca^{2+} influx with reduced phosphorylation. The integrated changes in ion channel activity lead to a decrease in the intrinsic excitability. Arrows in brackets indicate the changes in activity. (Abbreviations: AC, adenylyl cyclase; PKA, c-AMP-dependent protein kinase A; VSSC, voltage-sensitive Na^+ current; VSCC, voltage-sensitive Ca^{2+} current; IP_3R, IP_3 receptor; ER, endoplasmic reticulum; NCS-1, NCS protein 1; KChIPs, K^+ channel interacting protein (another type of NCS protein); circled p, phosphorylation.) **(B),** Repeated cocaine administration increases D1R signaling but decreases D2R function. **1.** Cocaine withdrawal without drug or certain excitatory stimuli: mPFC pyramidal cells show a decreased basal activity with decreased I_{Na} (X) and consequently reduced neuronal activity. Reduced D2R function also results in a decreased intracellular Ca^{2+} release (X) and a compensatory D2R proliferation, which is associated with increased protein levels of L-channels.

2. Cocaine withdrawal with drug or certain excitatory stimuli: increased Ca^{2+} influx via the L-channels and decreased VGKC through the A-channels occur in pyramidal cells, leading to an increase in firing. Whether the surface expression of the L-channels and A-channels is increased and decreased, respectively, requires further investigation. Filled arrows indicate cocaine-induced alterations in conductances.

Chronic Cocaine Exposure Decreases D2R Signaling but Increases D1R/PKA Signaling

Attenuated D2R Function in Medium Spiny NAc Cells and mPFC Pyramidal Neurons

Chronic cocaine alters motivated behaviors in drug-withdrawn human addicts, whereas drug abstinence causes the withdrawal symptoms, including but not limited to anergia, anhedonia, apathy, depression, and craving, which leads to relapse. Associated with these cocaine withdrawal effects, there is a marked decrease in the basal neuronal activity in the OFC of human addicts compared to drug-free control subjects. Meanwhile, D_2R availability is also remarkably reduced in the striatum of drug-withdrawn human addicts *(69–71)*, suggesting a decreased D2R function in this brain region. Under these conditions, a psychostimulant-induced "high"—a surge of euphoric pleasure that rapidly follows administration of the drug—is experienced by human addicts, but only in those who have decreased D_2R availability in the dorsal and ventral striatum *(70,72)*. Moreover, the decreased D2R availability is associated with a reduced $D_2R/G_{i/o}$ protein coupling and an increased gene expression of cAMP response element binding proteins in the NAc of cocaine-pretreated animals. Nestler and colleagues *(73,74)* have demonstrated that the activity and protein levels of D2R-coupled $G_{i/o}$ α-subunits are significantly reduced in medium spiny NAc neurons following repeated cocaine treatment. Additionally, D2R-mediated inhibition in evoked Na^+ spike firing is diminished in mPFC pyramidal neurons in cocaine-withdrawn rats *(75)*. These findings indicate that D2R function is reduced in the DA-innervated mPFC pyramidal neurons and medium spiny NAc cells of cocaine-withdrawn human addicts as well as in animal models.

However, unlike the decreased D2R availability in the NAc, an increase in the D2R protein levels is found in the mPFC of cocaine-withdrawn rats *(75)*. This unique change in the D2R levels lasts for at least 3 wk with drug abstinence, indicating a neuroadaptive proliferation of the D2R as a compensatory responsiveness to decreased D2R function in the mPFC (Fig. 2B). Interestingly, previous studies have shown that selectively increasing D2R activation in the mPFC blocks the initiation and attenuates the expression of cocaine-induced behavioral as well as neurochemical sensitization in cocaine-withdrawn rats *(76,77)*. Based on these findings, researchers have proposed that D2R regulation of neuronal activity is decreased in the drug-withdrawn mPFC and NAc following chronic exposure to cocaine. It is currently unknown why D2R proliferation occurs in the mPFC, whereas reduced D2R availability takes place in the striatum of cocaine-withdrawn subjects. However, decreased D2R function in the mPFC—especially a reduction in D2R-mediated dephosphorylation of L-type Ca^{2+} channels, which would lead to an increased Ca^{2+} influx in mPRFC pyramidal neurons—should play an important role in the increased glutamate output seen in response to the psychostimulant-reinstated increase in neuronal activity. The increased PFC neuronal activity and excitatory glutamate output could trigger drug-awarded behavioral activity, particularly craving and relapse.

Enhanced D1R Signaling in Medium Spiny NAc Neurons and mPFC Pyramidal Neurons

Earlier electrophysiological studies on medium spiny NAc neurons have shown that psychostimulant-induced behavioral sensitization is associated with a persistent attenuation of excitatory responsiveness to glutamate but enhanced inhibitory responses to D1R regulation *(78–81)*. Recent investigations have further revealed that in vitro evoked Na^+ spikes and HVA-Ca^{2+} plateau potentials are suppressed in NAc spiny cells of cocaine-withdrawn rats *(20,22)*. These findings indicate a decreased intrinsic excitability of these neurons, even in response to excitatory stimuli

(e.g., in anesthetized rats with iontophoretically applied glutamate or in brain slice preparations with membrane depolarization).

This decrease in NAc excitability during drug withdrawal results primarily from cocaine-induced alterations in the activity of voltage-gated ion channels in NAc spiny cells. We have determined that, along with suppressed Na^+ and Ca^{2+} spikes, whole-cell Na^+ and Ca^{2+} conductances are decreased in medium spiny NAc neurons of cocaine-withdrawn rats *(20–23)*. Meanwhile, the outward rectification during membrane depolarization, which reflects an increased outflow of I_K, is also enhanced in these cells. These findings suggest that NAc spiny cells become much less excitable in vitro following repeated noncontingent cocaine treatment and drug withdrawal, a result that is supported by investigations focusing on excitatory synaptic transmission. Evidence from an AMPA-receptor- to NMDA-receptor-mediated excitatory postsynaptic current ratio study showed that the magnitude of long-term depression was decreased at synapses made by PFC afferents onto these cells because the excitability of NAc spiny cells had been suppressed in cocaine-withdrawn animals *(82)*. Accordingly, another study indicated that enhanced long-term potentiation (LTP) in field potential recordings of excitatory postsynaptic potentials could be induced in psychostimulant-withdrawn NAc spiny cells *(83)*, apparently from reduced basal activity levels.

