From Signaling Molecules to Circuits and Behaviors: Cell-Type-Specific Adaptations to Psychostimulant Exposure in the Striatum

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Biological Psychiatry

Review

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ABSTRACT

Addiction is characterized by a compulsive pattern of drug seeking and consumption and a high risk of relapse after withdrawal that are thought to result from persistent adaptations within brain reward circuits. Drugs of abuse increase dopamine (DA) concentration in these brain areas, including the striatum, which shapes an abnormal memory trace of drug consumption that virtually highjacks reward processing. Long-term neuronal adaptations of gamma-aminobutyric acidergic striatal projection neurons (SPNs) evoked by drugs of abuse are critical for the development of addiction. These neurons form two mostly segregated populations, depending on the DA receptor they express and their output projections, constituting the so-called direct (D_1 receptor) and indirect (D_2 receptor) SPN pathways. Both SPN subtypes receive converging glutamate inputs from limbic and cortical regions, encoding contextual and emotional information, together with DA, which mediates reward prediction and incentive values. DA differentially modulates the efficacy of glutamate synapses onto direct and indirect SPN pathways by recruiting distinct striatal signaling pathways, epigenetic and genetic responses likely involved in the transition from casual drug use to addiction. Herein we focus on recent studies that have assessed psychostimulant-induced alterations in a cell-type–specific manner, from remodeling of input projections to the characterization of specific molecular events in each SPN subtype and their impact on long-lasting behavioral adaptations. We discuss recent evidence revealing the complex and concerted action of both SPN populations on drug-induced behavioral responses, as these studies can contribute to the design of future strategies to alleviate specific behavioral components of addiction.

Keywords: Addiction, Dopamine receptor, Gene regulation, Signaling, Striatal projection neuron, Striatum

https://doi.org/10.1016/j.biopsych.2019.11.001

Drug addiction is defined as a compulsive pattern of drug-seeking and drug-taking behavior, with recurrent episodes of abstinence and relapse, and a loss of control despite negative consequences. A current hypothesis is that persistent behavioral alterations characterizing addiction result from drug-evoked long-term changes in synaptic efficacy involving the early recruitment of specific signaling cascade and gene expression (1). This continuum between synaptic and nuclear events shapes an enduring remodeling of the reward circuitry likely involved in the transition from casual drug use to addiction (2–4). This drug-induced memory trace is persistent and tightly related to the context of drug consumption, partly explaining the high risk of relapse even after long periods of abstinence. Addictive drugs promote reinforcement by increasing dopamine (DA) in the mesocorticolimbic system (5), which alters excitatory glutamate transmission within the reward circuitry and hijacks reward processing (1). The striatum is a key target structure of drugs of abuse because it is at the crossroad of converging glutamate inputs from limbic, thalamic, and cortical regions, which encode components of drug-associated stimuli and environment, and DA, which mediates reward prediction error and incentive values. These signals are integrated in gamma-aminobutyric acidergic striatal projection neurons (SPNs), which receive glutamate and DA axons converging onto their dendritic spines (6,7). SPNs primarily form two mostly distinct populations based on the expression of either DA D_1 receptors (D_1Rs) or D_2 receptors (D_2Rs) (8,9) which are G protein–coupled receptors positively and negatively modulating adenylyl cyclase though their respective coupling to $G_{s/olf}$ and $G_{i/o}$ subtypes (10,11). While a classical view is that the two populations of SPNs act in parallel, playing antagonistic functional roles, the picture seems much more complex, as discussed below.

Initial studies based on the use of DA receptor agonists or antagonists led to somewhat confounding results on the role of D1R-SPNs and D2R-SPNs in drug-evoked adaptations (12). A major limitation of such strategies is the widespread effect of these compounds in the brain, with different specificity, pharmacokinetics, and downstream effects relative to presynaptic and postsynaptic receptors. Analyzing molecular events in identified neuronal populations was made possible by the development of reporter mouse lines expressing fluorescent proteins or Cre-recombinase under the control of cell-specific promoters *drd1a* and *drd2/adora2a* for D1R-SPNs and D2R-

ISSN: 0006-3223

SPNs, respectively (13–17). From circuits to molecules, cell-type–specific manipulations are now achievable owing to targeted chemogenetic and optogenetic technologies, conditional knockout, or acting on specific protein-protein interactions. This deepened our understanding of the differential impact of drug exposure on these two neuronal populations, from alterations of the inputs they receive to the pattern of signaling cascades and transcriptional landscape they exhibit at various stages of addiction. Focusing on studies using the psychostimulants cocaine and amphetamine, which constitute the vast majority of the work regarding cell-specific functions in addiction, we discuss the literature linking drug-induced behavioral alterations with adaptations in D1R-SPNs and D2R-SPNs at the circuit, cellular, and molecular levels.

CELL-TYPE–SPECIFIC MODULATION OF SPN ACTIVITY

A current hypothesis is that the transition from recreational to compulsive drug use relies on a gradual recruitment from ventromedial to dorsolateral striatal subregions (18,19). Despite the existence of behavioral features of vulnerability toward addiction (19), few preclinical studies support the intriguing possibility that individuals who develop behavioral traits of addiction display plasticity-related alterations in the striatum (20). Nonetheless, most studies have focused on remodeling of neural circuits in the ventral striatum (i.e., nucleus accumbens [NAc]) and dorsomedial striatum (DMS) induced by early drug exposure (Supplemental Table S1), which does not reflect addiction per se but might constitute a main trigger toward drug abuse.

SPNs constitute 95% of striatal neurons, the remaining 5% being local interneurons, which include large tonically active cholinergic interneurons. Even though cholinergic interneurons are major players in the modulation of striatal microcircuits (21), their implication in drug responses is beyond the scope of the current review.

In the dorsal striatum (DS), a common view is that D1R-SPNs and D2R-SPNs exert opposite effects through distinct output projections, with D2R-SPN reaching the midbrain through polysynaptic projections via the external globus pallidus (indirect pathway), while D1R-SPN directly project onto the internal globus pallidus and the substantia nigra (direct pathway). However, a subset of D1R-SPNs displays projections to the globus pallidus (22), which can be augmented under certain conditions (23). This dichotomy becomes even more erroneous regarding the NAc because 2% to 5% of core SPNs express both DA receptors (24,25), while in the shell, this proportion varies from 2% to 5% (25) up to 10% to 15% (24), depending on the methodologies used. Moreover, even though there exists a recent controversy as to whether they constitute a distinct subpopulation or they form collaterals, a large proportion of NAc D1R-SPNs projects in the ventral pallidum (VP), which is the canonical output of D2R-SPNs (25–27).

Consistent with the so-called Go/NoGo model of basal ganglia in which D1R-SPN activity facilitates and D2R-SPN activity inhibits movement planning/initiation (28–30), cell-type–specific manipulations initially supported that SPNs from the NAc and DMS play antagonistic roles on reward processing, including drug-induced behaviors (31,32). Indeed, the selective ablation of NAc D2R-SPNs (17) or their transient chemogenetic inhibition in the DS (33) enhances psychostimulant-induced conditioned place preference (CPP) and locomotor sensitization, respectively. Conversely, D2R-SPN chemogenetic activation in the entire striatum blocks amphetamine-induced sensitization (34), while their optogenetic stimulation in the NAc reduces cocaine CPP (31) and alleviates locomotor sensitization when applied during the withdrawal period (35). On the other hand, optogenetic activation of NAc D1R-SPNs enhances cocaine-induced CPP (31), whereas their inhibition in the DS (33) or the NAc (36,37) or their reversible blockade in the entire striatum (38) decreases psychostimulant-induced locomotor sensitization or CPP. Regarding operant behavior, NAc D2R-SPN chemogenetic inhibition enhances motivation to obtain cocaine, whereas their activation by optogenetics diminishes drug seeking (39). Conversely, chemogenetic inhibition of D1R-SPNs in the DMS reduces cue-induced reinstatement of cocaine seeking while sparing escalation, maintenance, and incentive components (40).

Altogether, these studies support a binary model in which D1R-SPNs and D2R-SPNs primarily form two parallel pathways promoting and reducing psychostimulant-induced adaptations, respectively. However, this dichotomic view overlooks several anatomofunctional findings. Notably, selective inhibition of NAc D1R-SPN projections to the VP reduces cue-induced reinstatement of cocaine seeking, suggesting that specific D1R-SPN projections might control distinct components of addictive behaviors (26). Accordingly, stimulating NAc shell projections to lateral hypothalamus [likely originating from D1R-SPNs (41)] enhances the motivation to self-administer cocaine and facilitates drug seeking, while global NAc shell activation accelerates extinction of this behavior (42). This latter finding could result from the concomitant stimulation of both SPN populations, which is likely to differentially impact striatal microcircuits. Along the same line, the “lateral inhibition” of D2R-SPNs onto D1R-SPNs in the NAc appears as a major mechanism by which D2R-SPNs could participate to locomotor sensitization by favoring D1R-SPN activity (43,44). Moreover, cocaine can inhibit D2R-SPN synaptic transmission onto VP neurons in a DA-independent but serotonin-dependent manner (45), the behavioral consequences of which remain unknown. Functionally, other findings challenge the view of opposite actions of the two SPN populations on drug-induced behavioral adaptations. In the DMS, ablation of D1R-SPNs decreases acute amphetamine-induced locomotor response without affecting sensitization, whereas ablating D2R-SPNs decreases sensitization but spares acute locomotor responses (46). Similarly, the transient inhibition of synaptic transmission of D2R-SPNs in the DMS delays the development of locomotor sensitization, although to a lower extent than D1R-SPN inhibition does (38).

