Development of the N-Methyl-D-Aspartate Receptor (NMDAR) Antagonist d-Methadone (REL 1017) for the Treatment of Depression and Other CNS Disorders.

Ottavio V. Vitolo[1], Paolo L. Manfredi[1], Charles Inturrisi[2], Gina DiGuglielmo[1], Taleen Hanania[3], Galina Bernstein[4], Sara DeMartin[5], Manoela Fogaca[6], Ron Duman[6], Sergio Traversa[1]

1. Relmada Therapeutics Inc., New York, New York, USA Department of Pharmaceutical and Pharmacological Sciences, University of Padova, Padova, Italy
2. Department of Pharmacology, Weill Cornell Medical College, New York, New York, USA
3. PyschoGenics Inc., New Jersey, NJ, USA
4. Syneos Health, Toronto, Canada
5. Department of Pharmaceutical and Pharmacological Sciences, University of Padova, Padova, Italy
6. Yale Medical School, New Haven, USA

Background

NMDA receptor (NMDAR) antagonists are potential agents for the treatment of several central nervous system (CNS) disorders including major depressive disorder.
Racemic methadone and its stereoisomers, l-methadone and d-methadone, bind NMDARs with an affinity similar to that of established NMDAR antagonists, while only l-methadone and racemic methadone bind to opioid receptors with high affinity.
D-methadone is expected to have no clinically significant opioid effects at therapeutic doses mediated by its NMDAR antagonism.
Relmada Therapeutics is developing d-methadone as a potential new treatment for depression and other CNS conditions.

Pre-clinical Studies

We conducted several pre-clinical studies comparing the effect of d-methadone and ketamine in different behavioral animal models commonly used to assess antidepressant activity.
These include the Forced Swim Test, the Female Urine Sniffing Test and the Novelty Suppressed Feeding Test.
We also performed behavioral analysis of the effect of both d-methadone and ketamine on rats exposed to a Chronic Unpredictable Stress (CUS) protocol.
In all of the aforementioned tests, d-methadone like ketamine produced significant improvements in drug treated vs. vehicle treated animals. In addition, we observed positive effects on the expression of synaptic proteins and receptors critically involved in synaptic plasticity.
These biochemical effects were also paralleled by favorable changes in electrophysiology (data not shown).

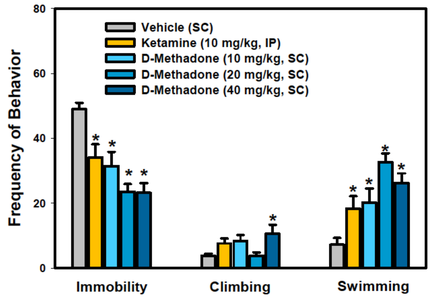


Fig. 1 Effects of ketamine and d-methadone on immobility, climbing and swimming counts in the FST. Data represent mean ± SEM. For immobility: *p=0.0034 for ketamine, 0.0007 for d-methadone 10 mg/kg, and <0.0001 for d-methadone 20 and 40 mg/kg compared to vehicle group, ANOVA. For climbing: *p<0.05 for d-methadone 40 mg/kg vs. vehicle. For swimming: *p<0.05 for ketamine and d-methadone 10 mg/kg, <0.0001 for d-methadone 20 mg/kg, and 0.0003 for d-methadone 40 mg/kg vs. vehicle, ANOVA.

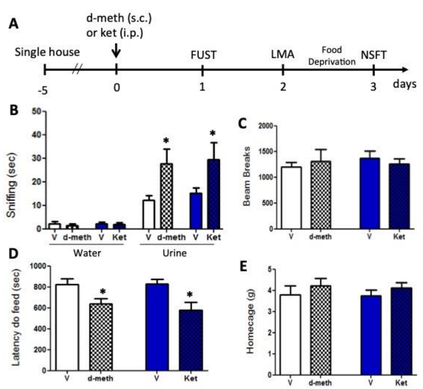


Fig 2. Influence of d-methadone and ketamine on FUST and NSFT.
(A) Rats were administered d-methadone or ketamine and tested in (B) FUST 24 hr later, (C) locomotor activity 2 d later, and in (D) NSFT 72 hr later; (E) home cage feeding.
The results are the mean ± S.E.M. FUST: One-way ANOVA, F3,42 = 3.26, p = 0.031; Fisher's LSD: Veh x Met, p = 0.025; Veh x Ket, p = 0.046; n = 9-12/group. NSFT: One-way ANOVA, F3,27 = 4.87, p = 0.008; Fisher's LSD: Veh x Met, p = 0.035; Veh x Ket, p = 0.005; n = 7-8/group.

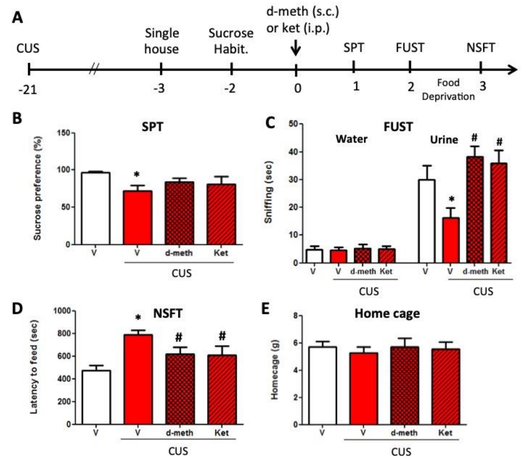


Fig 3. Single dose D-methadone prevents depressive behaviors induced by CUS exposure.
(A) Time-course for the CUS protocol, drug dosing, and behavioral analysis. D-methadone and ketamine prevented the behavioral effects of CUS in the (B) SPT (F3,45= 2.99), (C) FUST (F3,46= 5.43), and (D) NSFT (F3,46 = 6.79). (C) No difference was found for water sniffing or (E) home cage food consumption.
Results are the mean ± S.E.M., n = 9-15/group. P < 0.05, One-Way ANOVA and Duncan posthoc test.

