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What's the buzz about hydroxynorketamine? Is it the history, the story, the debate, or the promise?!

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The History

Early in the 1990s, a set of animal studies presented N-methyl-D-aspartate receptor (NMDAR) modulation as a common pathway to traditional antidepressants (1). Later in 1990s, a group of Yale scientists was set out to demonstrate the role of NMDAR modulation in clinical depression using subanesthetic doses of the NMDAR-antagonist ketamine. Surprisingly, they discovered that a single ketamine infusion exerted rapid acting antidepressant (RAAD) effects which were sustained for 3 days, well beyond the short half-life of the ketamine compound (2). Over the next decade, following the replication of the RAAD finding of ketamine and in the context of preclinical evidence of glutamate excitotoxicity, the inhibition of NMDAR gained attention as a putative novel mechanism to induce RAAD effects. However, increasingly it became evident that several non-ketamine NMDAR-antagonists do not possess RAAD properties (e.g. memantine). In addition, the antidepressant doses of ketamine induce a paradoxical glutamate neurotransmission surge, rather than inhibition. Late in the 2000s, a critical role for the ketamine-induced glutamate stimulation was supported first by showing that α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor (AMPA) blockers—which reduce both AMPAR and NMDAR signaling— inhibit the RAAD effects of ketamine (3), and second by demonstrating that ketamine RAAD properties are dependent on its induction of synaptogenesis, which is also inhibited by AMPAR blockers (4). Moreover, various non NMDAR-antagonist drugs—which presumably stimulate glutamate neurotransmission—were found to exert ketamine-like synaptogenesis and RAAD effects, including: 1) scopolamine a muscarinic cholinergic receptor (m-AChR) antagonist; 2) LY 341495 a metabotropic glutamate receptor 2 and 3 (mGluR_{2/3}) antagonist; and 3) Rapastinel an NMDAR modulator with partial agonist properties (Fig. 1). Further underscoring the role of

glutamate stimulation, AMPAR blockers were also found to inhibit the RAAD effects of scopolamine, mGluR_{2/3} antagonists, and rapastinel. Together, accumulating evidence over the past decade presented transient AMPAR and NMDAR stimulation as a primary underlying mechanism of RAADs (5). Complementing this model, it is believed that the RAAD effects of ketamine are further enhanced by its various pharmacodynamic properties, including its activity independent blockade of NMDAR (6) and its regulation of the inflammatory, opioid, and monoamine systems (5).

The Story

Ketamine is a racemic mixture of *R*- and *S*-ketamine. Each of the *R*- and *S*-ketamine enantiomers is metabolized into norketamine (NK), hydroxyketamine (HK), dehydronorketamine (DHNK), and hydroxynorketamine (HNK). The HNK metabolites include: Z-6-HNK, E-6-HNK, Z-5-HNK, E-5-HNK, Z-4-HNK, and E-4-HNK. Of these HNKs, Z-6-HNK [a.k.a., (2S,6S;2R,6R)-HNK] is the most abundant metabolite following ketamine administration, and its concentration is higher in females (7, 8). (2S,6S;2R,6R)-HNK lacks the anesthetic effects of ketamine and is not active on the NMDAR, with preliminary studies showing inhibitory effects on α 7-nicotinic-AChR; a finding that awaits replication. In contrast to the short half-life of ketamine, human plasma (2S,6S;2R,6R)-HNK is significantly present at 4h, and is quantifiable at 24h, post infusion of a subanesthetic dose of ketamine (8). In a pilot study of major depressive disorder (MDD; medication-free; 38% females) and bipolar depression (BD; medicated by valproate or lithium; 73% females) patients, the investigators found significantly higher (2S,6S;2R,6R)-HNK in the BD group compared to MDD, but there were no significant response or response-by-diagnosis effects. The lack of relationship between plasma (2S,6S;2R,6R)-HNK and response to ketamine remained after including gender in the statistical model. In the BD group, patients treated with the mood stabilizer valproate (a potent inhibitor of Cytochrome P450) were found to have higher plasma (2S,6S;2R,6R)-HNK levels (8). In summary, these pilot human data failed to show a statistically significant relationship between (2S,6S;2R,6R)-HNK and the RAAD effects of ketamine. However, there was evidence of higher plasma (2S,6S;2R,6R)-HNK in BD compared to MDD, although the groups were not well matched for gender and medication status. In rodents, it was previously reported that the (2S,6S)-HNK enantiomer increases the mammalian target of rapamycin complex 1 (mTORC1) function, an intracellular signaling pathway believed to play an essential role in the ketamine induced synaptogenesis and RAAD effects. Recently, Zanos et al. reported a more comprehensive rodent study which fully investigated the role of (2S,6S;2R,6R)-HNK in the antidepressant effects of ketamine (7). The authors found sustained (i.e., 24h post infusion) antidepressant-like effects in mice treated with R,S-ketamine or R-ketamine, but not with S-ketamine (all doses were 10mg/kg). They also found higher antidepressant-like effects and higher brain (2S,6S;2R,6R)-HNK concentration in females. Deuterated ketamine, with reduced conversion to (2S,6S;2R,6R)-HNK, failed to induce sustained antidepressant-like effects. Moreover, 5mg/kg of (2R,6R)-HNK induced sustained antidepressant-like effects in mice, while the effects of (2S,6S)-HNK were only evident at 25–75mg/kg. After confirming the low affinity of (2S,6S;2R,6R)-HNK to NMDAR, the investigator demonstrated that in vitro application of (2R,6R)-HNK to rat hippocampal slices induces an increase in excitatory

