Biological Psychiatry: Celebrating 50 Years

A Warning for Smoking Parents

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Drug addiction is a psychiatric disorder that affects millions of people worldwide and poses a challenge for current neuroscience research. Nicotine, commonly consumed by smoking tobacco, is considered one of the most commonly used addictive drugs. The mesolimbic dopamine (DA) system is a key brain circuit targeted by nicotine to induce behavioral changes. Upon acute exposure, nicotine binds and stimulates nicotinic acetylcholine receptors (nAChRs), resulting in an overall enhancement of DA signaling from the ventral tegmental area (VTA) to the nucleus accumbens (NAc) (1). VTA to NAc DA signaling stimulates appetitive and goal-directed behaviors and detects events that are key to motivation and reinforcement, such as an unpredicted reward or an anticipated reward (2). After chronic use, complicated neuronal alterations occur, one of which is the upregulation of voltage-gated calcium channels in VTA DA neurons. This upregulation not only increases intracellular calcium dynamics but also alters gene expression to steer undifferentiated VTA neurons to acquire a DAergic phenotype during development, a form of neuronal plasticity termed neurotransmitter switching (3).

Nicotine exposure during development induces behavioral and neurological changes in offspring, making it a well-known neuroteratogen that compromises the formation of critical neural pathways (4). Prenatal nicotine exposure has also been reported to induce various brain conditions, such as attention-deficit/hyperactivity disorder, learning disabilities, and an increased risk of future nicotine abuse. In animal models, offspring exposed to maternal nicotine through gestation and lactation exhibited metabolic, reproductive, pulmonary, and hypoxia-sensing malfunctions (5). These negative consequences prompt a thorough understanding of underlying neural and circuit mechanisms and the consequent effects of drug taking in exposed offspring.

In the current issue of *Biological Psychiatry*, Romoli *et al.* (6) tested the hypothesis that neonatal nicotine exposure enhances drug preference in adults by neural plastic changes involving non-DAergic neurons' acquiring a DAergic phenotype. This form of neural plasticity involving changes in neurotransmitter identities has previously been demonstrated during development and in the mature nervous system (7).

In Romoli *et al.*'s experiments (6), mouse pups were exposed to nicotine neonatally (NN) daily beginning at postnatal day 2 (P2) for 15 days through lactation. At P90, a two-bottle-choice test was performed between nicotine and water, revealing that the NN-exposed mice significantly preferred nicotine. It appears that NN presets these pups for nicotine preference. Romoli *et al.* (6) then moved on to test whether NN also presets the preference for other drugs of abuse. With another cohort of NN-exposed mice, Romoli *et al.* (6) tested the NN mice for a choice between ethanol or water and observed an increased consumption of ethanol over time despite the fact that the mice had never been exposed to ethanol. As an important control, these NN adult mice exhibit normal motivation to obtain natural rewards. These results not only demonstrate that NN enhances nicotine preference in later adult life but also raise a possibility that NN may also set up preference for drugs of abuse in general.

After establishing such important behavioral consequences of NN, Romoli *et al.* (6) moved on to explore the underlying mechanisms. VTA-originated DA signaling regulates the formation, consolidation, and reconsolidation of memories, including reward- and addiction-related memories (8). Given the cellular effects of nicotine on DA signaling and the possibility that some non-DAergic VTA neurons can acquire a DAergic phenotype upon altered DA signaling (5,7,9), Romoli *et al.* (6) tested the hypothesis that NN increases the number of DAergic neurons in the VTA, an alteration that induces neurotransmitter switching and redefines DA signaling. In support of this hypothesis, Romoli *et al.* (6) detected an increase in the expression of tyrosine hydroxylase, the rate-limiting enzyme in the production of DA, in the VTA of NN mice at P120. Furthermore, this increase was absent before adult nicotine re-exposure, suggesting that the neurotransmitter switching was initially primed by NN but only expressed upon re-exposure in adulthood. To determine the neuronal types that switched their neurotransmitter phenotypes, Romoli *et al.* (6) used transgenic mouse lines labeling the two major non-DAergic neurons in the VTA and observed that glutamatergic neurons, but not gamma-aminobutyric acidergic (GABAergic) neurons, were the players. Thus, NN primes a population of glutamatergic neurons in the VTA to convert to DAergic neurons upon a subsequent exposure to nicotine in the adulthood.

What mechanisms mediate the switch from one neuronal phenotype to another? To address this question, Romoli *et al.* (6) focused on Nuur1, which is sensitively regulated by intracellular calcium activities, as well as nAChRs with specific subunits (3). In Nurr1-expressing neurons, Romoli *et al.* (6) detected increased durations of calcium spikes, increased responsiveness and prolonged activation upon NN, and upregulation of the nAChR subunits. Furthermore, while NN affected calcium activity in both glutamatergic and GABAergic neurons, GABAergic neurons showed higher spontaneous calcium-spiking frequency. On the other hand, bath application of nicotine to brain slices increased the population activity of glutamatergic neurons and caused the opposite effect in GABAergic neurons. These results implicate calcium activities and nAChR as potential triggers for Nurr1 expression and neurotransmitter respecification after NN.