Previous findings have also uncovered an enhanced D1R signaling in the NAc of cocaine-withdrawn rats, which is generally regulated by increased PKA activity beyond the D1R. For example, increases in the activities of adenylate cyclase (AC), G_s proteins, cAMP, and PKA are found in NAc spiny neurons after repeated cocaine administration *(73,74)*. Increased PKA activity downstream from D1Rs could affect ion channel function in different ways in sensitized animals. First, it decreases I_{Na} by enhancing phosphorylation of the Na^+ channel *(20,23)*. Second, even with facilitated L-type Ca^{2+} channel activity, it actually decreases whole-cell I_{Ca} and suppresses HVA-Ca^{2+} plateau potentials by reducing N- and R-type I_{Ca} *(21,22)*. This reduction in Ca^{2+} influx is most likely regulated by PKA-activated protein phosphatase 1 (PP1) through dephosphorylation of non-L-type Ca^{2+} channels *(21,84)*. Finally, it may also increase activity of certain subtypes of Ca^{2+}-activated and voltage-gated K^+ channels by phosphorylation *(22,85,86)*. Notably, however, under these circumstances, direct stimulation of D1Rs no longer induces further decreases either in Na^+ or Ca^{2+} currents *(20,21)* or in LTP *(82)* in NAc spiny cells in cocaine-withdrawn rats. These findings suggest that a "ceiling effect" regarding adapted D1R modulation might have occurred, through which the increased PKA activity achieved its maximal effects on regulating ion channel activity in these cocaine-withdrawn neurons. Based on these findings, researchers have proposed that cocaine-induced neuroplasticity in the activity of Na^+, Ca^{2+}, and K^+ channels results partly from the increased D1R/PKA and decreased D2R signaling with reduced CaN activity, leading to a decreased intrinsic excitability in NAc spiny cells (Fig. 3B).

Conversely to that observed in medium spiny NAc cells, the intrinsic excitability of mPFC pyramidal neurons in cocaine-withdrawn rats is increased concurrently with increased whole-cell I_{Ca} and decreased I_K in response to excitatory stimuli *(17–19)*. These changes in mPFC excitability are also associated with a significant increase in PKA activity. However, the cellular mechanism underlying the increased mPFC excitability is not fully understood. First, although it is well-established that the activity of ion channels primarily depends on three major factors (the number of ion channels available for opening; the open probability of the channel; and the opening time of the channel when it is activated *[86]*), it is unknown which one plays a predominant role in the cocaine-induced alterations in Ca^{2+} and K^+ conductances. Second, it is unclear which subtypes of Ca^{2+} and K^+ channels are

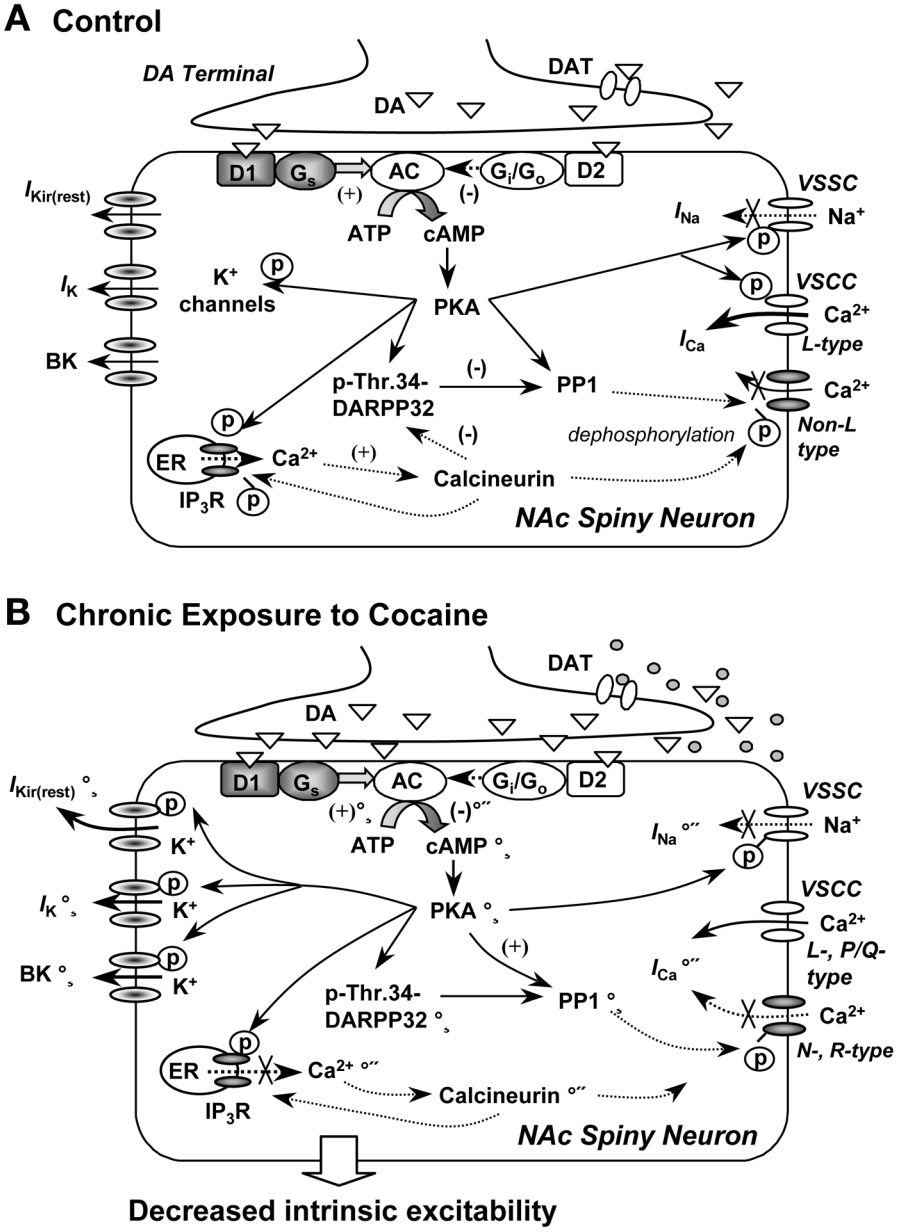
involved in the altered excitability. Third, it is not known whether other types of voltage-gated ion channels also participate in the altered PFC excitability. Finally, (and more importantly), given the differential alterations in excitability within the mPFC and NAc networks in cocaine-withdrawn animals, it needs to be determined whether and how DA/Ca^{2+} signaling (which regulates ion channel function) is interrupted.