Studies on the control of nondrug reward processing reinforce the controversy regarding the antagonistic roles of the two SPN populations. Indeed, optogenetic-mediated manipulations of D1R-SPNs and D2R-SPNs suggest that they can mediate “pro-rewarding” effects (47–52) or “aversion” depending on the type of manipulation (53), even though chemogenetic inhibition of D2R-SPNs leads to discrepant results (54,55). These studies highlight that D1R-SPNs and D2R-SPNs may rather act in a dynamic and concerted fashion

to control behavior. Accordingly, both populations are activated at the same time in the dorsolateral striatum during initiation of a reward-oriented action (49,56), which has been proposed to allow proper action sequence initiation (57). In addition to challenging the antagonism of both SPN populations, these findings call for caution regarding the interpretation of the results obtained by direct manipulations of SPNs on drug-related behaviors because they could result from perturbations of reward processing per se, rather than reflecting specific alterations of drug-induced behavioral adaptations.

In this context, Creed *et al.* (58) demonstrated that repeated cocaine exposure potentiates and depresses NAc D1R-SPN and D2R-SPN synapses, respectively, onto VP neurons. Optogenetically mediated depotentiation of D1R-SPN transmission onto the VP abolishes sensitized locomotor response, while potentiation of D2R-SPN-to-VP projections restores operant responding for sucrose in animals under cocaine withdrawal. These data suggest that D1R-SPN and D2R-SPN projections onto VP neurons mediate behavioral sensitization and cocaine-induced anhedonia, respectively (58). Yet, either chemogenetic activation of D1R-SPN or inhibition of D2R-SPN potentiates cocaine self-administration, which is reversed by chemogenetic inhibition of VP neurons (59).

Neurons specifically contacting each SPN subpopulation display distinct drug-induced structural plasticity, supporting input-specific alterations (60). In accordance, Luscher's group showed that cocaine-induced locomotor sensitization is associated with a potentiation of excitatory cortical inputs onto D1R-SPNs, but not D2R-SPNs, of the NAc (61). Optogenetically induced depotentiation of these synapses established the causality between drug-evoked plasticity in D1R-SPNs and behavioral sensitization (61). They also showed that mice trained to self-administer cocaine and subjected to a 30-day withdrawal display a potentiation of transmission at ventral hippocampal and cortical, but not amygdalar, excitatory projections onto NAc D1R-SPNs. Specific optogenetically induced patterns of stimulation decrease vigor response and abolish cocaine seeking when hippocampal and cortical inputs, respectively, are depotentiated (62). The same group also showed that compulsive self-stimulation of DA transmission, which models drug addiction, relies on the potentiation of excitatory projections from the orbitofrontal cortex onto D1R-SPNs of the ventrocentral striatum (63). These studies highlight that distinct components of drug addiction rely on drug-evoked synaptic adaptations at specific input projections onto D1R-SPNs. Accordingly, repeated cocaine exposure strengthens afferences from the basolateral amygdala onto D1R-SPNs but not D2R-SPNs (64). Inputs onto D2R-SPNs also seem to be altered by drug exposure because cocaine-induced CPP correlates with a strengthening of the coupling between hippocampal place cells and D2R-SPNs (65). However, further studies are necessary to establish causality between neurobiological adaptations onto D2R-SPNs and addictive behavior.

Beyond the comprehension of the mechanisms that underlie addiction, unraveling the precise alterations in synaptic connectivity between SPNs and input areas could inspire therapeutic strategies to reverse drug-induced synaptic changes, for instance through deep-brain or transcranial magnetic stimulation (66,67). Moreover, the identification of specific drug-evoked synaptic changes highlights that addiction involves perturbation in the balance between glutamatergic inputs and DA modulatory signaling onto SPNs. This calls for a better understanding of the underlying molecular mechanisms as it may lead to identification of innovative molecular target (68).

CELL-TYPE–SPECIFIC STRIATAL SIGNALING FROM THE MEMBRANE TOWARD THE NUCLEUS

Through differential coupling of DA receptors to $G_{s/olf}$ and $G_{i/o}$, DA activates the cAMP (cyclic adenosine monophosphate)/downstream PKA (protein kinase A) pathway in D1R-SPNs, while repressing it in D2R-SPNs, leading to opposite regulations of ion channels, including glutamate receptors (69–71). The consensus is that DA increase facilitates glutamate-dependent activation of D1R-SPNs and inhibits glutamate-dependent activation of D2R-SPNs. Accordingly, acute cocaine administration triggers a fast D_1R-mediated Ca^{2+} increase in D1R-SPNs and a slow D_2R-dependent deactivation of D2R-SPNs in the DS of anesthetized mice (72). In freely moving mice, acute cocaine administration does not influence global neuronal activity in either subpopulation of the DS, although compact clusters of activity were identified in discrete subregions of the DS (73). Thus, cocaine-induced hyperlocomotion correlates with increased activity in D1R-SPN clusters near the dorsolateral striatum and a decrease in D2R-SPN clusters near the DMS (73). During cocaine-induced CPP, a transient Ca^{2+} rise occurs in NAc D1R-SPNs before entry in the drug-paired compartment, while Ca^{2+} decreases in D2R-SPN when the animal stays in this compartment (36). The mechanisms by which DA controls Ca^{2+} signaling were initially investigated through cell-specific overexpression in D1R-SPNs of NMDA glutamate receptor (NMDAR) bearing reduced Ca^{2+} permeability, which prevents the sensitizing and rewarding effects of cocaine (74), whereas the same manipulation in D2R-SPNs did not impact amphetamine-mediated CPP (75). NMDAR knockout in D_1R-expressing cells also blocks amphetamine-induced sensitization, a phenotype that is rescued by restoring functional NMDAR in NAc D1R-SPNs or deleting NMDAR in all SPNs (76). By contrast, NMDAR knockout in either D_1R cells or adenosine A_{2A} receptor (A_{2A}R) cells (overlapping with D2R-SPNs), or both, preserves the development and extinction of cocaine-induced CPP (77). However, NMDAR deletion in D_1R, but not A_{2A}R, cells blunts CPP reinstatement, which is partially rescued by full NMDAR deletion (77). These findings highlight the critical role of a balanced NMDAR activity in each cell type for drug-induced responses. Although constitutive, and not striatal specific, these manipulations support a critical role of striatal DA and NMDAR signaling crosstalk in drug-evoked responses.

Downstream from these receptors, the ERK (extracellular signal-regulated kinase) pathway is activated by virtually all drugs of abuse (78). Global ERK inhibition blocks long-term potentiation of glutamate synapses impinging onto D1R-SPNs (61), cocaine-induced locomotor sensitization, and CPP (79,80) and the reconsolidation of drug-associated memories (81,82). Acute cocaine activates ERK in D1R-SPNs (24) via a mechanism that depends on both D1R and NMDAR (79,83). ERK activation thus behaves as a coincidence detector of