postsynaptic potentials (EPSP) and currents (EPSC), which were inhibited by an AMPAR blocker. Noticeably, the administration of AMPAR blocker both prior to, or 24h post, (2R,6R)-HNK infusion inhibited its sustained antidepressant-like effects. Similar findings were demonstrated with ketamine. Ketamine and (2R,6R)-HNK increased mature Brain Derived Neurotrophic Factor (BDNF) level and AMPAR GluA_{1/2} subunits in the hippocampus, but not in the prefrontal cortex. Finally, (2R,6R)-HNK appeared to lack the side effects evident in mice following ketamine administration.

The Debate

In the current issue, Collingridge and colleagues acknowledge the possibility that (2S,6S;2R,6R)-HNK may have RAAD properties, which remain to be demonstrated in clinical studies (9). However, they warn against prematurely rejecting the role of NMDAR inhibition in the mechanisms of RAADs. Collingridge et al. note that various NMDAR-antagonists (e.g. Traxoprodil; a.k.a. CP-101,606), which do not have (2S,6S;2R,6R)-HNK as metabolite, have shown RAAD effects in animal models and clinical trials. In addition, the authors highlight the difficulties in translating Zanos et al. findings in mice to humans. For example, while low dose S-ketamine and (2S,6S)-HNK failed to induce RAAD effects in the Zanos et al. mice study, it was already shown that S-ketamine—even at doses that are 40% of the regular ketamine antidepressant dose—has RAAD effects in depressed patients. Moreover, the authors raised concerns whether the (2S,6S;2R,6R)-HNK brain concentration required for efficacy in rodents can be achieved in humans. Finally, Collingridge et al. presented a hypothetical model in which depression is the result of LTD changes in hedonic regions and that ketamine blocks these LTD effects during reconsolidation (9). Of note, while the presented model is simple and conceivable, it is speculative in nature. It also does not account for extensive literature associating depression with LTP-like, not LTD, changes in the Nucleus Accumbens and that subanesthetic ketamine induces LTD-like changes in this hedonic brain region.

Zanos and colleagues responded by acknowledging the need for human evidence to firmly conclude that (2S,6S;2R,6R)-HNK is a RAAD (10). Then, they highlight that high doses of S-ketamine and (2S,6S)-HNK did show minimal antidepressant-like effects in their study. Considering that there are no human trials comparing S-ketamine to ketamine, the authors correctly refuted the statement by Collingridge et al. that “S-ketamine was roughly twice as potent as racemic intravenous (R,S)-ketamine”. The authors acknowledged that high concentrations were used to demonstrate the effects of (2R,6R)-HNK on AMPAR transmission, but they underscored the uncertainty about human brain concentration of (2S,6S;2R,6R)-HNK and suggested that this concentration may be higher in humans than in mice. Finally, the authors argued that the traxoprodil RAAD effects do not appear to be fully comparable to ketamine, and that these human findings were not replicated (10). However, it is important to note that there are no human trials comparing traxoprodil to ketamine and I am not aware of failed traxoprodil replication trials.

The Promise

Two decades of glutamate modulation research have provided great insight into the neurobiology of depression and the putative mechanisms of RAADs (5). However, to date, there are no FDA-approved RAAD agents. Some investigational drugs (e.g. ketamine, traxoprodil) have shown promising clinical results. Yet, for various economical, safety, and scientific reasons, these agents did not go through the scrutiny and rigorous assessment of the FDA approval process. In that context, the (2S,6S;2R,6R)-HNK findings are highly promising, potentially opening the door for a new line of drug development that may ultimately lead to effective and well tolerated RAADs. However, it is critical to reiterate that while the (2S,6S;2R,6R)-HNK preclinical findings are promising and that there is evidence of NMDAR-independent RAADs (e.g. scopolamine), the current literature data is not yet sufficient to provide evidence of clinical RAAD properties by (2S,6S;2R,6R)-HNK or to fully reject the role of NMDAR in the mechanisms of RAADs. In fact, the pilot human data available failed to show a relationship between plasma (2S,6S;2R,6R)-HNK and response to ketamine in MDD patients (8). Furthermore, currently there is preliminary clinical evidence to support the potential RAAD properties of the NMDAR-antagonists (S)-ketamine and traxoprodil [cited in (9, 10)], but not yet for (2S,6S;2R,6R)-HNK. So, what's the buzz about HNK?! I believe it is the indispensable promise of a rapid acting antidepressant with limited adverse events, a promise that —if realized— it could reduce suffering of millions of patients all around the world.