Chronic Exposure to Cocaine Interrupts Ca^{2+} Signaling and Ion Channel Function With Increased Phosphorylation and Decreased Dephosphorylation

Decreased Ca^{2+} Signaling in Medium Spiny NAc Neurons in Cocaine-Withdrawn Animals

A question arises from previous findings regarding drug-induced alterations in ion channel function: How does chronic cocaine exposure alter the intrinsic excitability of DA-innervated neurons in drug-withdrawn animals that leads to subsequent changes in neuronal activity? Based on the information described earlier, the decreased excitability of NAc spiny neurons in cocaine-withdrawn rats could be attributed to interruption of DA/Ca^{2+} signaling with altered phosphorylation and dephosphorylation. It has been established that D1R stimulation decreases I_{Na} in drug-naïve animals, whereas this decreased I_{Na} is normally regulated by D1R-coupled PKA activation with facilitated phosphorylation of the Na^+ channel *(87–89)*. However, in medium spiny NAc neurons in psychostimulant-withdrawn animals, the reduced I_{Na} is regulated not only by an enhanced phosphorylation following increased PKA activity *(20,21,73,74, 88,89)* but also by a decreased dephosphorylation of the Na^+ channel with diminished calcineurin activity *(23,66)*. Moreover, we have also discovered that the chronic cocaine-induced decrease in dephosphorylation results from a decreased Ca^{2+} influx passing N- and R-type Ca^{2+} channels as well as a reduced cytosolic Ca^{2+} release associated with suppressed D2R function *(21–23)*. Both changes lead to a decrease in Ca^{2+} signaling (Fig. 3B).

The decreased NAc excitability may also be regulated by psychostimulant-induced changes in presynaptic DA neurons. Gene expression of the L-type Ca^{2+} channel α1-subunit *(90)* is increased in midbrain DA neurons located within the VTA following repeated amphetamine treatment *(91)*. Such an elevation in messenger RNA (and possibly in protein levels) of L-type Ca^{2+} channels would facilitate spontaneous activity of VTA DA neurons, thereby increasing DA release in the NAc and mPFC, which usually decreases the firing of NAc neurons *(12,92,93)*. Accordingly, the increased L-channel gene expression is associated with a transient enhancement in glutamate (and AMPA)-induced firing in VTA DA neurons in either cocaine- or d-amphetamine-withdrawn rats *(94)*. Importantly, even a single cocaine exposure in vivo could induce LTP in DA neurons, which lasts for a short (5 d) but not a long period *(95)*. This unique transient change in VTA DA neurons has been considered to contribute to the mechanism underlying the early stage of the development of drug addiction as well as behavioral sensitization. Because the increased excitatory responsiveness of VTA DA neurons to glutamate lasts only a few days in drug-withdrawn rats, it will be interesting to see if the increased gene expression of $Ca_v1.2$ channels persists in DA cells after a long-term withdrawal. On the other hand, investigators need to determine whether a similar increase in gene expression of L-type Ca^{2+} channels also occurs in medium spiny NAc cells in cocaine-withdrawn animals as a compensatory response to decreased whole-cell I_{Ca} and intracellular Ca^{2+} release. Therefore, it is clear that besides increased PKA activity, a decreased D2R/Ca^{2+} signaling and increased presynaptic DA release could also play an important role in the decreased NAc excitability in cocaine-withdrawn rats.



Increased Ca^{2+} Influx in mPFC Pyramidal Neurons in Cocaine-Withdrawn Animals

The increased intrinsic excitability in mPFC pyramidal neurons of drug-withdrawn rats in response to excitatory stimuli should also be attributed to the interruption of DA/Ca^{2+} signaling, but through remarkably different mechanisms. It has been demonstrated that repeated cocaine administration prolongs the duration of evoked HVA-Ca^{2+} plateau potentials with increased density of whole-cell I_{Ca} in mPFC pyramidal neurons of rats after either a short-term (3-d) or a long-term (3-wk) withdrawal *(18,19)*. These results are consistent with and supportive of previous findings that have indicated an increased glutamatergic output from the PFC of sensitized animals in response to

Fig. 3. The cellular mechanisms underlying DA D1R/D2R modulation of ion channel activity in medium spiny NAc neurons in drug-free control and cocaine-withdrawn rats. The models incorporate some well-established previous findings from other research groups and recent findings from our laboratory. **(A),** The D1R and D2R modulate ion channel activity by regulating the cAMP/PKA cascade and Ca^{2+} signaling in medium spiny NAc neurons of drug-free rats. D1R stimulation increases cAMP/PKA activity, whereas D2R stimulation decreases activity of cAMP/PKA cascade but induces intracellular Ca^{2+} release. D1R-activated PKA phosphorylates various types of ion channels, IP_3 receptors *(105)*, and other signaling-related proteins (e.g., DARPP-32 at threonine 34). D2R/Ca^{2+}/calmodulin-coupled activation of calcineurin (PP2B) dephosphorylates these substrates. Phosphorylation and dephosphorylation dynamically modulate activity of ion channels and DA/Ca^{2+} signaling-related proteins in different ways, thereby regulating the intrinsic excitability of NAc neurons. The integrated D1R and D2R modulation leads to a balanced activity of ion channels and Ca^{2+} signaling in drug-naïve NAc spiny cells. **(B),** Chronic exposure to cocaine upregulates activity of the D1R/G_s/AC/cAMP/PKA pathway. Enhanced D1R/PKA activity and reduced D2R function interrupts activity of voltage-sensitive Na^+, Ca^{2+}, K^+ channels and IP_3 receptors in cocaine-withdrawn NAc spiny cells in several ways, causing a decrease in whole-cell VSSCs (I_{Na}), VSCCs (I_{Ca}), and intracellular Ca^{2+} release, but an increase in K^+ outflow. Decreased I_{Na} and intracellular Ca^{2+} release should be attributed to an enhanced phosphorylation by PKA and a decreased dephosphorylation of the Na^+ channel and IP_3 receptors by calcineurin, respectively. Diminished I_{Ca} mainly results from increased dephosphorylation of N- and R-type Ca^{2+} channels, probably by protein phosphatase 1 (PP1) activated by PKA. Moreover, chronic cocaine exposure may also reduce dendritic propagation of Ca^{2+} and Na^+ action potentials in drug-withdrawn NAc cells. Additionally, outflow of K^+ currents is enhanced by activating various K^+ channels, including K^+ channels activated at RMP ($I_{Kir\text{-}rest}$, "leak" currents), voltage-gated slowly inactivating rectifiers (I_{Ks}), and Ca^{2+}-activated BK channels. These changes in K^+ channel function may also be associated with increased PKA activity. Together, the integrated neuroadaptations in activity of ion channels and the D1R/Gs/AC/cAMP/PKA pathway lead to a decrease in the intrinsic excitability of medium spiny NAc neurons during cocaine withdrawal. Moreover, even with drug reinstatement or other excitatory stimuli, these cells are still less excitable than drug-naïve controls. (Open arrows, cocaine-induced alterations in the ion channel function and the membrane excitability.)

cocaine-associated challenge or other excitatory stimuli *(96)*. Additionally, it has also been noted that the increased Ca^{2+} influx and prolonged duration of evoked Ca^{2+} action potentials observed in mPFC pyramidal neurons differs dramatically from that in medium spiny NAc cells, which shows a decreased I_{Ca} and reduced duration of Ca^{2+} action potentials following repeated cocaine administration.