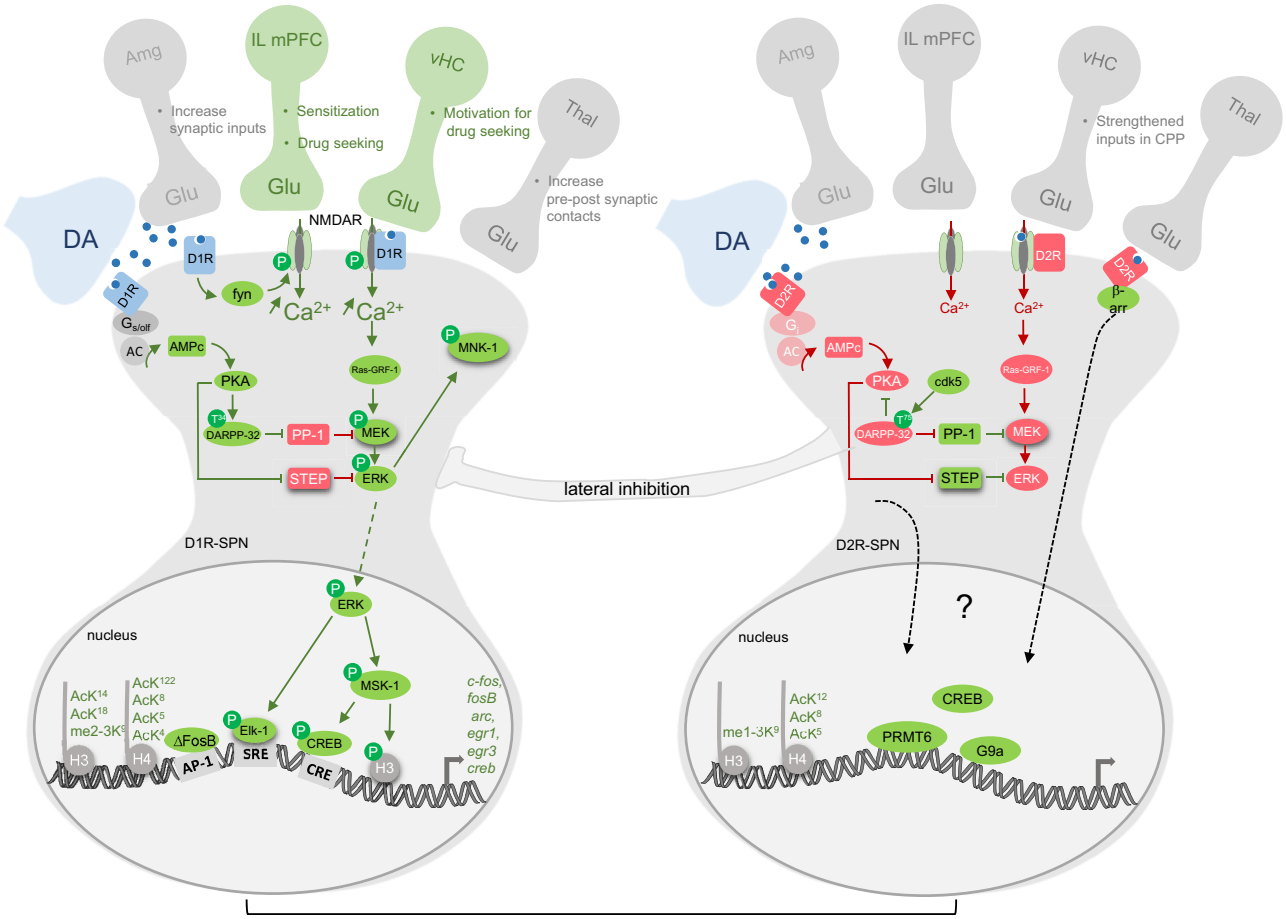

Figure 1. Cell-type–specific cellular and molecular events recruited from the plasma membrane to the nucleus in striatal projection neurons (SPNs) in response to psychostimulants. Diagram depicting major striatal signaling pathways and epigenetic and genic responses taking place from the membrane to the nucleus in (left) dopamine D_1 receptor (D1R)– or (right) D_2 receptor (D2R)–expressing SPNs in response to psychostimulants. Green and red arrows and lanes represent mechanisms described in the main text that are activated and inhibited, respectively, on dopamine release induced by psychostimulants. Glutamate afferences impinging onto each cell type are represented on top of each SPN subtype. Green afferences represent the ones for which a modulation of glutamate transmission induced by psychostimulants has been causally linked to distinct components of behavioral responses. Gray afferences represent the ones for which psychostimulant-induced changes in glutamate transmission have been either correlated to behavioral responses or not modified. AC, adenylate cyclase; Ack, acetyl-lysine; Amg, amygdala; AMPc, cyclic adenosine monophosphate; AP-1, activator protein-1; arc, activity-regulated cytoskeleton-associated protein; β-arr, β-arrestin; cdk5, cyclin-dependent kinase 5; CPP, conditioned place preference; CRE, calcium- and cyclic-AMP responsive element; CREB, cyclic adenosine monophosphate–responsive element binding protein; DA, dopamine; DARPP-32, dopamine and cyclic adenosine monophosphate–regulated phosphoprotein; egr1, early growth response 1; Elk-1, ETS-like-1 protein; ERK, extracellular signal-regulated kinase; Glu, glutamate; IL mPFC, infralimbic medial prefrontal cortex; me1-3K^9, mono-, di-, tri-methyl-lysine; me2-3K^9, di- tri-methyl-lysine 9; MEK, MAPKinase/ERK kinase; MNK-1, mitogen-activated protein kinase interacting protein-1; MSK-1, mitogen and stress-activated protein kinase-1; P, phosphorylation; PKA, protein kinase A; PP1, protein phosphatase 1; PRMT6, protein arginine methyltransferase 6; Ras-GRF-1, ras-guanine releasing factor-1; SRE, serum response element; STEP, striatal-enriched protein tyrosine phosphatase; T, threonine residue; Thal, thalamus; vHC, ventral hippocampus.

glutamate and DA signaling (84). This occurs through a cAMP-independent and D1R-mediated facilitation of GluN2B-containing NMDAR, triggering Ca^{2+}-dependent ERK activation (83,85,86) (Figure 1). Nevertheless, inhibiting D_1R-mediated potentiation of NMDAR targeting ERK activation in D1R-SPNs blocks the sensitizing and rewarding effects of cocaine (83). We also found that in NAc shell D1R-SPNs, acute cocaine triggers rapid de novo formation of dendritic spines contacting preexisting glutamate axon terminals (87). Stabilization of these newly formed synapses requires the targeting of the cytoplasmic MNK-1 (mitogen-activated protein kinase interacting protein-1) by ERK, which controls local translation independently of nuclear events, and likely influences responses to subsequent drug exposure (87).

Downstream from D_1R, the cAMP/PKA pathway also contributes to the facilitation of NMDAR functions by directly targeting specific subunits (88). Although PKA cannot directly target ERK, it can indirectly amplify its activation, notably via the cAMP-regulated phosphoprotein DARPP-32 [described in (85,86,89)]. Consistent with a D1R-SPN-specific ERK induction, cocaine increases DARPP-32 activity in D1R-SPNs and decreases it in D2R-SPNs (90). Deleting DARPP-32 in D1R-SPNs reduces basal locomotion and cocaine-induced hyperlocomotion, while its deletion in D2R-SPNs leads to an

opposite phenotype (91). Because DARPP-32 full knockout inhibits cocaine responses (89), cAMP/PKA/DARPP-32 signaling seems to prevail in D1R-SPNs over D2R-SPNs. Other possible pathways linking the D_1R and cAMP/PKA pathways to ERK activation include the striatal-enriched protein phosphatase STEP, a phosphatase of ERK that is inhibited by PKA (85,89,92), and the neuritogenic cAMP sensor NCS-Rapgef2, for which ablation blocks cocaine-induced ERK activation in the NAc (93).

Despite the importance of the crosstalk between striatal DA and glutamate signaling for drug-induced adaptations, targeting of cognate receptors in humans to alleviate addiction led to a lack of efficacy and/or side effects over time (86), likely due to perturbation of crucial physiological functions. The development of biased ligands, recruiting specific pathways, opens the way toward the modulation of specific behavioral components (94). Little is known about their relevance in addiction, notably whether they could alleviate specific drug-evoked behavioral components while sparing nondrug reward processing. Interestingly, transgenic approaches show that biasing D_2R toward arrestin signaling in the NAc affects cocaine-induced locomotion but not reward processing (95).

The physical interaction of DA receptors with other receptors also appears as a powerful mechanism by which receptors can mutually modify their functions through allosteric interactions resulting in functional selectivity. Hence, receptor heteromers are emerging as promising targets for fine-tuning of specific signaling pathways (96–98). One of the best-characterized receptor complexes is D_2R–$A_{2A}R$ heteromers, which are detected in vivo in the striatum (99) and whose implication in reward and addiction is well reviewed (98,100). The key role of DA receptor–NMDAR interaction for their reciprocal modulation (101–104) makes these heteromers particularly relevant for addiction. Endogenous heteromers formed by D_1R and the GluN1 NMDAR subunits are detected in the mouse striatum and act as a molecular bridge by which DA and glutamate exert their synergistic action on responses to cocaine in D1R-SPNs (105). Ex vivo electrophysiological recordings from D1R-SPN reporter mice show that the disruption of D_1R/GluN1 interaction impedes D_1R-mediated potentiation of NMDA postsynaptic currents; impedes long-term synaptic plasticity in D1R-SPNs, but not D2R-SPNs; and impedes cocaine-induced ERK activation (105). By contrast, D_2R/GluN2B interaction mediates the inhibition of NMDA currents by DA in D2R-SPNs and alters the acute hyperlocomotor effect of cocaine (106). Hence, DA receptor/NMDAR heteromers may play a role in the imbalance between D1R-SPNs and D2R-SPNs induced by drugs of abuse, even though further work is needed to characterize their impact on downstream signaling toward the nucleus and long-lasting behavioral responses in vivo.