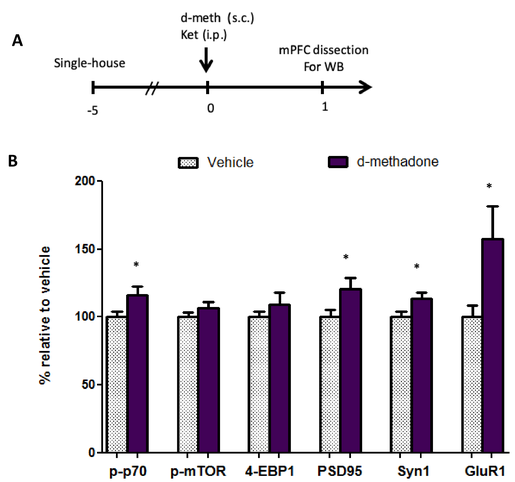


Fig 4. Influence of d-methadone on mTORC1 signaling and synaptic proteins.
(A) Rats were administered d-methadone and levels of mTORC1 signaling proteins and synaptic proteins were examined in the (B) PFC and (C) hippocampus.
Levels of phospho proteins were normalized to total proteins and levels of synaptic proteins were normalized the GAPDH. Results are the mean ± S.E.M., n = 10-12/group. P < 0.05 compared to vehicle (Student's t-test).

Clinical Studies

We then investigated the safety, tolerability and pharmacokinetic (PK) profile of d-methadone in healthy opioid-naïve volunteers in two Phase 1, double-blind, randomized, placebo-controlled, single and multiple ascending dose (SAD and MAD) studies.

d-Methadone exhibited linear PK with dose proportionality for most single dose and multiple dose parameters. Single doses up to 150 mg and daily doses up to 75 mg for 10 days were well tolerated with mostly mild treatment emergent adverse events and no severe or serious adverse events. At the tested doses, d-methadone did not cause dissociative or psychotomimetic adverse events, no clinically relevant opioid effects and no signs or symptoms of withdrawal upon abrupt discontinuation.

Single Ascending Dose (SAD) Study Design

Parallel group, double-blind, placebo controlled

Objectives

- Establish PK, PD and safety of single dose administration

Treatment Administration

- Cohorts 5, 20, 60, 100, 150, 200 mg and N = 42

Study Conclusions

- MTD = 150 mg (single dose)
- PK demonstrated linear proportionality of C_{max} and $AUC_{0\text{-}inf}$ vs. dose
- No clinically significant opioid effects of dextromethadone up to 150 mg

Multiple Ascending Dose (MAD) Study Design

Parallel group, double-blind, placebo controlled

Objectives

- Establish PK, PD and safety of once daily, 10 day administration

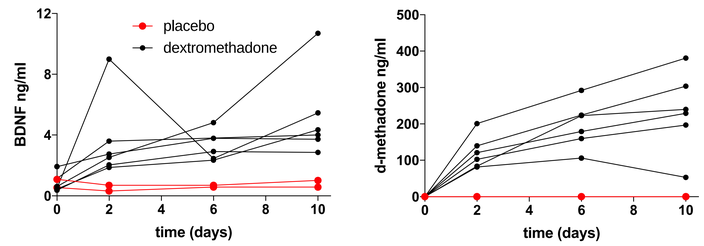
Treatment Administration

- Cohorts 25, 50, 75 mg and N = 24

Study Conclusions

- Doses up to 75mg per day well tolerated
- Dose proportionality was demonstrated for the single-dose parameters Cmax and AUCtau on Day 1 and for the steady state parameters Cmax, AUCtau, and Css on Day 10

Brain derived neurotrophic factor (BDNF) plasma levels from the 25 mg cohort of the MAD study were tested before any treatment and 4 hours after administration of d-methadone 25 mg (six patients) or placebo (two patients) on days 2, 6 and 10. In the d-methadone treatment group, 6 of 6 subjects showed an increase in BDNF levels post d-methadone treatment compared to pre-treatment levels, with post-treatment day 10 BDNF plasma levels ranging from twice to 17 times the pre-treatment BDNF levels. By contrast, in the two placebo subjects, the BDNF plasma levels remained unchanged. Plasma BDNF levels measured at day 2 and day 10 were significantly correlated to the plasma levels of d-methadone when placebo subjects are included in the analysis.



Treatment Arm	Average Plasma BDNF ng/ml (±SD)	
	Pre-treatment	Post-treatment
d-Methadone	0.84 (0.60)	5.84 (2.83)
Placebo	0.81 (0.38)	0.79 (0.30)

p=0.028 at day 2, p=0.043 at day 6, and p=0.028 at day 10, all vs BDNF plasma levels before treatment.

Future Directions

We are currently conducting a phase 2a, multicenter, randomized, double-blind, placebo controlled, 3-arm study to assess the safety, tolerability, PK profile, and antidepressant effect of 7-day dosing with d-methadone 25 mg QD and 50 mg QD as adjunctive therapy in the treatment of patients diagnosed with major depressive disorder, who have not responded to 1 to 3 courses of treatment with an antidepressant medication in the presenting episode. The findings of this study will be instrumental to determine the next step in the development of d-methadone

In summary, the evidence gathered so far supports the development of d-methadone in depression and other CNS conditions for which NMDAR antagonism could be an effective mechanism of action for a potential treatment.