This discrepancy in Ca^{2+} influx between mPFC pyramidal neurons and medium spiny NAc cells in cocaine-withdrawn animals is apparently related to the expression and activation of different subtypes of HVA-Ca^{2+} channels. Although decreased N- and R-type I_{Ca} in medium spiny striatal cells may be attributed to enhanced PP1 activity *(21,84)*, the increased Ca^{2+} influx in mPFC pyramidal neurons most likely results from chronic cocaine-induced adaptations in L-type Ca^{2+} channels. In fact, blocking the L-channel in mPFC pyramidal neurons of cocaine-withdrawn rats shortens the increased duration of Ca^{2+} plateau potentials to levels comparable to that found in saline-pretreated rats *(19)*. Combined with an increased PKA activity *(17)*, which facilitates phosphorylation of L-type Ca^{2+} channels *(50,51)*, these results strongly suggest that the L-type Ca^{2+} channel plays a predominant role in increasing Ca^{2+} influx in mPFC pyramidal neurons of cocaine-withdrawn rats, particularly in response to excitatory stimuli. Subsequent investigations have also revealed that the increased Ca^{2+} influx in cocaine-withdrawn mPFC pyramidal neurons is regulated not only by increased activity but also by proliferation of the L-type Ca^{2+} channel because the membrane protein levels of the L-type Ca^{2+} channel are

significantly increased in the rat mPFC *(97)*. Importantly, this distinctive change in the L-type Ca^{2+} channel is subtype- and region-specific because it is neither found in other subtypes of Ca^{2+} channels nor detected in the motor cortex of the same rats. These findings indicate that chronic cocaine-induced adaptations in DA and Ca^{2+} signaling are regulated by increased phosphorylation and decreased dephosphorylation along with integrated functional and conformational changes in certain types of voltage-gated ion channels (Figs. 2B and 3B). Further study should focus on determining whether the increased protein levels of L-type Ca^{2+} channels are functionally expressed on the surface of cocaine-withdrawn mPFC pyramidal cells.

Cocaine-Induced Neuro-Adaptations in Ion Channel Function and DA/Ca^{2+} Signaling Are Associated With Different Withdrawal Effects of Cocaine

Cocaine-induced neuroplasticity in ion channel activity that modulates the intrinsic excitability results in remarkable alterations in neuronal activity within the mPFC and NAc. These alterations in either basal or evoked neuronal activity are essential to behavioral changes in animals sensitized to psychostimulants. However, the withdrawal effects of cocaine are found to be dramatically different between drug-withdrawn subjects with certain stimuli (e.g., drug/cue challenge, increased DA or glutamate neurotransmission, and membrane depolarization) and those without such stimuli. For example, after chronic exposure to cocaine, anergia, anhedonia, apathy, and depression-like motional/behavioral changes are found in cocaine-withdrawn human addicts. Conversely, additional administration of psychostimulant (drug reinstatement) increases mood and drug craving that leads to persistent drug-seeking and drug-taking. Additionally, cocaine craving evoked by drug-associated cue also triggers an increase in PFC activity *(98–100)*. Moreover, expectation for cocaine, which is associated with drug-related learning and memory, also enhances the effects of drug reinstatement, not only on functional metabolism but also on its reinforcing action (e.g., self-report of drug-induced "high") in human addicts *(101,102)*.

Consistent with the changes in motivated behaviors in cocaine-withdrawn human addicts, cocaine-withdrawn rats show a comparable *nonsensitized* behavioral activity profile during abstinence. Morphine-abstained animals also exhibit withdrawal symptoms similar to those found in human heroin addicts. These abstinence (or "nonsensitized") behaviors are associated with a marked reduction in mesocorticolimbic DA release, which appears to be a common feature of ethanol, morphine, cocaine, and amphetamine abstinence in rats *(103)*. Moreover, as in cocaine-withdrawn human addicts, a challenge dose of reinstated cocaine also induces significant increases in locomotion and stereotypy in drug-withdrawn rats compared to saline-pretreated animals, indicating that "psychostimulant-sensitized" neurons localized in the mPFC and the NAc control and regulate the drug-stimulated behaviors.

Interestingly, recent studies have shown that with or without certain excitatory stimuli, behavioral changes in psychostimulant-sensitized animals correlate to the alterations in excitability (reflecting drug-induced maladaptations in ion channel function) and neuronal activity of both mPFC pyramidal neurons and NAc spiny cells. For example, a decreased basal neuronal activity (expressed by reduced functional glucose metabolism) is found in the PFC of cocaine-withdrawn human addicts with decreased mood, whereas an increased neuronal activity evoked by an additional drug challenge in the PFC of cocaine-withdrawn human addicts is associated with increased mood. Furthermore, in sensitized animals during withdrawal, psychostimulant-induced locomotion and stereotypy also correlate to increased excitability of mPFC pyramidal neurons in response to excitatory

stimuli. Although researchers need to identify whether the correlations observed in the subcellular (ion channels and receptors), neuronal (firing), system (DA, glutamate, and GABA), and behavioral (natural- and drug-awarded) levels are only a covariation, these findings clearly indicate involvement of altered membrane ion channel function and DA/Ca^{2+} signaling in the cellular mechanisms underlying the withdrawal effects of cocaine.

Decreased basal neuronal activity in the mPFC during cocaine withdrawal reduces the excitatory glutamatergic output to the NAc, resulting in a decrease in the excitability and basal activity of medium spiny NAc neurons. Previous studies have demonstrated that unlike mPFC pyramidal neurons, NAc spiny cells in withdrawn rats may not be able to fully recover from cocaine-induced decrease of excitability. Therefore, even with an overwhelmingly enhanced glutamatergic input from the mPFC in response to psychostimulant/cue-associated (or other excitatory) stimuli, their activity may still be lower than that found in drug-free control animals. Because cocaine-induced behavioral changes are regulated with neuroplasticity in ion channel function and DA/Ca^{2+} signaling, it is proposed that the neuro-adaptations found in mPFC pyramidal neurons and medium spiny NAc cells of drug-withdrawn animals are fundamental and critical in the development of cocaine addiction.

Conclusion

Reduced intrinsic excitability and basal activity in mPFC pyramidal neurons and medium spiny NAc neurons could contribute to the mechanism underlying certain "negative" withdrawal symptoms of cocaine regarding decreased mood, which include but are not limited to anergia, anhedonia, apathy, and depression. Therefore, medium spiny NAc neurons in withdrawn subjects after chronic exposure to cocaine would be less excitable and responsive to activation induced by non-drug-related motivational stimuli during drug abstinence. However, additional drug/cue-associated stimuli would evoke a remarkable increase in the intrinsic excitability and firing of mPFC pyramidal neurons, thereby increasing the excitatory PFC output to the NAc (and other subcortical areas). This change in the function of mPFC-NAc circuits would temporarily (and repetitively) reverse or at least alleviate the decreased NAc neuronal excitability. In response to direct psychostimulant/cue-associated stimuli, the increased neuronal activity in both the mPFC and NAc could lead to a dramatic change in motivational behaviors. Instead of a decreased motivation to seek biological rewards observed in drug-withdrawn subjects, craving would occur and uncontrollable drug-seeking behaviors would be reinforced *(96)*.