GENETIC AND EPIGENETIC REGULATION

Like other forms of memory, addiction-related memories require changes in gene regulation and protein expression (107,108) taking place downstream from the activation of cytoplasm-to-nucleus signaling. Once activated in D1R-SPNs (24), ERK directly targets the transcription factor Elk-1 (ETS-like-1 protein) and indirectly the Ca^{2+}-binding protein and CREB (cAMP-responsive element binding protein) via the MSK-1 (mitogen and stress-activated protein kinase-1) (109–111). Cocaine-triggered Elk-1 phosphorylation downstream of D_1R regulates the induction of immediate early genes (IEG), including *c-fos*, *zif268, Delta-fosB,* and *Arc* (110). As a consequence, inhibiting ERK-mediated Elk-1 phosphorylation blunts the sensitizing and rewarding effects of cocaine (110). By contrast, *Msk1*-deficient mice, which show altered histone H3 phosphorylation [a major cocaine-induced epigenetic mark in D1R-SPNs (24,111)], exhibit a down-regulation of c-Fos and dynorphin expression, but not of Zif268 expression, along with decreased locomotor sensitization but spared CPP (111).

The contribution of D1R-SPN-induced genes was initially established at the level of IEG through pharmacological studies and further confirmed using reporter mice (24,112). More comprehensive evidence came from the molecular profiling of FACS (fluorescence-activated cell sorting)–isolated c-Fos-positive neurons showing an enrichment of D1R-SPNs over D2R-SPN-specific genes, along with IEG (including Arc [activity-regulated cytoskeleton-associated protein] and FosB) in this cocaine-activated population (113). Functionally, D1R-SPN-specific knockout of *c-fos* alters cocaine-induced expression of neuronal plasticity-related gene, dendritic remodeling, and locomotor sensitization (114). ΔFosB, the stable spliced version of FosB IEG, persistently accumulates in NAc D1R-SPNs after chronic psychostimulant exposure and plays a key role in cocaine addiction (107). This cell-type–specific ΔFosB expression was further confirmed in reporter lines (115,116) or using ribosomal tagging approaches (117). On the other hand, D1R-SPN-specific overexpression of ΔFosB led to increased silent synapses and immature spines formation (118), along with enhanced behavioral responses to cocaine and an increased sensitivity to this drug at low doses (118–120).

Psychostimulant exposure also affects IEG encoding effector proteins such as Arc, which is induced in the striatum downstream of D_1R and ERK activation (121–123). Owing to its peculiar dendritic expression, the role of Arc in neuronal plasticity has been extensively described at synapses (124,125). However, acute cocaine induces an enrichment of Arc in the nucleus, where it colocalizes with active transcription regions and phosphorylated histones H3 (123). Consistently, *Arc*-deficient mice exhibit decreased chromatin compaction, higher RNA-polymerase II activity, and enhanced c-Fos expression, along with exacerbated cocaine-mediated CPP (123), revealing a homeostatic role of Arc in cocaine-induced gene expression. Other psychostimulant-regulated IEGs include genes encoding proteins involved in various cell functions, including mitochondrial or metabolic, which are differentially regulated in each SPN subpopulation after cocaine and play a key role in neuronal and behavioral adaptations to this drug (126–129).

Cell-type–specific genome-wide approaches, including FACS array profiling from D1R-SPN and D2R-SPN reporter lines (130) or cell-type–specific affinity purification of polysomal messenger RNAs (117), revealed major differences in transcriptional landscapes between each subpopulation. A similar approach based on the use of Ribotag mice identified the transcription factor Egr3 (early growth response 3) as

differentially regulated by chronic cocaine use in the two SPN subtypes with an increase in D1R-SPNs and a decrease in D2R-SPNs (131). Chromatin immunoprecipitation demonstrated the binding of Egr3 to promoters of neuronal plasticity-associated genes CamK2α (calcium/calmodulin-dependent protein kinase IIα) and CREB, which were further shown increased in D1R-SPNs and decreased in D2R-SPNs (131).

The persistent and experience-dependent features of addiction have suggested a role for epigenetic modifications, as they mediate stable transcriptional alterations supporting long-term remodeling of neuronal networks (4). Drugs affect multiple epigenetic processes including histone post-translational modifications, long-range chromatin reorganization, and noncoding RNAs (132).

Histone posttranslational modifications, including acetylation, methylation, or phosphorylation, gate DNA accessibility for transcription (133) and have been extensively studied in addiction (134). In particular, cocaine exposure induces a global increase in H3 and H4 acetylation (135–137), a transcription-permissive epigenetic mark (138). Chromatin immunoprecipitation–based analyses have been instrumental in understanding the contribution of these epigenetic changes to specific candidate gene regulation (132), as illustrated by H4 hyperacetylation at the *c-fos* promoter after acute, but not chronic, cocaine exposure (135). A genome-wide analysis of H3 and H4 pan-acetylation extended the mapping of cocaine-induced histone acetylation alterations at many genes predominantly induced in D1R-SPNs including *Arc*, *c-fos*, and *Egr3* (137). Direct assessment of cell-specific histone acetylation on FACS D1R-SPNs or D2R-SPNs (139) shows an increase in H3 and H4 acetylation in both populations after acute cocaine exposure, while H4 acetylation remain enriched only in D2R-SPNs after chronic exposure. This study also confirms the D1R-SPN-specific increase in Ser10-PH3 associated with *c-fos* transcriptional activation (140).

Histone lysine methylation is another cocaine-regulated modification that can either activate or repress transcription depending on the specific lysine (in this case, lysine K) residue targeted and its valence of methylation (141). In the NAc, chronic cocaine decreases the global level of dimethylation and trimethylation of H3K9 residue and downregulates its catalyzing enzymes, G9a histone methyltransferase (142). A ribosomal affinity purification approach shows that G9a expression is predominantly affected in D2R-SPNs in the whole striatum after cocaine exposure, and its specific knockdown in D2R-SPNs shifts these SPNs to a D1R-SPN phenotype, resulting in enhanced response to cocaine (143). NAc-specific profiling further demonstrates that the reduction of G9a specifically occurs in D1R-SPNs (131). Similarly, histone lysine methylation profile shows cell-specific regulations, depending on time and regimen of cocaine (139). Arginine methylation is decreased in D2R-SPNs together with downregulation of both PRMT6 (protein arginine methyltransferase 6) and its associated mark, asymmetric demethylation of R2 on histone H3 (H3R2me2a), after repeated exposure to cocaine (144). While direct methylation of DNA at specific promoter regions also plays a key role in gene regulation and cocaine-related behaviors (4), its cell-type specificity is yet unknown.

Growing evidence suggests a critical role for persistent changes in chromatin architecture in modulating subsequent neuronal responses during drug reexposure via a mechanism referred to as “gene priming.” This process could be involved in cell-type–specific long-term transcriptional alterations as recent observations show differential changes in chromatin accessibility between D1R-SPNs and D2R-SPNs after chronic cocaine exposure (145). Overall these data indicate that epigenetic remodeling could be critical in shaping the transcriptional program of D1R-SPNs and D2R-SPNs, although further investigations are now required to understand the cell-specific contribution of these processes.

Finally, much attention has been paid to microRNAs (miRNAs), a category of 21- to 25-nucleotides-long noncoding RNAs, which are altered in several psychiatric conditions (146), including addiction (147). miRNA, by repressing messenger RNA translation (148), are considered as “master regulators” of long-lasting transcriptional adaptations. miRNA-mediated gene regulation plays a role in cocaine-related changes in neurotransmission and behavior (148), but their cell-type–specific role in the striatum remains unknown. One study began to broadly address this issue by ablating a key protein in miRNA processing, Ago2, in D2R-SPNs (149). These mice showed loss of motivation to self-administer cocaine and a decrease of 23 (of 63) miRNAs induced by acute cocaine exposure. Questions as to whether and how miRNA regulation in D1R-SPNs occurs, along with the specific role of miRNAs in this neuronal population, remain to be answered.

CONCLUSIONS AND PERSPECTIVES

While converging data point at a critical role for D1R-SPN-specific plasticity, cell-type–specific approaches reveal evident molecular alterations in D2R-SPNs in favor of their active role in the reshaping of striatal circuits in addiction. However, we still lack a comprehensive model for the contribution of intracellular pathways involved in drug-induced transcriptional alterations in D1R-SPNs and D2R-SPNs at various stages of addiction. Cell-type–specific approaches and timed-controlled interventions combined with genome-wide analysis of gene expression, epigenetic marks, and chromatin structure should deepen our understanding of the synergistic role of these two populations in drug-induced behavioral alterations. Establishing causality between drug-induced behavioral adaptations and discrete molecular and cellular mechanisms specific to each SPN population could allow the design of novel treatment strategies to alleviate selective behavioral components of addiction.

