with depressive symptoms. However, ketamine's dissociative side effects and abuse potential has led to the search for identification of alternative compounds that trigger rapid antidepressant effects without the psychotomimetic side effects. A successful effort will require elucidation of the molecular mechanisms that elicit the antidepressant effects of ketamine. In 2011, our group proposed a synaptic signaling pathway that can account for ketamine's antidepressant-like effects in preclinical animal models (Autry *et al*, 2011). Specifically, we showed that ketamine-mediated block of resting synaptic NMDA receptor (NMDAR) activity—driven by spontaneous glutamate release—leads to deactivation of eEF2 kinase resulting in dephosphorylation of its sole known target eEF2. The dephosphorylation of eEF2 results in desuppression of dendritic protein translation and a rapid subsequent increase in brain-derived neurotrophic factor (BDNF) expression. Released BDNF activates TrkB receptors and triggers a subsequent potentiation of AMPA receptor (AMPA)-mediated synaptic transmission in the hippocampus, providing a synaptic basis for the antidepressant effects of ketamine (Autry *et al*, 2011; Nosyreva *et al*, 2013). Consistent with the premise, ketamine's antidepressant effects have previously been shown to require AMPARs as NBQX, an AMPAR antagonist, attenuates the antidepressant effects in rodents (Maeng *et al*, 2008; Autry *et al*, 2011). Importantly, the model we proposed can account for the ineffectiveness of the NMDAR blocker memantine as an antidepressant in the clinic, as memantine—unlike ketamine—has a limited ability to block resting NMDAR function. However, a recent study challenged a key component of the model by suggesting that ketamine's antidepressant action is mediated by its metabolite 2R,6R-hydroxynorketamine (2R,6R-HNK), via the same signaling pathway and AMPAR-mediated synaptic potentiation as outlined above (Autry *et al*, 2011) but in a NMDAR-independent manner (Zanos *et al*, 2016). Validation of the same

signaling pathway and effects on AMPAR-mediated potentiation—sans NMDAR block—prompted us to investigate the impact of 2R,6R-HNK on NMDAR-mediated neurotransmission. In recent experiments, we showed that 2R,6R-HNK can swiftly inhibit NMDAR transmission up to 50% at concentrations needed to trigger the intracellular signaling pathway involved in the potentiation of synaptic AMPAR responses, strongly supporting a role for NMDAR-blockade in this effect (Suzuki *et al*, 2017). Further characterization showed that the effect of 2R,6R-HNK on individual NMDA-mEPSCs mimicked those of ketamine, demonstrating that 2R,6R HNK—like ketamine—selectively inhibits NMDARs while they are open (Suzuki *et al*, 2017). Our study demonstrated that 50 μ M 2R,6R-HNK blocks synaptic NMDARs and thus 2R,6R-HNK is not 'NMDAR-independent' in its action. Although we used a higher concentration than the 10 μ M 2R,6R-HNK—suggested as the brain concentration in rodents following a behaviorally effective antidepressant dose of 10 mg/kg 2R,6R-HNK (Zanos *et al*, 2016)—a recent study was not able to detect antidepressant effects of 10 mg/kg 2R,6R-HNK in the same behavioral paradigms (Yang *et al*, 2017) used by Zanos and colleagues, raising questions about the dose of 2R,6R-HNK necessary to mediate antidepressant effects and the brain concentration necessary to trigger the molecular pathway. Moreover, a mechanism, other than the demonstrated NMDAR-blockade of 2R,6R-HNK, remains to be demonstrated. The selective action of 2R,6R-HNK on open NMDARs indicate its efficacy as an antidepressant will strongly depend on factors that impact synaptic NMDAR channel opening. These factors may include glutamate release-probability, subunit composition of synaptic NMDARs, as well as levels of coagonists. Future studies will need to take into account these synaptic factors to elucidate the specific circuits involved in the antidepressant action of ketamine as well as 2R,6R-HNK. Overall, our findings demonstrate a critical

role of NMDAR-block-mediated signaling in triggering the antidepressant effects of ketamine and 2R,6R-HNK.