Together, these findings indicate that chronic cocaine-induced neuro-adaptations in the mesocorticolimbic DA system *(104)* result partly from the neuroplasticity in ion channel function and DA/Ca^{2+} signaling. This neural plasticity alters the intrinsic excitability and responsiveness of the cells located in the reward pathway to excitatory inputs. Although the molecular mechanism(s) underlying the alterations in neuronal excitability remain unknown, psychostimulant-induced neuroadaptations in voltage-gated ion channel function may play a critical role in the development of the cocaine-withdrawal effects. Based on the findings discussed in this article, it is concluded that cocaine-induced neuro-adaptations in mPFC pyramidal neurons and medium spiny NAc cells result partly from enhanced D1R signaling but reduced D2R function, both of which are related to dyregulation of intracellular signaling. Because cocaine-induced changes in DA neurotransmission affect both D1R and D2R population at the same time period, neuro-adaptations in the function of the two receptor classes could occur concurrently. Additionally, because an early change in the activity of VTA DA neurons may be regulated by increased glutamatergic inputs from the PFC, it is possible that functional alterations in the PFC may initiate the cocaine-induced maladaptations in the reward pathway. Figure 1 schematically summarizes the

findings in which chronic exposure to cocaine causes neuro-adaptations not only in behavioral activity of human addicts and sensitized animals but also in the neuronal activity and ion channel function of mPFC pyramidal neurons and medium spiny NAc cells, either with or without drug-associated or other excitatory stimuli (Fig. 1).

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References

1. Tzschentke T. M. (2001) Pharmacology and behavioral pharmacology of the mesocortical dopamine system, *Prog. Neurobiol.* **63,** 241–320.
2. Robbins T. W. and Everitt B. J. (2002) Limbic-striatal memory systems and drug addiction, *Neurobiol. Learn. Mem.* **78,** 625–636.
3. Volkow N. D., Fowler J. S., Wang G. J., and Goldstein R. Z. (2002) Role of dopamine, the frontal cortex and memory circuits in drug addiction: insight from imaging studies, *Neurobiol. Learn. Mem.* **78,** 610–624.
4. Nestler E. J. (2004) Molecular mechanisms of drug addiction, *Neuropharmacology* **47 (Suppl 1),** 24–32.
5. Wolf M. E. (2002) Addiction: making the connection between behavioral changes and neuronal plasticity in specific pathways, *Mol. Interv.* **2,** 146–157.
6. Pierce R. C., Reeder D. C., Hicks J., Morgan Z. R., and Kalivas P. W. (1998) Ibotenic acid lesions of the dorsal prefrontal cortex disrupt the expression of behavioral sensitization to cocaine, *Neuroscience* **82,** 1103–1114.
7. Robinson T. E., Browman K. E., Crombag H. S., and Badiani A. (1998) Modulation of the induction or expression of psychostimulant sensitization by the circumstances surrounding drug administration, *Neurosci. Biobehav. Rev.* **22,** 347–354.
8. Robinson T. E. and Berridge K. C. (2000) The psychology and neurobiology of addiction: an incentive-sensitization view, *Addiction* **95 (Suppl 2),** S91–S117.
9. Lu L., Grimm J. W., Hope B. T., and Shaham Y. (2004) Incubation of cocaine craving after withdrawal: a review of preclinical data. *Neuropharmacology* **47(Suppl 1),** 214–226.
10. Wolf M. E., Dahlin S. L., Hu X. T., Xue C. J., and White K. (1995) Effects of lesions of prefrontal cortex, amygdala, or fornix on behavioral sensitization to amphetamine: Comparison with *N*-methyl-D-aspartate antagonists. *Neuroscience* **69,** 417–439.
11. Li Y., Hu X. T., Berney T. G., et al. (1999) Both glutamate receptor antagonists and prefrontal cortex lesions prevent induction of cocaine sensitization and associated neuroadaptations. *Synapse* **34,** 169–180.
12. Xu M., Hu X. -T., Cooper D. C., et al. (1994) Elimination of cocaine-induced hyperactivity and dopamine- mediated neurophysiological effects in dopamine D1 receptor mutant mice. *Cell* **79,** 945–955.
13. Volkow N. D. (2004) Imaging the addicted brain: from molecules to behavior. *J Nucl. Med.* **45,** 13N-20N, 22N.
14. Volkow N. D., Fowler J. S., and Wang G. J. (2004) The addicted human brain viewed in the light of imaging studies: brain circuits and treatment strategies. *Neuropharmacology* **47 (Suppl 1),** 3–13.
15. Porrino L. J., Daunais J. B., Smith H. R., and Nader M. A. (2004) The expanding effects of cocaine: studies in a nonhuman primate model of cocaine self-administration. *Neurosci. Biobehav. Rev.* **27,** 813–820.
16. Porrino L. J., Lyons D., Smith H. R., Daunais J. B., and Nader M. A. (2004) Cocaine self-administration produces a progressive involvement of limbic, association, and sensorimotor striatal domains. *J Neurosci.* **24,** 3554–3562.
17. Dong Y., Nasif F. J., Tsui J. J., et al. (2005) Cocaine-induced plasticity of intrinsic membrane properties in prefrontal cortex pyramidal neurons: adaptations in potassium currents. *J Neurosci.* **25,** 936–940.
18. Nasif F. J., Hu X. T., and White F. J. (2005) Repeated cocaine administration increases voltage-sensitive calcium currents in response to membrane depolarization in medial prefrontal cortex pyramidal neurons. *J Neurosci.* **4,** 3674–3679.
19. Nasif F. J., Sidiropoulou K., Hu X. T., and White F. J. (2005) Repeated cocaine administration increases membrane excitability of pyra-

