severely perturbed in the adult, indicating that there is a critical period for columnar specification during the first postnatal week and that correlated activity is required in this period for column formation.

This study signifies the dawn of a new era—one can imagine that in the near future different populations of RGCs labeled with XFPs will allow visual neuroscientists to discern, in a single preparation, the relationship of axon terminals of different types of RGCs in the SC, LGN, or other visual centers, therefore revealing convergence of information coded by different channels from the retina. Furthermore, it will be possible to visualize developmental interactions between terminals of different populations of RGCs to establish maps in higher visual centers. Molecular mechanisms found to regulate map formation (Luo and Flanagan, 2007) can now be tested in these preparations at the global rather than individual level.

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Confused about NMDA and Addiction? Targeted Knockouts Provide Answers and New Questions

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NMDA-dependent plasticity in VTA dopamine neurons has been hypothesized to be an important first step in the development of long-term changes in the brain reward circuitry that underlie addiction. Two papers from Zweifel et al. and Engblom et al. in this issue of *Neuron* raise new questions concerning the role of NMDA receptors within VTA dopamine neurons in mediating the behavioral effects of drugs of abuse.

Many theories on the development of addiction to drugs of abuse suggest that repeated exposure to these substances co-opts and overpowers the neural circuitry utilized by natural rewards to motivate behavior. By their association with the behavioral effects of the drug, stimuli in the environment become strongly associated with the drug's reinforcing properties. The development of these learned drug associations is thought to contribute to the progression from casual drug use to compulsive drug relapse. Supporting this view is a large body of evidence showing that drugs of abuse can alter learning-related synaptic plasticity mechanisms within the brain's reward processing circuitry. One key area for the development and expression of behaviors associated with drug addiction is the ventral tegmental area (VTA). The VTA contains the dopamine (DA)-containing neurons that project to reward-associated areas of the brain such as the prefrontal cortex and nucleus accumbens. Stimulation of glutamate receptors within the VTA appears to be a critical first step in the development of drug-induced behaviors in experimental animals thought to model the development of compulsive drug seeking, such as conditioned place preference (CPP—a preference for environments associated with the drug) and locomotor sensitization (the progressive increase in the locomotor effects of psychostimulant drugs such as cocaine or amphetamine). Correspondingly, most previous studies reveal that injections of NMDA receptor antagonists directly into the VTA block the development of these addiction-related behaviors.

The cellular mechanisms whereby NMDA receptor stimulation in the VTA is necessary to develop CPP or sensitization was first provided in a series of papers by Bonci and colleagues (Borgland et al., 2004; Ungless et al., 2001). These authors reported that a single injection of cocaine or other addictive drugs increases the strength of glutamatergic synapses on VTA DA neurons. Similar to the strengthening of glutamate synapses in the hippocampus by long-term potentiation (LTP), this increase in synaptic strength was produced by the addition of new AMPA

receptors to the synapse and required the activation of NMDA receptors. Blocking NMDA receptors during exposure to cocaine prevented both the augmentation of glutamatergic synapses as well as the expression of locomotor sensitization, thus suggesting a causal link between the two phenomena.

To extend the experimental proof for an obligatory role of NMDA receptors in the VTA in developing addiction, two papers published in this issue of *Neuron* (Engblom et al., 2008; Zweifel et al., 2008) genetically delete NMDAR1 (NR1) specifically within DA cells by using a Cre recombinase system that selectively prevents the synthesis of NR1 in cells expressing the DA transporter (DAT), a marker for most populations of DA neurons. This constitutes an elegant extension of previous studies where the role of NMDA receptors could not be specifically linked to the DA cells due to the fact that local injection of NMDA blockers into the VTA inactivates NMDA receptors in all cell types. Thus, the initial hypothesis for both of these studies was that elimination of NR1 specifically in VTA DA cells would inhibit the development of CPP and behavioral sensitization. Surprisingly, what at the outset seemed to be a relatively straightforward experimental proof of the critical role of NR1 has instead raised new questions about the role of NMDA-dependent synaptic plasticity within VTA DA neurons in the development of drug-related behaviors. Thus, although the loss of NR1 prevented the cocaine-induced augmentation in the strength of glutamate synapses onto VTA DA cells, both papers report that the initial development of locomotor sensitization following repeated cocaine exposure was unchanged compared to wild-type mice. Moreover, the role of NR1 in the development of CPP was also uncertain, as the two reports strongly differ in their findings. Zweifel et al. (2008) report that the loss of NR1 expression in DA neurons completely blocked cocaine-induced expression of CPP, while Engblom et al. (2008) found that the development of CPP was unaltered in the knockout mice. Although these two labs utilized slightly different approaches in generating DA cell-specific NR1 knockout mice, it is unclear that these differences can account for their discrepant findings.

In light of these mixed results, the authors were hard-pressed to fully reconcile their findings with the prevailing view of a potent role for NMDA receptors in the VTA in regulating the development of behaviors thought to model aspects of addiction. In combination with possibilities proposed by the authors, we offer the following suggestions. The neuroplasticity previously observed in VTA glutamatergic synapses is short lasting and dissipates over a few days after the last injection (Borgland et al., 2004). Thus, it is thought unlikely that cocaine induced plasticity in VTA glutamatergic synapses is the primary cellular substrate for long-term drug-induced behavioral changes; rather, the LTP-like state might constitute an important first step for triggering subsequent long-term adaptations elsewhere within reward circuitry (Kauer, 2004). Given the transient nature of the cellular response induced by the drug itself, it is important to consider that the gene deletions produced do not mimic this time course. Both groups are to be applauded for going to great lengths to extend the constitutively deleted animal model to a more transient model either using viral transfection in the VTA or a tamoxifen-inducible deletion. However, even these latter models will produce stable deletions over many days that have the potential to induce cellular adaptations that may countermand the expected biological effect of DA-selective NR1 deletion. For example, both authors report a surprising increase in AMPA EPSCs in drug-naive NR1 knockout mice.