ACKNOWLEDGMENTS AND DISCLOSURES

The work is supported by the Centre National de la Recherche Scientifique (to MS, PV, and JC), Institut National de la Santé et de la Recherche Médicale (to PV and JC), Fondation pour la Recherche Médicale (Grant No. DEQ20150734352 [to JC]), and the Bio-Psy Labex cluster of excellence (to MS, JC, and PV); Sorbonne Université—Paris VI (to JC and PV), Agence Nationale pour la Recherche (Grant Nos. ANR-15-CE16-001 [to PT and PV] and ANR-18-CE37-0003-02 [to PV]); and the Institut National de la Recherche Agronomique, University of Bordeaux (to PT), Idex Bordeaux

"chaire d'installation" (Grant No. ANR-10-IDEX-03-02) [to PT], and Région Aquitaine (Grant No. 2014-1R30301-00003023 [to PT]).

JC and PV disclose consulting fees from MELKin Pharmaceuticals but reported no potential conflicts of interest. The other authors report no biomedical financial interests or potential conflicts of interest.

ARTICLE INFORMATION

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Received Jun 7, 2019; revised Oct 30, 2019; accepted Nov 1, 2019.

Supplementary material cited in this article is available online at https://doi.org/10.1016/j.biopsych.2019.11.001.

REFERENCES

1. Lüscher C, Malenka RC (2011): Drug-evoked synaptic plasticity in addiction: From molecular changes to circuit remodeling. Neuron 69:650–663.
2. Nestler EJ (2001): Molecular basis of long-term plasticity underlying addiction. Nat Rev Neurosci 2:119–128.
3. Milton AL, Everitt BJ (2012): The persistence of maladaptive memory: Addiction, drug memories and anti-relapse treatments. Neurosci Biobehav Rev 36:1119–1139.
4. Nestler EJ, Lüscher C (2019): The molecular basis of drug addiction: Linking epigenetic to synaptic and circuit mechanisms. Neuron 102:48–59.
5. Di Chiara G, Imperato A (1988): Drugs abused by humans preferentially increase synaptic dopamine concentrations in the mesolimbic system of freely moving rats. Proc Natl Acad Sci U S A 85:5274–5278.
6. Moss J, Bolam JP (2008): A dopaminergic axon lattice in the striatum and its relationship with cortical and thalamic terminals. J Neurosci 28:11221–11230.
7. Doig NM, Moss J, Bolam JP (2010): Cortical and thalamic innervation of direct and indirect pathway medium-sized spiny neurons in mouse striatum. J Neurosci 30:14610–14618.
8. Gerfen C, Engber T, Mahan L, Susel Z, Chase T, Monsma F, Sibley D (1990): D1 and D2 dopamine receptor-regulated gene expression of striatonigral and striatopallidal neurons. Science 250:1429–1432.
9. Moine CL, Bloch B (1995): D1 and D2 dopamine receptor gene expression in the rat striatum: Sensitive cRNA probes demonstrate prominent segregation of D1 and D2 mRNAS in distinct neuronal populations of the dorsal and ventral striatum. J Comp Neurol 355:418–426.
10. Felder CC, Williams HL, Axelrod J (1991): A transduction pathway associated with receptors coupled to the inhibitory guanine nucleotide binding protein Gi that amplifies ATP-mediated arachidonic acid release. Proc Natl Acad Sci U S A 88:6477–6480.
11. Corvol JC, Studler JM, Schonn JS, Girault JA, Hervé D (2001): Gαolf is necessary for coupling D1 and A2a receptors to adenylyl cyclase in the striatum. J Neurochem 76:1585–1588.
12. Self DW (2010): Dopamine receptor subtypes in reward and relapse. In: Neve KA, editor. The Dopamine Receptors. Totowa, NJ: Humana Press, 479–524.
13. Gong S, Zheng C, Doughty ML, Losos K, Didkovsky N, Schambra UB, *et al.* (2003): A gene expression atlas of the central nervous system based on bacterial artificial chromosomes. Nature 425:917–925.
14. Gong S, Doughty M, Harbaugh CR, Cummins A, Hatten ME, Heintz N, Gerfen CR (2007): Targeting Cre recombinase to specific neuron populations with bacterial artificial chromosome constructs. J Neurosci 27:9817–9823.
15. Lemberger T, Parlato R, Dassesse D, Westphal M, Casanova E, Turiault M, *et al.* (2007): Expression of the Cre recombinase in dopaminoceptive neurons. BMC Neurosci 8:4.
16. Shuen JA, Chen M, Gloss B, Calakos N (2008): Drd1a-tdTomato BAC transgenic mice for simultaneous visualization of medium spiny neurons in the direct and indirect pathways of the basal ganglia. J Neurosci 28:2681–2685.
17. Durieux PF, Bearzatto B, Guiducci S, Buch T, Waisman A, Zoli M, *et al.* (2009): D2R striatopallidal neurons inhibit both locomotor and drug reward processes. Nat Neurosci 12:393–395.
18. Belin D, Jonkman S, Dickinson A, Robbins TW, Everitt BJ (2009): Parallel and interactive learning processes within the basal ganglia: Relevance for the understanding of addiction. Behav Brain Res 199:89–102.
19. Everitt BJ, Robbins TW (2016): Drug addiction: Updating actions to habits to compulsions ten years on. Annu Rev Psychol 67:23–50.
20. Kasanetz F, Deroche-Gamonet V, Berson N, Balado E, Lafourcade M, Manzoni O, Piazza PV (2010): Transition to addiction is associated with a persistent impairment in synaptic plasticity. Science 328:1709–1712.
21. Lim SAO, Kang UJ, McGehee DS (2014): Striatal cholinergic interneuron regulation and circuit effects. Front Synaptic Neurosci 6:22.
22. Kawaguchi Y, Wilson C, Emson P (1990): Projection subtypes of rat neostriatal matrix cells revealed by intracellular injection of biocytin. J Neurosci 10:3421–3438.
23. Cazorla M, de Carvalho FD, Chohan MO, Shegda M, Chuhma N, Rayport S, *et al.* (2014): Dopamine D2 receptors regulate the anatomical and functional balance of basal ganglia circuitry. Neuron 81:153–164.
24. Bertran-Gonzalez J, Bosch C, Maroteaux M, Matamales M, Herve D, Valjent E, Girault J-A (2008): Opposing patterns of signaling activation in dopamine D1 and D2 receptor-expressing striatal neurons in response to cocaine and haloperidol. J Neurosci 28:5671–5685.
25. Kupchik YM, Brown RM, Heinsbroek JA, Lobo MK, Schwartz DJ, Kalivas PW (2015): Coding the direct/indirect pathways by D1 and D2 receptors is not valid for accumbens projections. Nat Neurosci 18:1230–1232.
26. Pardo-Garcia TR, Garcia-Keller C, Penaloza T, Richie CT, Pickel J, Hope BT, *et al.* (2019): Ventral pallidum is the primary target for accumbens D1 projections driving cocaine seeking. J Neurosci 39:2041–2051.
27. Baimel C, McGarry LM, Carter AG (2019): The Projection Targets of Medium Spiny Neurons Govern Cocaine-Evoked Synaptic Plasticity in the Nucleus Accumbens. Cell Rep 28:2256–2263.e3.
28. Kravitz AV, Freeze BS, Parker PRL, Kay K, Thwin MT, Deisseroth K, Kreitzer AC (2010): Regulation of Parkinsonian motor behaviours by optogenetic control of basal ganglia circuitry. Nature 466:622–626.
29. Albin RL, Young AB, Penney JB (1989): The functional anatomy of basal ganglia disorders. Trends Neurosci 12:366–375.
30. DeLong MR (1990): Primate models of movement disorders of basal ganglia origin. Trends Neurosci 13:281–285.
31. Lobo MK, Covington HE, Chaudhury D, Friedman AK, Sun H, Damez-Werno D, *et al.* (2010): Cell type-specific loss of BDNF signaling mimics optogenetic control of cocaine reward. Science 330:385–390.
32. Kravitz AV, Tye LD, Kreitzer AC (2012): Distinct roles for direct and indirect pathway striatal neurons in reinforcement. Nat Neurosci 15:816–818.
33. Ferguson SM, Eskenazi D, Ishikawa M, Wanat MJ, Phillips PEM, Dong Y, *et al.* (2011): Transient neuronal inhibition reveals opposing roles of indirect and direct pathways in sensitization. Nat Neurosci 14:22–24.
34. Farrell MS, Pei Y, Wan Y, Yadav PN, Daigle TL, Urban DJ, *et al.* (2013): A Gαs DREADD mouse for selective modulation of cAMP production in striatopallidal neurons. Neuropsychopharmacology 38:854–862.