midal neurons in the rat medial prefrontal cortex. *J Pharmacol. Exp. Ther.* **312,** 1305–1313.
20. Zhang X. -F., Hu X. -T., and White F. J. (1998) Whole-cell plasticity in cocaine withdrawal: reduced sodium currents in nucleus accumbens neurons. *J Neurosci* **18,** 488–498.
21. Zhang X. F., Cooper D. C., and White F. J. (2002) Repeated cocaine treatment decreases whole-cell calcium current in rat nucleus accumbens neurons. *J. Pharmacol. Exp. Ther.* **301,** 1119–1125.
22. Hu X. -T., Basu S., and White F. J. (2004) Repeated cocaine administration suppresses HVA-Ca2+ potentials and enhances activity of K+ channels in rat nucleus accumbens neurons. *J Neurophysiol.* **92,** 1597–1608.
23. Hu X. -T., Ford K., and White F. J. (2005) Repeated Cocaine Administration Decreases Calcineurin (PP2B) but Enhances DARPP-32 Modulation of Sodium Currents in Rat Nucleus Accumbens Neurons. *Neuropsychopharmacology* **30,** 916–926.
24. Fuster J. M. (2001) The prefrontal cortex—an update: time is of the essence. *Neuron* **30,** 319–333.
25. Miller E. K. and Cohen J. D. (2001) An integrative theory of prefrontal cortex function. *Annu. Rev. Neurosci.* **24,** 167–202.
26. Seamans J. K. and Yang C. R. (2004) The principal features and mechanisms of dopamine modulation in the prefrontal cortex. *Prog. Neurobiol.* **74,** 1–58.
27. Neve K. A., Seamans J. K., and Trantham-Davidson H. (2004) Dopamine receptor signaling. *J Recept. Signal. Transduct. Res.* **24,** 165–205.
28. Yang C. R. and Seamans J. K. (1996) Dopamine D1 receptor actions in layers V-VI rat prefrontal cortex neurons in vitro: modulation of dendritic-somatic signal integration. *J Neuroscience* **16,** 1922–1935.
29. Yang C. R., Seamans J. K., and Gorelova N. (1999) Developing a neuronal model for the pathophysiology of schizophrenia based on the nature of electrophysiological actions of dopamine in the prefrontal cortex. *Neuropsychopharmacology* **21,** 161–194.
30. Gorelova N. A. and Yang C. R. (2000) Dopamine D1/D5 receptor activation modulates a persistent sodium current in rat prefrontal cortical neurons in vitro. *J Neurophysiol.* **84,** 75–87.
31. Dong Y. and White F. J. (2003) Dopamine D1–class receptors selectively modulate a slowly inactivating potassium current in rat medial prefrontal cortex pyramidal neurons. *J Neurosci.* **23,** 2686–2695.
32. Dong Y., Cooper D., Nasif F., Hu X. T., and White F. J. (2004) Dopamine modulates inwardly rectifying potassium currents in medial prefrontal cortex pyramidal neurons. *J Neurosci.* **24,** 3077–3085.
33. Brady A. M. and O'Donnell P. (2004) Dopaminergic modulation of prefrontal cortical input to nucleus accumbens neurons in vivo. *J Neurosci.* **24,** 1040–1049.
34. Wolf M. E., Sun X., Mangiavacchi S., and Chao S. Z. (2004) Psychomotor stimulants and neuronal plasticity. *Neuropharmacology* **47 (Suppl 1),** 61–79.
35. Tseng K. Y. and O'Donnell P. (2004) Dopamine-glutamate interactions controlling prefrontal cortical pyramidal cell excitability involve multiple signaling mechanisms. *J Neurosci.* **24,** 5131–5139.
36. Wang J. and O'Donnell P. (2001) D(1) dopamine receptors potentiate nmda-mediated excitability increase in layer V prefrontal cortical pyramidal neurons. *Cereb. Cortex* **11,** 452–462.
37. Seamans J. K., Durstewitz D., Christie B. R., Stevens C. F., and Sejnowski T. J. (2001) Dopamine D1/D5 receptor modulation of excitatory synaptic inputs to layer V prefrontal cortex neurons. *Proc. Natl. Acad. Sci. U. S. A* **98,** 301–306.
38. Lavin A. and Grace A. A. (2001) Stimulation of D1–type dopamine receptors enhances excitability in prefrontal cortical pyramidal neurons in a state-dependent manner. *Neuroscience* **104,** 335-346.
39. Maurice N., Tkatch T., Meisler M., Sprunger L. K., and Surmeier D. J. (2001) D1/D5 dopamine receptor activation differentially modulates rapidly inactivating and persistent sodium currents in prefrontal cortex pyramidal neurons. *J Neurosci.* **21,** 2268–2277.
40. Seamans J. K., Gorelova N. A., and Yang C. R. (1997) Contributions of voltage-gated Ca^{2+} channels in the proximal versus distal dendrites to synaptic integration in prefrontal cortical neurons. *J. Neurosci.* **17,** 5936–5948.
41. Young C. E. and Yang C. R. (2004) Dopamine D1/D5 receptor modulates state-dependent switching of soma-dendritic Ca^{2+} potentials via differential protein kinase A and C activation in rat prefrontal cortical neurons. *J. Neurosci.* **24,** 8–23.
42. Gorelova N., Seamans J. K., and Yang C. R. (2002) Mechanisms of dopamine activation of fast-spiking interneurons that exert inhibition