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Cognitive Signaling in Cerebellar Granule Cells

The cerebellum contains over three quarters of the neurons in the human brain (Herculano-Houzel, 2010), but it has traditionally been studied mainly in sensorimotor contexts. Yet in humans, cerebellar circuits activate during and are required for verbal and spatial processing tasks (eg, Stoodley *et al* (2012)), and multiple lines of evidence point to cerebellar links

to psychoses such as schizophrenia as well as to autism (reviewed in Sokolov *et al* (2017)). Moreover, the cerebellum is widely connected with multiple cognitive regions of the neocortex (eg, Strick *et al* (2009)), with lateral cerebellar regions preferentially connected to frontal neocortical areas, both of which are expanded in the primate lineage. Despite this evidence, little is known about the putative cognitive signals that reach the cerebellum at the cellular physiological level.

In particular, the cerebellar input layer that receives external information consists of granule cells. Cerebellar granule cells receive only a few inputs that can arise from a multitude of places throughout the brain and sensory periphery, indicating they may be well positioned to receive and transmit an array of non-sensorimotor signals to the cerebellum. Because of the longstanding technical difficulties arising from their small size and dense packing, however, granule cell responses have not been recorded during cognitive tasks.

We recently performed two-photon calcium imaging in ensembles of individual cerebellar granule cells during conditioning tasks where mice learned to expect upcoming rewards (Wagner *et al*, 2017). In one such task, thirsty mice used their forepaw to move a small handle forward for delayed receipt of a water reward. Surprisingly, some granule cells preferentially responded to expected or unexpected rewards, or to the omission of expected rewards. Other cells selectively encoded the cognitive state of expectant waiting for an upcoming reward. Multiple experiments and analyses indicated that these reward signals were unexplained by motor signals such as licking or body movement. Granule cell reward signals were present in multiple different reward-related behavioral tasks, and emerged over the course of task learning.

These results raise a host of new questions. Foremost, it will be critical to understand the origin of reward-related signaling in the cerebellum. Anatomical tracing did not reveal

direct input to the cerebellar cortex from midbrain dopamine cells like those of the ventral tegmental area that are known to convey reward signals (Cohen *et al*, 2012). On the other hand, the cerebellar regions we studied received robust input from the pontine nuclei, which relay projections to cerebellum from many regions of the neocortex (Strick *et al*, 2009), which is a prime candidate for further study. As important for future work will be determining how the cerebellum uses cognitive signals in its own computations, and what role any cerebellar cognitive output has in downstream circuits like those of the cortex. This would help determine whether cerebellar output could be a translational target for schizophrenia or psychosis. Integrating cerebellar circuits into the brain's broader reward-related computations may help to shed light on the mechanisms of cerebellar involvement in cognitive processes.

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The Promise of Genome Editing for Modeling Psychiatric Disorders

A sizeable fraction of the risk for psychiatric illness can be attributed to the genes that we inherit (Polderman *et al*, 2015). The challenge of identifying gene variants that influence vulnerability to psychiatric illness, which often operate only after interacting with environmental risk factors, is daunting. Nevertheless, advances in genomics are yielding new insights into these risk-modifying gene variants.

A major breakthrough that will facilitate our understanding in this area is the development of CRISPR/Cas9 (clustered regularly interspaced short palindromic repeats) (Cong *et al*, 2013; Jinek *et al*, 2013); Heidenreich and Zhang, 2016). CRISPR technology utilizes a bacterial borne RNA-guided DNA nuclease defense mechanism to elicit specific gene mutations. Scientists have exploited the mechanism by which bacteriophages transcribe part of a pathogenic genome into guide RNAs (gRNAs) that, as their name implies, serve to guide the endonuclease, Cas9, to cut the DNA of the invading pathogen. The resulting DNA breaks are repaired by either non-homologous end joining (NHEJ) or homology-directed repair (HDR). The NHEJ pathway efficiently ligates broken ends of DNA strands, but often introduces nucleotide insertions or deletions resulting in frameshift mutations that can disrupt expression of the repaired gene. The less efficient but higher fidelity HDR pathway includes a specific DNA sequence to be inserted at the break site.

Using CRISPR-mediated NHEJ, mutations in genes of interest can be generated efficiently, thus reducing the time between gene discovery and investigation of mechanisms of action. Delivery of the CRISPR components (gRNA, tracrRNA, and Cas9) to cells or model organisms is efficient and can, for example, be accomplished through the transfection of mRNA or delivery via adeno-associated viruses