Sigma-1 Receptor Agonists as Potential Treatment Options for Autism Spectrum Disorders: Pre-clinical Studies with ANAVEX 2-73 in a Fragile X Model

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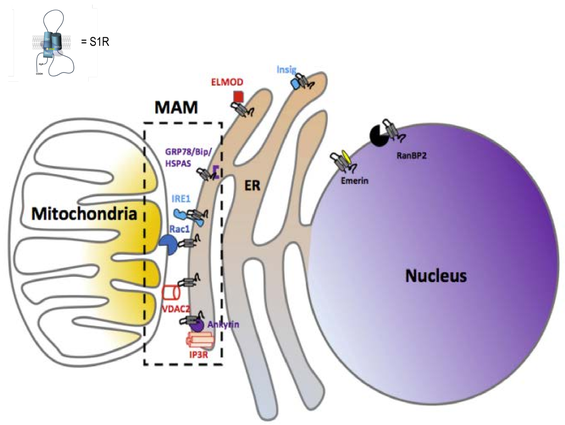
Background

Autism spectrum disorders (ASD) occur in up to 2/3 of males and 1/3 of females with Fragile X syndrome (FXS). FXS is the most common form of inherited intellectual disability and the most frequent single gene cause of autism, affecting approximately 1 in 4,000 males and 1 in 6,000 females. In addition to the clinical overlap between FXS and ASD, there is likely a substantial overlap in the molecular pathology of the two disorders. Molecules aimed at targets in these shared pathways are expected to have therapeutic overlap in subsets of individuals with ASD or neurodevelopmental disorders.

The Fragile X gene FMR1 was cloned in 1991 and has been studied since then using techniques from biochemistry through genetics to model organisms in an effort to elucidate the functions of the FMR1 protein (FMRP). In the brain FMRP is highly expressed in neurons and actively transported as part of a messenger ribonucleoprotein (mRNP)-complex throughout the dendritic fields to the synaptic spines, where its main function appears to be the regulation of protein synthesis. Insufficient expression of FMRP leads to deregulated translation and a broad array of effects on cellular signaling pathways, ultimately leading to abnormalities in brain connectivity and neurodevelopmental processes (Grossman AW et al 2006, Bhakar AL et al 2012).

The sigma-1 receptor (S1R) is an intracellular chaperone protein located at the endoplasmic reticulum–mitochondria interface with important roles in inter-organelle communication. S1R is also involved in transcriptional regulation at the nuclear envelop and restores homeostasis and stimulates recovery of cell function when activated. ANAVEX 2-73 is a S1R agonist that recently demonstrated significant improvements in an array of behavioral and gait paradigms in a mouse model for the neurodevelopmental disease Rett syndrome. In addition, ANAVEX 2-73 has exhibited anticonvulsant, anti-amnesic, neuroprotective and antidepressant properties in various animal models (Lahmy V et al 2013, 2015, Maurice T 2016, Villard V et al 2011). Clinically, ANAVEX 2-73 has demonstrated good safety, bioavailability, and tolerability in Phase 1 and Phase 2a trials, and data from the Phase 2a clinical trial in Alzheimer's disease has demonstrated dose-dependent cognitive improvements.

Sigma-1 Receptor: Key Upstream Cellular Modulator



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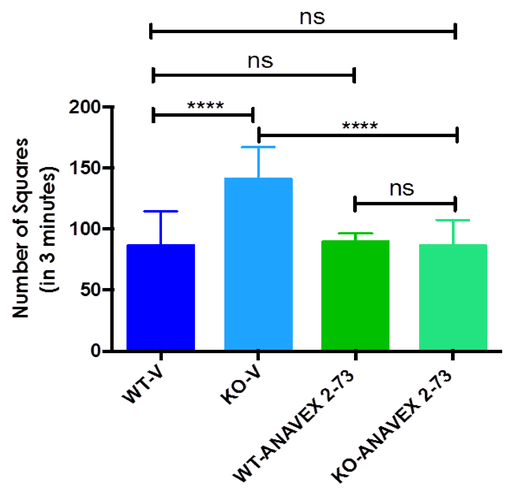
Experimental Procedures

Fmr1-KO2 mice (Mientjes MJ et al 2006) and wild type (WT) littermates, generated on a C57BL/6J background, were used throughout (N=10 per treatment arm). ANAVEX 2-73 was dosed at 1 mg/kg IP twice daily for 14 days. Testing was conducted on male mice that were approximately 2 months old, and the experimenters were blind to both genotype and treatment during all testing and data analyses. Drug-related safety was first assessed and then followed by a battery of 3 behavioral tests served to characterize efficacy-related endpoints: open field test (hyperactivity), contextual fear conditioning (associative learning), and marble-burying (species-specific behavior). All data were analyzed via a one-way ANOVA followed post hoc by a Tukey's multiple comparison test.