in rat prefrontal cortex. *J Neurophysiol.* **88,** 3150–3166.
43. Kroner S., Rosenkranz J. A., Grace A. A., and Barrionuevo G. (2005) Dopamine modulates excitability of basolateral amygdala neurons in vitro. *J Neurophysiol.* **93,** 1598–1610.
44. Mangiavacchi S. and Wolf E. M. (2004) D1 dopamine receptor stimulation increases the rate of AMPA receptor insertion onto the surface of cultured nucleus accumbens neurons through a pathway dependent on protein kinase A. *J. Neurochemastry* **88,** 1261–1271.
45. Chen L. and Yang C. R. (2002) Interaction of dopamine D1 and NMDA receptors mediates acute clozapine potentiation of glutamate EPSPs in rat prefrontal cortex. *J Neurophysiol.* **87,** 2324–2336.
46. Chen G., Greengard P., and Yan Z. (2004) Potentiation of NMDA receptor currents by dopamine D1 receptors in prefrontal cortex. *Proc. Natl. Acad. Sci. U. S. A* **101,** 2596–2600.
47. Gonzalez-Islas C. and Hablitz J. J. (2003) Dopamine enhances EPSCs in layer II-III pyramidal neurons in rat prefrontal cortex. *J. Neurosci.* **23,** 867–875.
48. Lewis B. L. and O'Donnell P. (2000) Ventral tegmental area afferents to the prefrontal cortex maintain membrane potential 'up' states in pyramidal neurons via D(1) dopamine receptors. *Cereb. Cortex* **10,** 1168–1175.
49. Tseng K. Y. and O'Donnell P. (2005) Postpubertal emergence of prefrontal cortical up states induced by D1-NMDA co-activation. *Cereb. Cortex* **15,** 49–57.
50. Hernández-López S., Bargas J., Surmeier D. J., Reyes A., and Galarraga E. (1997) D_1 receptor activation enhances evoked discharge in neostriatal medium spiny neurons by modulating an L-type Ca^{2+} conductance. *J. Neurosci.* **17,** 3334–3342.
51. Hernández-López S., Tkatch T., Perez-Garci E., et al. (2000) D_2 dopamine receptors in striatal medium spiny neurons reduce L-type Ca^{2+} currents and excitability via a novel PLCb1-IP_3-calcineurin-signaling cascade. *J. Neurosci.* **20,** 8987–8995.
52. Sun X., Gu X. Q., and Haddad G. G. (2003) Calcium influx via L- and N-type calcium channels activates a transient large-conductance Ca2+-activated K+ current in mouse neocortical pyramidal neurons. *J Neurosci.* **23,** 3639–3648.
53. Seamans J. K., Gorelova N., Durstewitz D., and Yang C. R. (2001) Bidirectional dopamine modulation of GABAergic inhibition in prefrontal cortical pyramidal neurons. *J Neurosci.* **21,** 3628–3638.
54. Trantham-Davidson H., Neely L. C., Lavin A., and Seamans J. K. (2004) Mechanisms underlying differential D1 versus D2 dopamine receptor regulation of inhibition in prefrontal cortex. *J Neurosci.* **24,** 10,652–10,659.
55. Hu X. -T. and White F. J. (1994) Loss of D1/D2 dopamine receptor synergisms following repeated administration of D1 or D2 receptor selective antagonists: electrophysiological and behavioral studies. *Synapse* **17,** 43–61.
56. Gulledge A. T. and Jaffe D. B. (1998) Dopamine decreases the excitability of layer V pyramidal cells in the rat prefrontal cortex. *J. Neurosci.* **18,** 9139–9151.
57. Henze D. A., Gonzalez-Burgos G. R., Urban N. N., Lewis D. A., and Barrionuevo G. (2000) Dopamine increases excitability of pyramidal neurons in primate prefrontal cortex. *J Neurophysiol.* **84,** 2799–2809.
58. Lavin A., Nogueira L., Lapish C. C., Wightman R. M., Phillips P. E., and Seamans J. K. (2005) Mesocortical dopamine neurons operate in distinct temporal domains using multimodal signaling. *J Neurosci.* **25,** 5013–5023.
59. An W. F., Bowlby M. R., Betty M., et al. (2000) Modulation of A-type potassium channels by a family of calcium sensors. *Nature* **403,** 553–556.
60. Bahring R., Dannenberg J., Peters H. C., Leicher T., Pongs O., and Isbrandt D. (2001) Conserved Kv4 N-terminal domain critical for effects of Kv channel-interacting protein 2. 2 on channel expression and gating. *J Biol. Chem.* **276,** 23,888–23,894.
61. Holmqvist M. H., Cao J., Hernandez-Pineda R., et al. (2002) Elimination of fast inactivation in Kv4 A-type potassium channels by an auxiliary subunit domain. *Proc. Natl. Acad. Sci. U. S. A* **99,** 1035-1040.
62. Morohashi Y., Hatano N., Ohya S., et al. (2002) Molecular cloning and characterization of CALP/KChIP4, a novel EF-hand protein interacting with presenilin 2 and voltage-gated potassium channel subunit Kv4. *J Biol. Chem.* **277,** 14,965-14,975.
63. O'Callaghan D. W., Hasdemir B., Leighton M., and Burgoyne R. D. (2003) Residues within the myristoylation motif determine intracellular targeting of the neuronal Ca2+ sensor protein KChIP1 to post-ER transport vesicles and traffic of Kv4 K+ channels. *J Cell Sci.* **116,** 4833–4845.

64. Takimoto K., Yang E. K., and Conforti L. (2002) Palmitoylation of KChIP splicing variants is required for efficient cell surface expression of Kv4. 3 channels. *J Biol. Chem.* **277,** 26,904–26,911.
65. Wang X., Zhong P., Gu Z. L., and Yan Z. (2003) Regulation of NMDA receptors by dopamine D_4 signaling in prefrontal cortex. *J. Neurosci.* **23,** 9852–9861.
66. Hu X. -T., Dong Y., Zhang X. F., and White F. J. (2005) Dopamine D2 receptor-activated Ca2+ signaling modulates voltage-sensitive sodium currents in rat nucleus accumbens neurons. *J Neurophysiol*. **93,** 1406–1417.
67. Wang X., Zhong P., and Yan Z. (2002) Dopamine D_4 receptors modulate GABAergic signaling in pyramidal neurons of prefrontal cortex. *J. Neurosci.* **22,** 9185-9193.
68. Greengard P., Allen P. B., and Nairn A. C. (1999) Beyond the dopamine receptor: the DARPP-32/protein phosphatase-1 cascade. *Neuron* **23,** 435-447.
69. Volkow N. D., Fowler J. S., Wang G. J., et al. (1993) Decreased dopamine D2 receptor availability is associated with reduced frontal metabolism in cocaine abusers. *Synapse* **14,** 169–177.
70. Volkow N. D., Wang G. J., Fowler J. S., et al. (1999) Prediction of reinforcing responses to psychostimulants in humans by brain dopamine D2 receptor levels. *Am. J Psychiatry* **156,** 1440–1443.
71. Volkow N. D., Chang L., Wang G. J., et al. (2001) Low level of brain dopamine D2 receptors in methamphetamine abusers: association with metabolism in the orbitofrontal cortex. *Am. J Psychiatry* **158,** 2015-2021.
72. Volkow N. D., Wang G. J., Fowler J. S., et al. (2002) Brain DA D2 receptors predict reinforcing effects of stimulants in humans: replication study. *Synapse* **46,** 79–82.
73. Nestler E. J., Terwilliger R. Z., Walker J. R., Sevarino K. A., and Duman R. S. (1990) Chronic cocaine treatment decreases levels of the G protein subunits G_{ia} and G_{oa} in discrete regions of rat brain. *J. Neurochem.* **55,** 1079–1082.
74. Terwilliger R., Beitner-Johnson D., Sevarino K. A., Crain S. M., and Nestler E. J. (1991) A general role for adaptations in G-proteins and the cyclic AMP system in mediating the chronic actions of morphine and cocaine on neuronal function. *Brain Res.* **548,** 100–110.
75. Nasif,F. J., White,F. J., and Hu,X. -T. (2005) Reduced dopamine D2-class receptor mediation of excitability in medial prefrontal cortex pyramidal neurons after repeated cocaine administration. Soc. Neurosci. Abstr. Program # 562. 16. Abstract
76. Beyer C. E. and Steketee J. D. (2000) Intramedial prefrontal cortex injection of quinpirole, but not SKF 38393, blocks the acute motor-stimulant response to cocaine in the rat. *Psychopharmacology (Berl)* **151,** 211–218.
77. Beyer C. E. and Steketee J. D. (2002) Cocaine sensitization: modulation by dopamine D2 receptors. *Cereb. Cortex* **12,** 526–535.
78. Henry D. J. and White F. J. (1991) Repeated cocaine administration causes persistent enhancement of D1 dopamine receptor sensitivity within the rat nucleus accumbens. *J. Pharmacol. Exp. Ther.* **258,** 882–890.
79. White F. J., Hu X. T., Zhang X. F., and Wolf M. E. (1995) Repeated administration of cocaine or amphetamine alters neuronal responses to glutamate in the mesoaccumbens dopamine system. *J. Pharmacol. Exp. Ther.* **273,** 445-454.
80. Hu X. -T., Koeltzow T. E., Cooper D. C., Robertson G. S., White F. J., and Vezina P. (2002) Repeated ventral tegmental area amphetamine administration alters dopamine D1 receptor signaling in the nucleus accumbens. *Synapse* **45,** 159–170.
81. Beurrier C. and Malenka R. C. (2002) Enhanced inhibition of synaptic transmission by dopamine in the nucleus accumbens during behavioral sensitization to cocaine. *J. Neurosci.* **22,** 5817–5822.
82. Thomas M. J., Beurrier C., Bonci A., and Malenka R. C. (2001) Long-term depression in the nucleus accumbens: a neural correlate of behavioral sensitization to cocaine. *Nat. Neurosci.* **4,** 1217–1223.
83. Yao W. D., Gainetdinov R. R., Arbuckle M. I., et al. (2004) Identification of PSD-95 as a regulator of dopamine-mediated synaptic and behavioral plasticity. *Neuron* **41,** 625-638.
84. Surmeier D. J., Bargas J., Hemmings H. C. Jr., Nairn A. C., and Greengard P. (1995) Modulation of calcium currents by a D1 dopaminergic protein kinase/phosphatase cascade in rat neostriatal neurons. *Neuron* **14,** 385-397.
85. Koh S. D., Sanders K. M., and Carl A. (1996) Regulation of smooth muscle delayed rectifier K+ channels by protein kinase A. *Pflugers Arch.* **432,** 401–412.
86. Hille B. (2001),Ionic Channels of Excitable Membrane, Sinauer Associates, Inc., Sunderland, MA, pp. 131–167.
87. Cantrell A. R., Scheuer T., and Catterall W. A. (1999) Voltage-dependent neuromodulation of

