Ketamine persists within neuronal compartments long after systemic clearance

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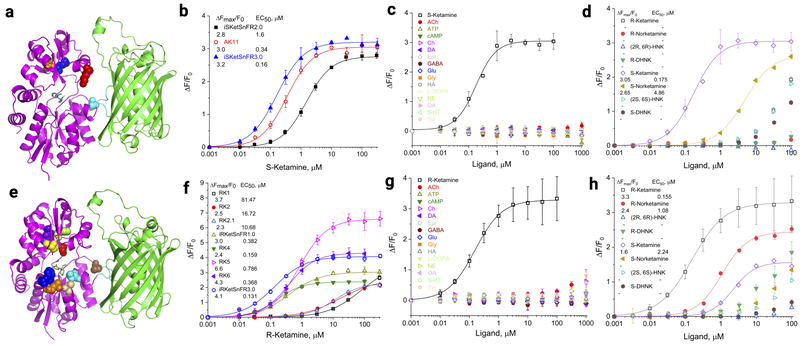
Question -> hypothesis

- Ketamine produces rapid and sustained antidepressant effects after a single dose.
- Plasma membrane NMDA receptors are the presumptive primary molecular targets, yet how ketamine distributes, localizes, and engages neurons across cellular and subcellular compartments remains largely unknown.
- Ketamine, a lipophilic weak base, is thought to accumulate within acidic intracellular compartments, suggesting potential unrecognized sites of action.
- Hypothesis: Ketamine's rapid and sustained antidepressant effects are mediated not only by transient blockade of plasma membrane NMDA receptors, but also by prolonged intracellular accumulation and engagement of neuronal targets within specific subcellular compartments

Ketamine (iKetSnFR) biosensor development

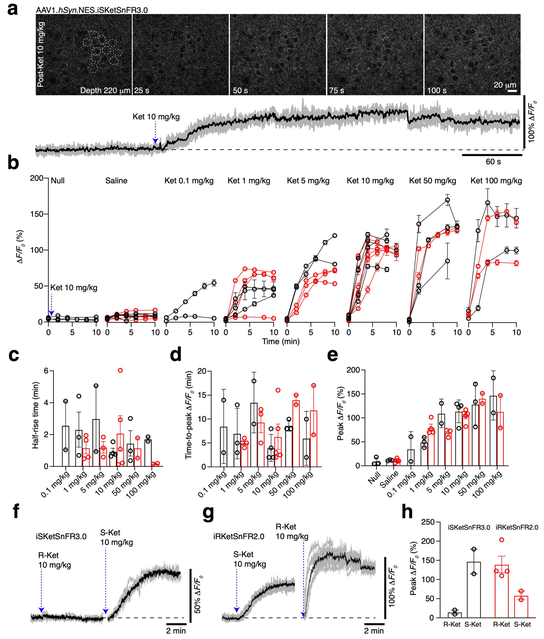
- We developed a toolbox of genetically encoded intensity-based ketamine-sensing fluorescent reporters (iKetSnFRs) that specifically and robustly report R- or S-ketamine with cellular and subcellular resolution *in vitro* and *in vivo*, with excellent sensitivity and fluorescence change.

Structure-guided engineering and characterization of ketamine-sensitive fluorescent sensors.

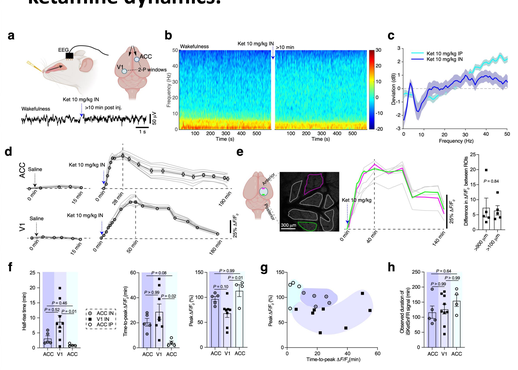


In vivo results

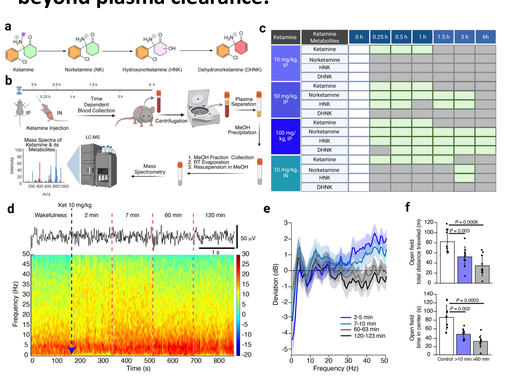
Rapid cortical detection of ketamine in mouse prefrontal cortex following administration.



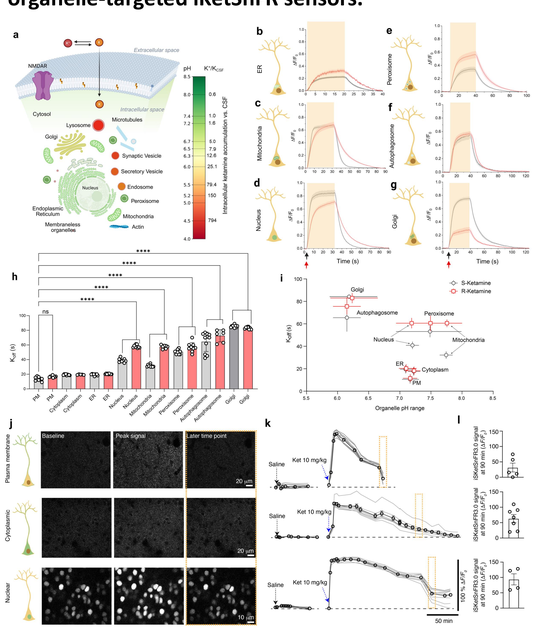
Intranasal ketamine induces rapid cortical desynchronization and sustained, region-specific ketamine dynamics.



Prolonged brain intracellular retention of ketamine beyond plasma clearance.



Subcellular compartmentalization and persistence of ketamine accumulation in neurons revealed by organelle-targeted iKetSnFR sensors.



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Conclusion

- This persistent intracellular engagement coincides with the prolonged timeframe of behavioral effects. Together, these findings link a single ketamine exposure to persistent intracellular drug dynamics and begin to provide a mechanistic basis for its rapid and enduring cellular and behavioral effects.