A second consideration related to the time course of how NMDA receptor stimulation in the VTA may affect the development of addiction-related behaviors is that although both sets of authors have divergent data regarding CPP, when more enduring measures of cocaine-induced behavioral plasticity were measured, both research teams showed that NR1 deletion impaired long-term behavioral plasticity. This is important because the enduring quality of drug-induced behavioral changes is thought to be a critical characteristic of addiction-related behaviors, for example, the vulnerability to relapse that can endure for years after the last drug exposure in addicts. Thus, Zweifel et al. (2008) show that locomotor sensitization is attenuated if animals undergo a withdrawal period that would normally not affect or enhance behavioral sensitization, and Engblom et al. (2008) found that the reinstatement of CPP by a cocaine injection was abolished after a withdrawal period in which animals were extinguished to the cocaine-preferring side. Thus, while there is confusion between the papers regarding the short-term induction of CPP or sensitization, NR1 deletion successfully abolished the capacity of these behaviors to endure. This is consistent with the possibility that NR1 deletion is preventing the translation from initial drug-induced plasticity in DA cells in the VTA to more widespread enduring plasticity in other regions of reward circuitry, such as the prefrontal cortex or nucleus accumbens.

A final potential contribution to how NR1 in the VTA contributes to the development of drug-induced behavioral changes that was not considered in these studies is that VTA DA neurons projecting to the prefrontal cortex express little or no detectable levels of DAT (Lammel et al., 2008). Since both labs used a knockout strategy that utilized DAT expression as the switch to turn off the expression of NMDA receptors, it is possible that NMDA receptors within prefrontal-projecting DA neurons were spared in these mice, as was reported by Engblom et al. (2008) for the hypothalamic DA neurons which also contain little DAT. Also, the prefrontal projecting DA neurons were likely not evaluated electrophysiologically in these papers, as they do not express the hyperpolarization-induced voltage sag that was used as the electrophysiological marker to distinguish DA cells (Lammel et al., 2008). The prefrontal cortex appears to play important roles in behavioral sensitization. For example, pharmacological manipulations in the prefrontal cortex or lesions block sensitization, while repeated electrical stimulation of the prefrontal sensitizes animals to subsequent cocaine exposure (Steketee, 2005; Tzschentke, 2001). DA release within the prefrontal cortex can modulate synaptic plasticity within this cortical area, and prefrontal neurons projecting to the VTA can regulate the activity of DA neurons (Gurden et al., 2000). Thus, in mice in which a gene deletion is driven by the DAT promoter, it is possible that NR1-mediated neuroplasticity in prefrontal circuitry will remain intact.

Taken together, these two papers are consistent in showing that NR1 deletion selectively in VTA DA cells impacts drug-induced behaviors, such as CPP and sensitization, especially with regards to altering the ability of behavioral neuroplasticity to endure after a period of withdrawal. In contrast, the studies are in disagreement on the role of NR1 in some aspects of short-term behavioral plasticity, notably CPP. In general, these studies constitute an elegant proof that is consistent with the body of work indicating an important role for NMDA receptors in the VTA in developing addiction-related behaviors. However, as with all experimental proofs, when looking at discrepancies between studies, it is important to consider possible caveats that may influence the data outcome. In the case of the present studies, this includes possibilities that the neuroplasticity induced by deleting NR1 over the course of days or weeks may impact the subsequent drug-induced behaviors in unpredictable ways and the fact that the potentially critical prefrontal projecting DA cells may not sustain NR1 deletion since they have low or nonexistent expression of DAT. Regardless, it is a rare opportunity to view two such excellent studies side by side and be afforded the opportunity for direct comparisons in how two leading laboratories in addiction research use similar animal models to develop support (or lack thereof) for a long-standing hypothesis; namely, the role played by NMDA receptor-dependent plasticity in the development of addiction.

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Time to Change: Retina Sends a Messenger to Promote Plasticity in Visual Cortex

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Maturation of GABA inhibitory circuitry in primary visual cortex activates the critical period of plasticity, but the underlying mechanisms are not well understood. In the August 8th issue of *Cell*, Sugiyama et al. demonstrate that visual experience promotes the passage of a retina-derived homeoprotein along the visual pathway, which nurtures subclasses of cortical interneurons implicated in regulating critical period plasticity.

The assembly of neural circuits is often shaped by experience in postnatal life. For example, during a brief postnatal period, the closure of one eye can permanently shift the response property of neurons in the primary visual cortex (V1) to favor inputs from the open eye (ocular dominance shift). Since the discovery of ocular dominance plasticity several decades ago (Wiesel and Hubel, 1963), generations of neuroscientists have been making progress toward understanding how a mere imbalance of inputs from the two eyes, a seemingly innocuous manipulation, can profoundly alter neural circuit structures in the cortex, and why this occurs only during a defined critical period.

To shift their eye preference following monocular deprivation (MD), visual cortical neurons must first be able to detect the imbalance of converging visual inputs, relayed to the cortex as altered spiking patterns in thalamic axons, before they can engage a cascade of molecular, cellular, and circuitry mechanisms to weaken the deprived-eye-associated inputs, strengthen the open-eye-associated inputs, and reorganize a balanced network accordingly. GABAergic interneurons are crucial in shaping and detecting the precise spatiotemporal patterns of electrical signaling in the network, including those involved in synaptic plasticity. In recent years, accumulating evidence suggests that proper functioning of GABAergic inhibitory neurons in V1 are critical to establishing the physiological circuit architecture that allows OD plasticity to proceed.