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| Experiment | Species | Experimental groups (all naïve at start) | Timeline | Statistics |
| Blood brain barrier penetration | Mouse: C57/BL6Rat: Sprague Dawley | 1. JNJ-63533054 (10 mg/kg) | 0.25, 0.5, 1,2, 4, 8 h: samples collected in mouse0.5, 2, 6 h: samples collected in rat | NA |
| c-Fos  | C57/BL6 | 1. 10 mg/kg JNJ-635330542. 30 mg/kg JNJ-635330543. Vehicle4. 2 mg/kg Amphetamine (c-fos positive control) | 1 h: Samples collected | Medial habenula: No effect of treatment on c-Fos+ cell number (F(3, 12)=1.589, p=0.24)Dorsal striatum:Significant effect of amphetamine treatment only (F(3, 12)=41.11, p<0.0001) |
| Microdialysis | Sprague Dawley | PFC:1. JNJ-63533054 (10 mg/kg) + Desipramine (10 mg/kg)2. Vehicle + Desipramine (10 mg/kg)NAC:3. JNJ-63533054 (10 mg/kg) + Amphetamine4. Vehicle + Desipramine (10 mg/kg) | PFC:0 h: Baseline started2 h: Experimental treatment injected 6 h: Desipramine injected7.5 h: Collection terminated NAC:0 h: Baseline started2 h: Experimental treatment injected2.5 h: Amphetamine injected8 h: Collection terminated | PFC:No effect of JNJ-63533054(F(12, 84)=0.27, p=0.99)NAC:No effect of JNJ-63533054 (F(16, 160)=1.10, p=0.36) |
| Naloxone conditioning | C57/BL6 | 1. Morphine (20 mg/kg) + JNJ-63533054 (10 mg/kg) + Naloxone (10 mg/kg)2. Morphine (20 mg/kg) + Vehicle + Naloxone (10 mg/kg)3. Morphine (20 mg/kg) + JNJ-63533054 (10 mg/kg) + Saline4. Morphine (20 mg/kg) + Vehicle + Saline | Day 1 afternoon: HabituationDay 1 evening: Morphine injectionDay 2 - 4 morning: Saline conditioningDay 2 – 4 afternoon: JNJ-63533054 + Naloxone conditioningDay 2 – 3 evening: Morphine injectionDay 5 afternoon: Test | No effect of JNJ-63533054 on withdrawal (H(1, N=24)=2.26, p=0.13) or conditioning (F(1, 44)=4.75, p>0.05 for post-hoc) |
| Marble burying | C57/BL6 | 1. JNJ-63533054 (10 mg/kg) + 10 mg/kg fluoxetine2. JNJ-63533054 (10 mg/kg) + 3 mg/kg fluoxetine 3. JNJ-63533054 (10 mg/kg) + saline4. Vehicle + 10 mg/kg fluoxetine5. Vehicle + 3 mg/kg fluoxetine 6. Vehicle + saline | 0 h: Co-injection of treatments0.5 h: Start of marble burying test1 h: End of test | Minor decrease in marbles buried (F(1, 54)=8.70, p=0.0047) |
| EPM | Sprague Dawley | 1. JNJ-63533054 (30 mg/kg)2. JNJ-63533054 (10 mg/kg)3. Vehicle4. Diazepam | 0 min: injection of treatments30 min: Start of EPM for 5 min | No effect of JNJ-63533054 (F(3, 36)=4.60, p>0.05 for post-hoc) |
| Urine sniffing | C57/BL6 | 1. JNJ-63533054 (10 mg/kg)2. Vehicle | 0 min: Habituation30 min: Injection60 min: Test started63 min: Test ended | Decreased urine sniffing (F(1, 10)=13.66, p=0.00018) |
| Tail suspension | C57/BL6 | 1. JNJ-63533054 (10 mg/kg) + 10 mg/kg imipramine2. JNJ-63533054 (10 mg/kg) + 3 mg/kg imipramine3. JNJ-63533054 (10 mg/kg) +saline4. Vehicle + 10 mg/kg imipramine5. Vehicle + 3 mg/kg imipramine6. Vehicle + saline | 0 min: Co-injection30 min: Test started36 min: Test ended | No effect of JNJ-63533054 (F(1, 66)=3.60, p=0.06) |
| Learned helpless | Wistar Kyoto | Reversal:1. JNJ-63533054 (10 mg/kg)2. VehiclePrevention:3. JNJ-63533054 (10 mg/kg)4. Vehicle | Reversal:Day 1: Inescapable shocksDay 2-3: ScreenDay 4 0 min: InjectionDay 4 15 min: Test startedDay 4 ~60 min: Test endedDay 5 -8: Repeat of Day 4Prevention:Day 1 0 min: InjectionDay 1 15 min: Inescapable shocks startedDay 2: Test | Reversal:No effect of JNJ-63533054 on test day (F(1, 20)=0.45 p=0.51) or extinction (F(5, 50)=0.89, p=0.49)Prevention:No effect of JNJ-63533054 (F(1, 22)=1.10, p=0.31) |

Supplemental table summarizing the experimental details.