35. Song SS, Kang BJ, Wen L, Lee HJ, Sim H, Kim TH, *et al.* (2014): Optogenetics reveals a role for accumbal medium spiny neurons expressing dopamine D2 receptors in cocaine-induced behavioral sensitization. Front Behav Neurosci 8:336.
36. Calipari ES, Bagot RC, Purushothaman I, Davidson TJ, Yorgason JT, Peña CJ, *et al.* (2016): In vivo imaging identifies temporal signature of D1 and D2 medium spiny neurons in cocaine reward. Proc Natl Acad Sci U S A 113:2726–2731.
37. Chandra R, Lenz JD, Gancarz AM, Chaudhury D, Schroeder GL, Han M-H, *et al.* (2013): Optogenetic inhibition of D1R containing nucleus accumbens neurons alters cocaine-mediated regulation of Tiam1. Front Mol Neurosci 6:13.
38. Hikida T, Kimura K, Wada N, Funabiki K, Nakanishi S (2010): Distinct roles of synaptic transmission in direct and indirect striatal pathways to reward and aversive behavior. Neuron 66:896–907.
39. Bock R, Shin JH, Kaplan AR, Dobi A, Markey E, Kramer PF, *et al.* (2013): Strengthening the accumbal indirect pathway promotes resilience to compulsive cocaine use. Nat Neurosci 16:632–638.
40. Yager LM, Garcia AF, Donckels EA, Ferguson SM (2019): Chemogenetic inhibition of direct pathway striatal neurons normalizes pathological, cue-induced reinstatement of drug-seeking in rats. Addict Biol 24:251–264.
41. O'Connor EC, Kremer Y, Lefort S, Harada M, Pascoli V, Rohner C, Lüscher C (2015): Accumbal D1R neurons projecting to lateral hypothalamus authorize feeding. Neuron 88:553–564.
42. Larson EB, Wissman AM, Loriaux AL, Kourrich S, Self DW (2015): Optogenetic stimulation of accumbens shell or shell projections to lateral hypothalamus produce differential effects on the motivation for cocaine. J Neurosci 35:3537–3543.
43. Dobbs LK, Kaplan AR, Lemos JC, Matsui A, Rubinstein M, Alvarez VA (2016): Dopamine regulation of lateral inhibition between striatal neurons gates the stimulant actions of cocaine. Neuron 90:1100–1113.
44. Burke DA, Rotstein HG, Alvarez VA (2017): Striatal local circuitry: A new framework for lateral inhibition. Neuron 96:267–284.
45. Matsui A, Alvarez VA (2018): Cocaine inhibition of synaptic transmission in the ventral pallidum is pathway-specific and mediated by serotonin. Cell Rep 23:3852–3863.
46. Durieux PF, Schiffmann SN, de Kerchove d'Exaerde A (2012): Differential regulation of motor control and response to dopaminergic drugs by D1R and D2R neurons in distinct dorsal striatum subregions: Dorsal striatum D1R- and D2R-neuron motor functions. EMBO J 31:640–653.
47. Soares-Cunha C, Coimbra B, Domingues AV, Vasconcelos N, Sousa N, Rodrigues AJ (2018): Nucleus accumbens microcircuit underlying D2-MSN-driven increase in motivation. eNeuro 5(2).
48. Cole SL, Robinson MJF, Berridge KC (2018): Optogenetic self-stimulation in the nucleus accumbens: D1 reward versus D2 ambivalence. PLoS One 13:e0207694.
49. Vicente AM, Galvão-Ferreira P, Tecuapetla F, Costa RM (2016): Direct and indirect dorsolateral striatum pathways reinforce different action strategies. Curr Biol 26:R267–R269.
50. Soares-Cunha C, Coimbra B, David-Pereira A, Borges S, Pinto L, Costa P, *et al.* (2016): Activation of D2 dopamine receptor-expressing neurons in the nucleus accumbens increases motivation. Nat Commun 7:11829.
51. Natsubori A, Tsutsui-Kimura I, Nishida H, Bouchekioua Y, Sekiya H, Uchigashima M, *et al.* (2017): Ventrolateral striatal medium spiny neurons positively regulate food-incentive, goal-directed behavior independently of D1 and D2 selectivity. J Neurosci 37:2723–2733.
52. Tsutsui-Kimura I, Takiue H, Yoshida K, Xu M, Yano R, Ohta H, *et al.* (2017): Dysfunction of ventrolateral striatal dopamine receptor type 2-expressing medium spiny neurons impairs instrumental motivation. Nat Commun 8:14304.
53. Soares-Cunha C, de Vasconcelos NAP, Coimbra B, Domingues AV, Silva JM, Loureiro-Campos E, *et al.* (2019): Nucleus accumbens medium spiny neurons subtypes signal both reward and aversion [published online ahead of print Aug 28; published correction Sep 18.]. Mol Psychiatry.
54. Carvalho Poyraz F, Holzner E, Bailey MR, Meszaros J, Kenney L, Kheirbek MA, *et al.* (2016): Decreasing striatopallidal pathway function enhances motivation by energizing the initiation of goal-directed action. J Neurosci 36:5988–6001.
55. Gallo EF, Meszaros J, Sherman JD, Chohan MO, Teboul E, Choi CS, *et al.* (2018): Accumbens dopamine D2 receptors increase motivation by decreasing inhibitory transmission to the ventral pallidum. Nat Commun 9:1086.
56. Cui G, Jun SB, Jin X, Pham MD, Vogel SS, Lovinger DM, Costa RM (2013): Concurrent activation of striatal direct and indirect pathways during action initiation. Nature 494:238–242.
57. Tecuapetla F, Jin X, Lima SQ, Costa RM (2016): Complementary contributions of striatal projection pathways to action initiation and execution. Cell 166:703–715.
58. Creed M, Ntamati NR, Chandra R, Lobo MK, Lüscher C (2016): Convergence of reinforcing and anhedonic cocaine effects in the ventral pallidum. Neuron 92:214–226.
59. Heinsbroek JA, Neuhofer DN, Griffin WC, Siegel GS, Bobadilla A-C, Kupchik YM, Kalivas PW (2017): Loss of plasticity in the D2-accumbens pallidal pathway promotes cocaine seeking. J Neurosci 37:757–767.
60. Barrientos C, Knowland D, Wu MMJ, Lilascharoen V, Huang KW, Malenka RC, Lim BK (2018): Cocaine-induced structural plasticity in input regions to distinct cell types in nucleus accumbens. Biol Psychiatry 84:893–904.
61. Pascoli V, Turiault M, Lüscher C (2012): Reversal of cocaine-evoked synaptic potentiation resets drug-induced adaptive behaviour. Nature 481:71–75.
62. Pascoli V, Terrier J, Espallergues J, Valjent E, O'Connor EC, Lüscher C (2014): Contrasting forms of cocaine-evoked plasticity control components of relapse. Nature 509:459–464.
63. Pascoli V, Hiver A, Van Zessen R, Loureiro M, Achargui R, Harada M, *et al.* (2018): Stochastic synaptic plasticity underlying compulsion in a model of addiction. Nature 564:366–371.
64. MacAskill AF, Cassel JM, Carter AG (2014): Cocaine exposure reorganizes cell type– and input-specific connectivity in the nucleus accumbens. Nat Neurosci 17:1198–1207.
65. Sjulson L, Peyrache A, Cumpelik A, Cassataro D, Buzsáki G (2018): Cocaine place conditioning strengthens location-specific hippocampal coupling to the nucleus accumbens. Neuron 98:926–934.e5.
66. Creed M, Pascoli VJ, Luscher C (2015): Refining deep brain stimulation to emulate optogenetic treatment of synaptic pathology. Science 347:659–664.
67. Diana M, Raij T, Melis M, Nummenmaa A, Leggio L, Bonci A (2017): Rehabilitating the addicted brain with transcranial magnetic stimulation. Nat Rev Neurosci 18:685–693.
68. Südhof TC (2017): Molecular neuroscience in the 21st century: A personal perspective. Neuron 96:536–541.
69. Gerfen CR, Surmeier DJ (2011): Modulation of striatal projection systems by dopamine. Annu Rev Neurosci 34:441–466.
70. Gardoni F, Bellone C (2015): Modulation of the glutamatergic transmission by dopamine: A focus on Parkinson, Huntington and Addiction diseases. Front Cell Neurosci 9:25.
71. van Huijstee AN, Mansvelder HD (2015): Glutamatergic synaptic plasticity in the mesocorticolimbic system in addiction. Front Cell Neurosci 8:466.
72. Luo Z, Volkow ND, Heintz N, Pan Y, Du C (2011): Acute cocaine induces fast activation of D1 receptor and progressive deactivation of D2 receptor striatal neurons: In vivo optical microprobe [Ca2+]i imaging. J Neurosci 31:13180–13190.
73. Barbera G, Liang B, Zhang L, Gerfen CR, Culurciello E, Chen R, *et al.* (2016): Spatially compact neural clusters in the dorsal striatum encode locomotion relevant information. Neuron 92:202–213.
74. Heusner CL (2005): Expression of mutant NMDA receptors in dopamine D1 receptor-containing cells prevents cocaine sensitization and decreases cocaine preference. J Neurosci 25:6651–6657.
75. Lambot L, Chaves Rodriguez E, Houtteman D, Li Y, Schiffmann SN, Gall D, de Kerchove d'Exaerde A (2016): Striatopallidal neuron NMDA receptors control synaptic connectivity, locomotor, and goal-directed behaviors. J Neurosci 36:4976–4992.
76. Beutler LR, Wanat MJ, Quintana A, Sanz E, Bamford NS, Zweifel LS, Palmiter RD (2011): Balanced NMDA receptor activity in dopamine