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How are the expression of Nurr1 and increased calcium signals or neuronal activities coordinated in neuronal phenotype switching after NN? Using chemogenetic tools, Romoli *et al.* (6) observed that chronic activation of VTA glutamatergic neurons boosted Nurr1 expression, which led to the switch of neuronal phenotypes. Therefore, switching from glutamatergic to DAergic neurons is primed by a multistep process.

Romoli *et al.* (6) depict a novel mechanism through which NN primes the reward circuit to fundamentally change DA signaling in adulthood upon re-exposure to nicotine. These findings also raise several important questions for future addiction research. First, does the neurotransmitter respecification between glutamate and DA occur naturally without exposure to NN, or does NN exposure induce a fate for neurons that they would not normally adopt without exposure to NN? Research has not provided a clear and concise answer, but Romoli *et al.* (6) conclude that the latter is true where the population of glutamatergic neurons are primed to a fate that they would not have normally acquired without NN exposure. Further evidence suggests that activity-dependent transmitter specification does not result in neurogenesis or apoptosis but possibly showcases a "transfating or repurposing" of the reserve pool neurons in which the switch takes place (7). This neuronal plasticity involving DAergic neurons has also occurred in response to physiological levels of environmental illumination and in response to lesions in rodent models of Parkinson's disease in different brain regions. Therefore, it has been hypothesized that neurotransmitter switching might have evolved in the brain to respond to physiological demands and to repair damage. However, research still needs to be conducted to address whether this novel form of plasticity between glutamate and DA occurs everywhere in the brain, in which circuits it occurs, and what machinery or lack thereof the reserve pool of neurons possess.

Second, how are VTA GABAergic neurons affected by NN? While Romoli *et al.* (6) did not detect the DA switch in GABAergic neurons, they detected sensitive responses from these neurons upon NN. For instance, an upregulation of nAChR transcripts of α4 subunit was also observed in GABAergic neurons. nAChRs are critical for neurotransmitter release, neuronal excitability, and activity-dependent plasticity; in the midbrain, they modulate DA neuronal activity and DA release (2). As in the case of nicotine, these receptors have been critically implicated in the reinforcement, tolerance, and sensitization to nicotine. Future studies are warranted to examine the circuit and behavioral consequences of NN-induced adaptations in these VTA GABAergic neurons.

Third, are corresponding morphological changes also induced in VTA neurons that switch phenotypes after NN? Glutamatergic and DA projections to the NAc exhibited anatomic differences after chronic exposure to drugs (10). It is important for future studies to determine whether the NN-induced neurotransmitter switch is accompanied by morphologic switches to adapt to the newly acquired DAergic transmission. Romoli *et al.* (6) provide a foundation to address these and other important questions, and call loudly for smoking parents to think twice when pulling out the next cigarette.

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Article Information

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References

1. Nestler EJ (2005): Is there a common molecular pathway for addiction? J Nat Neurosci 8:1445–1449.
2. Faure P, Tolu S, Valverde S, Naudé J (2014): Role of nicotinic acetylcholine receptors in regulating dopamine neuron activity. Neuroscience 282:86–100.
3. West AE, Griffith EC, Greenberg ME (2002): Regulation of transcription factors by neuronal activity. Nat Rev Neurosci 3:921–931.
4. Behnke M, Smith VC, Committee on Substance Abuse, Committee on Fetus and Newborn (2013): Prenatal substance abuse: Short-and long-term effects on the exposed fetus. Pediatrics 131:e1009–e1024.
5. Bruin JE, Gerstein HC, Holloway AC (2010): Long-term consequences of fetal and neonatal nicotine exposure: A critical review. Toxicol Sci 116:364–374.
6. Romoli B, Lozada AF, Sandoval IM, Manfredsson FP, Hnasko TS, Berg DK, Dulcis D (2019): Neonatal nicotine exposure primes midbrain neurons to a dopaminergic phenotype and increases adult drug consumption. Biol Psychiatry 86:344–355.
7. Dulcis D, Spitzer NC (2012): Reserve pool neuron transmitter respecification: Novel neuroplasticity. Dev Neurobiol 72:465–474.
8. Arias-Carrión O, Stamelou M, Murillo-Rodríguez E, Menéndez-González M, Pöppel E (2013): Dopaminergic reward system: A short integrative review. Int Arch Med 3:24.
9. Borodinsky LN, Root CM, Cronin JA, Sann SB, Gu X, Spitzer NC (2004): Activity-dependent homeostatic specification of transmitter expression in embryonic neurons. Nature 429:523–530.
10. Volkow N, Morales M (2015): The brain on drugs: From reward to addiction. Cell 162:712–725.