ANAVEX 2-73 Safety Profile

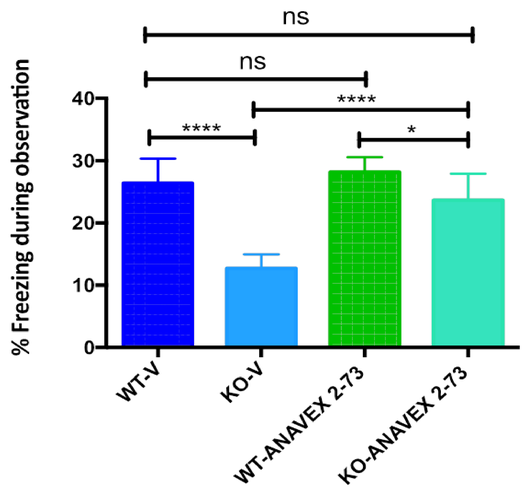
ANAVEX 2-73 was well tolerated by both Fmr1-KO2 mice and WT mice. No signs of a drug effect were observed when mice were assessed for weight loss, fur loss, lacrimal discharges, or changes in gait, alertness, and general behavior.

ANAVEX 2-73 Reverses the Hyperactivity of the Fmr1 KO2 mice to Normal



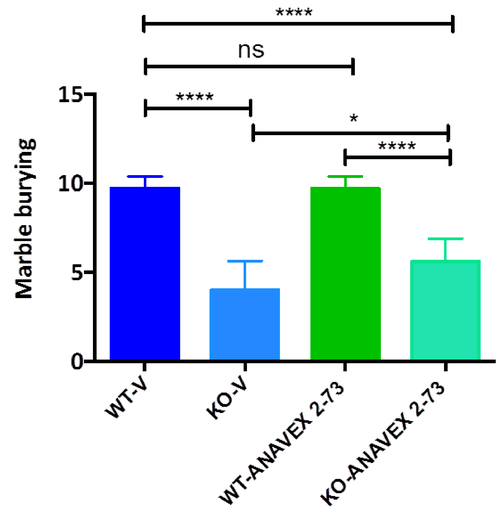
Compared to the corresponding WT animals, vehicle-treated Fmr1-KO2 mice displayed an increase in total distance traveled (number of squares), a measure of general hyperactivity (**** p≤0.0001). Chronic treatment with ANAVEX 2-73 significantly reduced the increased locomotor activity of the Fmr1-KO2 mice (**** p≤0.0001), to the same levels observed in vehicle-treated WT mice.

ANAVEX 2-73 Normalizes the Impairment in Associative Learning Characteristic of Fmr1 KO2 mice



Under conditions of acute stress, ANAVEX 2-73 treatment rescued Fmr1 KO2 mice from the learning deficits observed in the vehicle-treated Fmr1 KO2 group (measured as % freezing during 5 min, **** p≤0.0001). Compared to the vehicle treated WT animals, mice chronically treated with ANAVEX 2-73 exhibited a similar percentage of freezing behavior compared to the vehicle-treated WT mice in the contextual fear conditioning paradigm.

ANAVEX 2-73 Reduces Impairments in Species-Specific Behavior in Fmr1 KO2 Mice



The Fmr1 KO2 mice buried significantly fewer marbles than WT mice (**** p≤0.0001). This behavior was partially rescued by ANAVEX 2-73; the drug-treated Fmr1-KO2 group was statistically different from the Fmr1-KO2 animals that received vehicle (* p≤0.05).

Conclusions

Overall, these findings demonstrate that chronic treatment with ANAVEX 2-73 to Fmr1-KO2 mice has a robust effect on their characteristic hyperactivity and deficits in learning and memory. At the dose tested, ANAVEX 2-73 also yielded a partial effect on species-specific behavior in the form of marble burying.

In conclusion, the ability of ANAVEX 2-73 to significantly reverse the pathophysiological signs of FXS as observed in an animal model of this disorder, together with the positive safety and cognition data obtained in human trials, suggest that this agent could be of interest to investigate clinically in FXS patients and potentially other autism disorders.

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Acknowledgment

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