Na^+ channels by D1-like dopamine receptors in rat hippocampal neurons. *J. Neurosci.* **19,** 5301–5310.
88. Cantrell A. R., Tibbs V. C., Yu F. H., et al. (2002) Molecular mechanism of convergent regulation of brain Na(+) channels by protein kinase C and protein kinase A anchored to AKAP-15. *Mol. Cell Neurosci.* **21,** 63–80.
89. Cantrell A. R. and Catterall W. A. (2001) Neuromodulation of Na+ channels: an unexpected form of cellular plasticity, *Nat. Rev. Neurosci.* **2,** 397–407.
90. Day M., Olson P. A., Platzer J., Striessnig J., and Surmeier D. J. (2002) Stimulation of 5-HT_2 receptors in prefrontal pyramidal neurons inhibits Ca_v1. 2 L-type Ca^{2+} currents via a PLCb/IP3/calcineurin signaling cascade. *J. Neurophysiol.* **87,** 2490–2504.
91. Rajadhyaksha A. M. and Kosofsky B. E. (2005) Psychostimulants, L-type calcium channels, kinases, and phosphatases. *Neuroscientist.* **11,** 494–502.
92. White F. J., Hu X. T., and Henry D. J. (1993) Electrophysiological effects of cocaine in the rat nucleus accumbens: microiontophoretic studies. *J. Pharmacol. Exp. Ther.* **266,** 1075–1084.
93. Hu X. -T. and White F. J. (1997) Dopamine enhances glutamate-induced excitation of rat striatal neurons by cooperative activation of D1 and D2 class receptors. *Neurosci. Lett.* **224,** 61–65.
94. Zhang X. F., Hu X. T., White F. J., and Wolf M. E. (1997) Increased responsiveness of ventral tegmental area dopamine neurons to glutamate after repeated administration of cocaine or amphetamine is transient and selectively involves AMPA receptors. *J. Pharmacol. Exp. Ther.* **281,** 699–706.
95. Ungless M. A., Whistler J. L., Malenka R. C., and Bonci A. (2001) Single cocaine exposure *in vivo* induces long-term potentiation in dopamine neurons. *Nature* **411,** 583–587.
96. Kalivas, P. W. and Hu, X. -T. (2006) Exciting inhibition in psychostimulant addiction. *Trends Neurosci.* **29,** 610–616.
97. Grevers,C. M., Nasif,F. J., White,F. J., and Hu,X. -T. (2005) Repeated cocaine administration increases L-type calcium channel protein levels in rat medial prefrontal cortex. *Soc. Neurosic. Abstr.* Program # 562. 17. Abstract
98. Grant S., London E. D., Newlin D. B., et al. (1996) Activation of memory circuits during cue-elicited cocaine craving. *Proc. Natl. Acad. Sci. U. S. A* **93,** 12,040–12,045.
99. Maas L. C., Lukas S. E., Kaufman M. J., et al. (1998) Functional magnetic resonance imaging of human brain activation during cue-induced cocaine craving. *Am. J Psychiatry* **155,** 124–126.
100. Childress A. R., Mozley P. D., McElgin W., Fitzgerald J., Reivich M., and O'Brien C. P. (1999) Limbic activation during cue-induced cocaine craving. *Am. J Psychiatry* **156,** 11–18.
101. Volkow N. D., Wang G. J., Ma Y., et al. (2003) Expectation enhances the regional brain metabolic and the reinforcing effects of stimulants in cocaine abusers. *J Neurosci.* **23,** 11,461–11,468.
102. Volkow N. D., Fowler J. S., Wang G. J., and Swanson J. M. (2004) Dopamine in drug abuse and addiction: results from imaging studies and treatment implications. *Mol. Psychiatry* **9,** 557–569.
103. Rossetti Z. L., Hmaidan Y., and Gessa G. L. (1992) Marked inhibition of mesolimbic dopamine release: a common feature of ethanol, morphine, cocaine and amphetamin abstinence in rats. *Eur. J. Pharmacol.* **221,** 227–234.
104. Simmons J. M., Ackermann R. F., and Gallistel C. R. (1998) Medial forebrain bundle lesions fail to structurally and functionally disconnect the ventral tegmental area from many ipsilateral forebrain nuclei: Implications for the neural substrate of brain stimulation reward. *J. Neurosci.* **18,** 8515–8533.
105. Xia J. M., Simonyi A., and Sun G. Y. (1998) Changes in IP_3R1 and SERCA2b mRNA levels in the gerbil brain after chronic ethanol administration and transient cerebral ischemia-reperfusion. *Mol. Brain Res.* **56,** 22–28.