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Abbreviations

Glu	glutamate
Gln	glutamine
NR1	NMDA receptor unit 1
NR2B	NMDA receptor unit 2B
VSOAC	volume-sensitive organic anion channel
EAAT	excitatory aminoacid transporter

References

1. Skolnick P, Layer RT, Popik P, Nowak G, Paul IA, Trullas R. Adaptation of N-methyl-D-aspartate (NMDA) receptors following antidepressant treatment: implications for the pharmacotherapy of depression. *Pharmacopsychiatry*. 1996; 29:23–26. [PubMed: 8852530]
2. Berman RM, Cappiello A, Anand A, Oren DA, Heninger GR, Charney DS, et al. Antidepressant effects of ketamine in depressed patients. *Biol Psychiatry*. 2000; 47:351–354. [PubMed: 10686270]
3. Maeng S, Zarate CA Jr, Du J, Schloesser RJ, McCammon J, Chen G, et al. Cellular mechanisms underlying the antidepressant effects of ketamine: role of alpha-amino-3-hydroxy-5-methylisoxazole-4-propionic acid receptors. *Biol Psychiatry*. 2008; 63:349–352. [PubMed: 17643398]
4. Li N, Lee B, Liu RJ, Banasr M, Dwyer JM, Iwata M, et al. mTOR-dependent synapse formation underlies the rapid antidepressant effects of NMDA antagonists. *Science*. 2010; 329:959–964. [PubMed: 20724638]
5. Abdallah CG, Adams TG, Kelmendi B, Esterlis I, Sanacora G, Krystal JH. Ketamine's Mechanism of Action: A Path to Rapid-Acting Antidepressants. *Depress Anxiety*. 2016
6. Autry AE, Adachi M, Nosyreva E, Na ES, Los MF, Cheng PF, et al. NMDA receptor blockade at rest triggers rapid behavioural antidepressant responses. *Nature*. 2011; 475:91–95. LID - 101038/nature10130 [doi]. [PubMed: 21677641]
7. Zanos P, Moaddel R, Morris PJ, Georgiou P, Fischell J, Elmer GI, et al. NMDAR inhibition-independent antidepressant actions of ketamine metabolites. *Nature*. 2016; 533:481–486. [PubMed: 27144355]
8. Zarate CA Jr, Brutsche N, Laje G, Luckenbaugh DA, Venkata SL, Ramamoorthy A, et al. Relationship of ketamine's plasma metabolites with response, diagnosis, and side effects in major depression. *Biol Psychiatry*. 2012; 72:331–338. [PubMed: 22516044]
9. Collingridge GL, Lee Y, Bortolotto ZA, Kang H, Lodge D. Antidepressant Actions of Ketamine Versus Hydroxynorketamine. *Biol Psychiatry*. 2016
10. Zanos P, Moaddel R, Morris PJ, Wainer IW, Albuquerque EX, Thompson SM, et al. Reply to: Antidepressant Actions of Ketamine Versus Hydroxynorketamine. *Biol Psychiatry*. 2016

Figure 1. Glutamatergic Mechanisms Underlying the Effects of Rapid Acting Antidepressants (RAADs)

It is believed that the rapid increase of synaptogenesis, by modulating prefrontal and hippocampal glutamatergic synapses, is a convergent mechanistic pathway underlying the beneficial behavioral effects of RAADs. Ketamine and scopolamine have been shown to precipitate glutamate neurotransmission surge by blocking interneuronal N-methyl-D-aspartate (NMDA) or muscarinic acetylcholine (mACh) receptors, respectively, leading to inhibition of GABAergic input on glutamatergic neurons. This glutamate surge stimulates α -amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors and activity dependent release of brain derived neurotrophic factor (BDNF), which in turn stimulates mammalian target of rapamycin complex 1 (mTORC1) signaling and increases translation of synaptic proteins. LY341495 induces a glutamate neurotransmission surge presumably by blocking presynaptic metabotropic glutamate receptors (mGluR_{2/3}). Rapastinel is believed to increase postsynaptic glutamate neurotransmission by exerting partial agonist properties on intrasynaptic NMDA receptors. Ketamine, traxoprodil, and Ro25-6981 were proposed to increase synaptogenesis by blocking NR2B-containing NMDA receptors activated by ambient glutamate. (2S,6S;2R,6R)-Hydroxynorketamine [a.k.a., Z-6-HNK] has been shown to increase glutamate transmission and elongation eukaryotic factor 2 (eEF2), however, the underlying pathways leading to these changes are not fully known. Alpha 7 nicotinic ACh ($\alpha 7$ -nACh) receptors induce presynaptic glutamate release as well as increase ambient extrasynaptic glutamate by activating astrocytic glutamate release. Considering preliminary

evidence of $\alpha 7$ -nACh inhibition by (2S,6S;2R,6R)-HNK, future studies should examine whether the (2S,6S;2R,6R)-HNK induced increase in eEF2 is related to its effects on astrocytic $\alpha 7$ -nicotinic-ACh receptors.