D1 receptor (D1R)- and D2R-expressing medium spiny neurons is required for amphetamine sensitization. Proc Natl Acad Sci U S A 108:4206–4211.
77. Joffe ME, Vitter SR, Grueter BA (2017): GluN1 deletions in D1- and A2A-expressing cell types reveal distinct modes of behavioral regulation. Neuropharmacology 112:172–180.
78. Valjent E, Pages C, Herve D, Girault J-A, Caboche J (2004): Addictive and non-addictive drugs induce distinct and specific patterns of ERK activation in mouse brain. Eur J Neurosci 19:1826–1836.
79. Valjent E, Corvol J-C, Pagès C, Besson M-J, Maldonado R, Caboche J (2000): Involvement of the extracellular signal-regulated kinase cascade for cocaine-rewarding properties. J Neurosci 20:8701–8709.
80. Valjent E, Corvol J-C, Trzaskos JM, Girault A, Hervé D (2006): Role of the ERK pathway in psychostimulant-induced locomotor sensitization. BMC Neurosci 7:20.
81. Miller CA, Marshall JF (2005): Molecular substrates for retrieval and reconsolidation of cocaine-associated contextual memory. Neuron 47:873–884.
82. Valjent E, Corbille A-G, Bertran-Gonzalez J, Herve D, Girault J-A (2006): Inhibition of ERK pathway or protein synthesis during reexposure to drugs of abuse erases previously learned place preference. Proc Natl Acad Sci U S A 103:2932–2937.
83. Pascoli V, Besnard A, Hervé D, Pagès C, Heck N, Girault J-A, *et al.* (2011): Cyclic adenosine monophosphate–independent tyrosine phosphorylation of NR2B mediates cocaine-induced extracellular signal-regulated kinase activation. Biol Psychiatry 69:218–227.
84. Girault J, Valjent E, Caboche J, Herve D (2007): ERK2: A logical AND gate critical for drug-induced plasticity? Curr Opin Pharmacol 7:77–85.
85. Pascoli V, Cahill E, Bellivier F, Caboche J, Vanhoutte P (2014): Extracellular signal-regulated protein kinases 1 and 2 activation by addictive drugs: A signal toward pathological adaptation. Biol Psychiatry 76:917–926.
86. Cahill E, Salery M, Vanhoutte P, Caboche J (2014): Convergence of dopamine and glutamate signaling onto striatal ERK activation in response to drugs of abuse. Front Pharmacol 4:172.
87. Dos Santos M, Salery M, Forget B, Garcia Perez MA, Betuing S, Boudier T, *et al.* (2017): Rapid synaptogenesis in the nucleus accumbens is induced by a single cocaine administration and stabilized by mitogen-activated protein kinase interacting kinase-1 activity. Biol Psychiatry 82:806–818.
88. Flores-Hernández J, Cepeda C, Hernández-Echeagaray E, Calvert CR, Jokel ES, Fienberg AA, *et al.* (2002): Dopamine enhancement of NMDA currents in dissociated medium-sized striatal neurons: Role of D1 receptors and DARPP-32. J Neurophysiol 88:3010–3020.
89. Valjent E, Pascoli V, Svenningsson P, Paul S, Enslen H, Corvol J-C, *et al.* (2005): Regulation of a protein phosphatase cascade allows convergent dopamine and glutamate signals to activate ERK in the striatum. Proc Natl Acad Sci U S A 102:491–496.
90. Bateup HS, Svenningsson P, Kuroiwa M, Gong S, Nishi A, Heintz N, Greengard P (2008): Cell type–specific regulation of DARPP-32 phosphorylation by psychostimulant and antipsychotic drugs. Nat Neurosci 11:932–939.
91. Bateup HS, Santini E, Shen W, Birnbaum S, Valjent E, Surmeier DJ, *et al.* (2010): Distinct subclasses of medium spiny neurons differentially regulate striatal motor behaviors. Proc Natl Acad Sci U S A 107:14845–14850.
92. Pulido R (1998): PTP-SL and STEP protein tyrosine phosphatases regulate the activation of the extracellular signal-regulated kinases ERK1 and ERK2 by association through a kinase interaction motif. EMBO J 17:7337–7350.
93. Jiang SZ, Xu W, Emery AC, Gerfen CR, Eiden MV, Eiden LE (2017): NCS-Rapgef2, the protein product of the neuronal Rapgef2 gene, is a specific activator of D1 dopamine receptor-dependent ERK phosphorylation in mouse brain. eNeuro 4(5).
94. Beaulieu JM (2016): In vivo veritas, the next frontier for functionally selective GPCR ligands. Methods 92:64–71.
95. Donthamsetti P, Gallo EF, Buck DC, Stahl EL, Zhu Y, Lane JR, *et al.* (2018): Arrestin recruitment to dopamine D2 receptor mediates locomotion but not incentive motivation [published online ahead of print Aug 17]. Mol Psychiatry.
96. Wang M, Wong AH, Liu F (2012): Interactions between NMDA and dopamine receptors: A potential therapeutic target. Brain Res 1476:154–163.
97. Borroto-Escuela DO, Fuxe K (2017): Diversity and bias through dopamine D2R heteroreceptor complexes. Curr Opin Pharmacol 32:16–22.
98. Andrianarivelo A, Saint-Jour E, Walle R, Trifilieff P, Vanhoutte P (2018): Modulation and functions of dopamine receptor heteromers in drugs of abuse-induced adaptations. Neuropharmacology 152:42–50.
99. Trifilieff P, Rives M-L, Urizar E, Piskorowski R, Vishwasrao H, Castrillon J, *et al.* (2011): Detection of antigen interactions ex vivo by proximity ligation assay: Endogenous dopamine D2-adenosine A2A receptor complexes in the striatum. Biotechniques 51:111–118.
100. Ferré S, Bonaventura J, Zhu W, Hatcher-Solis C, Taura J, Quiroz C, *et al.* (2018): Essential control of the function of the striatopallidal neuron by pre-coupled complexes of adenosine A2A-dopamine D2 receptor heterotetramers and adenylyl cyclase. Front Pharmacol 9:243.
101. Lee FJS, Xue S, Pei L, Vukusic B, Chéry N, Wang Y, *et al.* (2002): Dual regulation of NMDA receptor functions by direct protein-protein interactions with the dopamine D1 receptor. Cell 111:219–230.
102. Pei L (2004): Regulation of dopamine D1 receptor function by physical interaction with the NMDA receptors. J Neurosci 24:1149–1158.
103. Cepeda C, Levine MS (2006): Where do you think you are going? The NMDA-D1 receptor trap. Sci STKE 2006:pe20.
104. Ladepeche L, Dupuis JP, Bouchet D, Doudnikoff E, Yang L, Campagne Y, *et al.* (2013): Single-molecule imaging of the functional crosstalk between surface NMDA and dopamine D1 receptors. Proc Natl Acad Sci U S A 110:18005–18010.
105. Cahill E, Pascoli V, Trifilieff P, Savoldi D, Kappès V, Lüscher C, *et al.* (2014): D1R/GluN1 complexes in the striatum integrate dopamine and glutamate signalling to control synaptic plasticity and cocaine-induced responses. Mol Psychiatry 19:1295–1304.
106. Liu X-Y, Chu X-P, Mao L-M, Wang M, Lan H-X, Li M-H, *et al.* (2006): Modulation of D2R-NR2B interactions in response to cocaine. Neuron 52:897–909.
107. Robison AJ, Nestler EJ (2011): Transcriptional and epigenetic mechanisms of addiction. Nat Rev Neurosci 12:623–637.
108. Dong Y, Nestler EJ (2014): The neural rejuvenation hypothesis of cocaine addiction. Trends Pharmacol Sci 35:374–383.
109. Brami-Cherrier K, Roze E, Girault J-A, Betuing S, Caboche J (2009): Role of the ERK/MSK1 signalling pathway in chromatin remodelling and brain responses to drugs of abuse. J Neurochem 108:1323–1335.
110. Besnard A, Bouveyron N, Kappes V, Pascoli V, Pages C, Heck N, *et al.* (2011): Alterations of molecular and behavioral responses to cocaine by selective inhibition of Elk-1 phosphorylation. J Neurosci 31:14296–14307.
111. Brami-Cherrier K (2005): Parsing molecular and behavioral effects of cocaine in mitogen- and stress-activated protein kinase-1-deficient mice. J Neurosci 25:11444–11454.
112. Chandra R, Lobo MK (2017): Beyond neuronal activity markers: Select immediate early genes in striatal neuron subtypes functionally mediate psychostimulant addiction. Front Behav Neurosci 11:112.
113. Guez-Barber D, Fanous S, Golden SA, Schrama R, Koya E, Stern AL, *et al.* (2011): FACS identifies unique cocaine-induced gene regulation in selectively activated adult striatal neurons. J Neurosci 31:4251–4259.
114. Zhang J, Zhang L, Jiao H, Zhang Q, Zhang D, Lou D, *et al.* (2006): c-Fos facilitates the acquisition and extinction of cocaine-induced persistent changes. J Neurosci 26:13287–13296.
115. Lee K-W, Kim Y, Kim AM, Helmin K, Nairn AC, Greengard P (2006): Cocaine-induced dendritic spine formation in D1 and D2 dopamine receptor-containing medium spiny neurons in nucleus accumbens. Proc Natl Acad Sci 103:3399–3404.

116. Lobo MK, Zaman S, Damez-Werno DM, Koo JW, Bagot RC, DiNieri JA, *et al.* (2013): FosB induction in striatal medium spiny neuron subtypes in response to chronic pharmacological, emotional, and optogenetic stimuli. J Neurosci 33:18381–18395.
117. Heiman M, Schaefer A, Gong S, Peterson JD, Day M, Ramsey KE, *et al.* (2008): A translational profiling approach for the molecular characterization of CNS cell types. Cell 135:738–748.
118. Grueter BA, Robison AJ, Neve RL, Nestler EJ, Malenka RC (2013): FosB differentially modulates nucleus accumbens direct and indirect pathway function. Proc Natl Acad Sci 110:1923–1928.
119. Kelz MB, Chen J, Carlezon WA Jr, Whisler K, Gilden L, Beckmann AM, *et al.* (1999): Expression of the transcription factor DFosB in the brain controls sensitivity to cocaine. Nature 401:272–276.
120. Colby CR, Whisler K, Steffen C, Nestler EJ, Self DW (2003): Striatal cell type-specific overexpression of deltaFosB enhances incentive for cocaine. J Neurosci 23:2488–2493.
121. Fosnaugh JS, Bhat RV, Yamagata K, Worley PF, Baraban JM (1995): Activation of arc, a putative "effector" immediate early gene, by cocaine in rat brain. J Neurochem 64:2377–2380.
122. Fumagalli F, Bedogni F, Frasca A, Di Pasquale L, Racagni G, Riva MA (2006): Corticostriatal up-regulation of activity-regulated cytoskeletal-associated protein expression after repeated exposure to cocaine. Mol Pharmacol 70:1726–1734.
123. Salery M, Dos Santos M, Saint-Jour E, Moumné L, Pagès C, Kappès V, *et al.* (2017): Activity-regulated cytoskeleton-associated protein accumulates in the nucleus in response to cocaine and acts as a brake on chromatin remodeling and long-term behavioral alterations. Biol Psychiatry 81:573–584.
124. Korb E, Finkbeiner S (2011): Arc in synaptic plasticity: From gene to behavior. Trends Neurosci 34:591–598.
125. Bramham CR, Alme MN, Bittins M, Kuipers SD, Nair RR, Pai B, *et al.* (2010): The Arc of synaptic memory. Exp Brain Res 200:125–140.
126. Arango-Lievano M, Schwarz JT, Vernov M, Wilkinson MB, Bradbury K, Feliz A, *et al.* (2014): Cell-type specific expression of p11 controls cocaine reward. Biol Psychiatry 76:794–801.
127. Chandra R, Engeln M, Schiefer C, Patton MH, Martin JA, Werner CT, *et al.* (2017): Drp1 mitochondrial fission in D1 neurons mediates behavioral and cellular plasticity during early cocaine abstinence. Neuron 96:1327–1341.e6.
128. Parekh PK, Logan RW, Ketchesin KD, Becker-Krail D, Shelton MA, Hildebrand MA, *et al.* (2019): Cell-type-specific regulation of nucleus accumbens synaptic plasticity and cocaine reward sensitivity by the circadian protein, NPAS2. J Neurosci 39:4657–4667.
129. Chandra R, Engeln M, Francis TC, Konkalmatt P, Patel D, Lobo MK (2017): A role for peroxisome proliferator-activated receptor gamma coactivator-1α in nucleus accumbens neuron subtypes in cocaine action. Biol Psychiatry 81:564–572.
130. Lobo MK, Karsten SL, Gray M, Geschwind DH, Yang XW (2006): FACS-array profiling of striatal projection neuron subtypes in juvenile and adult mouse brains. Nat Neurosci 9:443–452.
131. Chandra R, Francis TC, Konkalmatt P, Amgalan A, Gancarz AM, Dietz DM, Lobo MK (2015): Opposing role for Egr3 in nucleus accumbens cell subtypes in cocaine action. J Neurosci 35:7927–7937.
132. Nestler EJ (2014): Epigenetic mechanisms of drug addiction. Neuropharmacology 76:259–268.
133. Bannister AJ, Kouzarides T (2011): Regulation of chromatin by histone modifications. Cell Res 21:381–395.
134. Walker DM, Cates HM, Heller EA, Nestler EJ (2015): Regulation of chromatin states by drugs of abuse. Curr Opin Neurobiol 30:112–121.
135. Kumar A, Choi K-H, Renthal W, Tsankova NM, Theobald DEH, Truong H-T, *et al.* (2005): Chromatin remodeling is a key mechanism underlying cocaine-induced plasticity in striatum. Neuron 48:303–314.
136. Shen H-Y, Kalda A, Yu L, Ferrara J, Zhu J, Chen J-F (2008): Additive effects of histone deacetylase inhibitors and amphetamine on histone H4 acetylation, cAMP responsive element binding protein phosphorylation and ΔFosB expression in the striatum and locomotor sensitization in mice. Neuroscience 157:644–655.
137. Renthal W, Kumar A, Xiao G, Wilkinson M, Covington HE, Maze I, *et al.* (2009): Genome-wide analysis of chromatin regulation by cocaine reveals a role for sirtuins. Neuron 62:335–348.
138. Rogge GA, Wood MA (2013): The role of histone acetylation in cocaine-induced neural plasticity and behavior. Neuropsychopharmacology 38:94–110.
139. Jordi E, Heiman M, Marion-Poll L, Guermonprez P, Cheng SK, Nairn AC, *et al.* (2013): Differential effects of cocaine on histone posttranslational modifications in identified populations of striatal neurons. Proc Natl Acad Sci 110:9511–9516.
140. Mahadevan LC, Clayton AL, Hazzalin CA, Thomson S (2008): Phosphorylation and acetylation of histone H3 at inducible genes: Two controversies revisited. In: Bock G, Goode J, editors. Novartis Foundation Symposia. Chichester, UK: John Wiley & Sons, Ltd., 102–114
141. Barski A, Cuddapah S, Cui K, Roh T-Y, Schones DE, Wang Z, *et al.* (2007): High-resolution profiling of histone methylations in the human genome. Cell 129:823–837.
142. Maze I, Covington HE, Dietz DM, LaPlant Q, Renthal W, Russo SJ, *et al.* (2010): Essential role of the histone methyltransferase G9a in cocaine-induced plasticity. Science 327:213–216.
143. Maze I, Chaudhury D, Dietz DM, Von Schimmelmann M, Kennedy PJ, Lobo MK, *et al.* (2014): G9a influences neuronal subtype specification in striatum. Nat Neurosci 17:533–539.
144. Damez-Werno DM, Sun H, Scobie KN, Shao N, Rabkin J, Dias C, *et al.* (2016): Histone arginine methylation in cocaine action in the nucleus accumbens. Proc Natl Acad Sci 113:9623–9628.
145. Mews P, Walker DM, Nestler EJ (2019): Epigenetic priming in drug addiction. Cold Spring Harb Symp Quant Biol 83:131–139.
146. Im H-I, Kenny PJ (2012): MicroRNAs in neuronal function and dysfunction. Trends Neurosci 35:325–334.
147. Smith ACW, Kenny PJ (2018): MicroRNAs regulate synaptic plasticity underlying drug addiction. Genes Brain Behav 17:e12424.
148. Bartel DP (2009): MicroRNAs: Target recognition and regulatory functions. Cell 136:215–233.
149. Schaefer A, Im H-I, Venø MT, Fowler CD, Min A, Intrator A, *et al.* (2010): Argonaute 2 in dopamine 2 receptor–expressing neurons regulates cocaine addiction. J Exp Med 207:1843–1851.