Gestational Exposure to Oxycodone Affects Later Socio-communication Behaviors in Mice

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INTRODUCTION

- The use of opioids during pregnancy can lead to neonatal opioid withdrawal syndrome (NOWS).[1]
- Birth outcomes associated with opioid use disorder (OUD) include preterm birth, poor fetal growth, low birthweight, and possible birth defects.[2]
- Even if they do not demonstrate outright problems at birth, OUD newborns may be plagued with health consequences later on, otherwise considered Developmental Origin of Health and Disease (DOHaD) effects.[3]
- Oxycodone is one of the primary opioids overprescribed for pain, including in pregnant women.[4]

HYPOTHESIS

We hypothesize that as an opioid, oxycodone exposure impairs fetal brain development that will manifest as behavioral problems later on in life

METHODS

- **Gestational Exposure:** We exposed 12 females CF1 mice to 1.5mg/mL per oxycodone and 12 female CF1 mice to 0.9% saline. Doses were calculated as 5mg/kg of the mouse body weight.[5] Treatments were administered daily via intraperitoneal injections for two weeks prior to pairing and continued throughout gestation. Treatments ceased after each dam gave birth

- **Postnatal Assessments**: On postnatal days (PND) 1 through 5, 7, and 14, twenty-three dams and litters were assessed (13 saline, 10 oxycodone).
- Such measurements included dam weight, litter weight, number of pups, nest size, and the amount of time the dam spent in and out of nest in a five-minute interval.

- **Socio-communicative behaviors**: At PND21, all pups were weaned and one male and one female pup from each litter were randomly selected for further behavioral analysis (13 males, 13 females for saline and 10 males, 10 females for oxycodone). Socio-communicative behaviors of the test mice were measured in Crawley's three-chambered social test[6] (Figure 1a). Three trials were recorded. The apparatus was cleaned with 70% ethanol between trials. The first trial controlled for the mouse behavior and acclimation the testing apparatus for 5 minutes. The second trial measured test mouse behavior with one stranger mouse for 10 minutes. The third trial measured preferential behavior of the test mouse with the first stranger mouse and a second stranger mouse for 10 minutes. The social test apparatus was followed by a 5 minute recording in a soundproof Ultrasonic Vocalization(USV) chamber using MATLAB software (Figure 1b). MATLAB was also used to analyze number, duration, and frequency of recorded USVs.

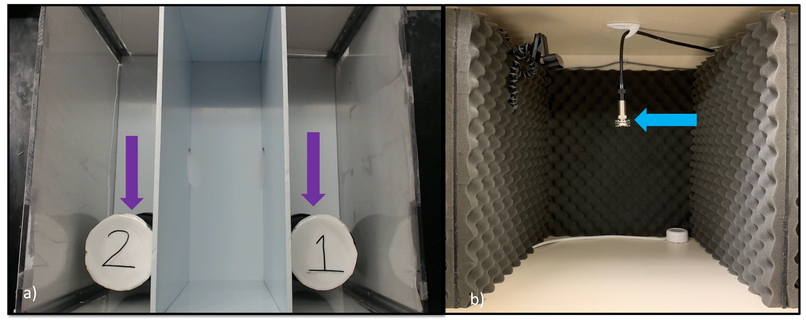

Figure 1 (a) Crawley's Three Chamber social apparatus: the test mouse was placed in the center of the apparatus. Stranger mice 1 and 2 were placed under their respective cups, indicated by the purple arrows. **(b) Sound recording chamber:** the test mouse was placed in the center of the soundproof chamber and recorded by the microphone, indicated by the blue arrow.

ANALYSIS

- **Social-communicative behaviors:** data in the three-chamber social testing, such as nose-to-nose interaction frequency and interaction duration with stranger mouse in Trials 2 and 3, and vocalization parameters were analyzed by using the PROG GLM procedure of SAS. Mean differences were determined using Fisher's Protected Least Significant Difference (LSD). All data are presented as actual means ($\bar{x}$). The error bars for all figures and reported data represent the standard error of the mean (SEM).

RESULTS

- **Social-communicative behaviors:** Female mice exposed to oxycodone during gestation showed a significant decrease in duration of time spent investigating Stranger 1 of trial 2 in comparison to female control mice (Figure 2). In trial 3, gestational exposure to oxycodone significantly impaired the sociability of the male mice compared to male control mice (Figure 3). Frequency (in kHz) of calls show a significant difference between males and females exposed to oxycodone in gestation compared to control mice (Figure 4). Saline mice vocalized under 20kHz, while oxycodone mice vocalized considerably above 20kHz.

Oxycodone Exposure Disrupts Social Behaviors in Female Mice during Trial 2

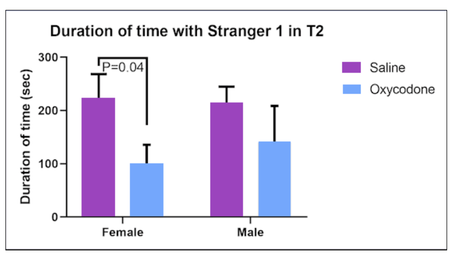

Figure 2: Duration of time spent investigating stranger 1 during Trial 2. Data are presented as means ± the standard errors. P = 0.04.

Oxycodone Exposure Disrupts Social Behaviors in Male Mice during Trial 3

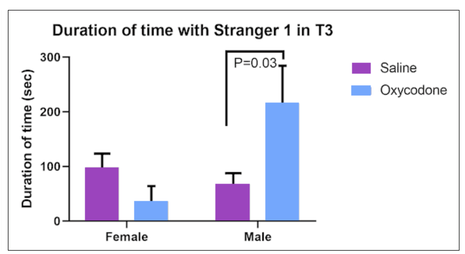

Figure 3: Duration of time spent investigating stranger 1 during Trial 3. Data are presented as means ± standard errors. P = 0.03.

Oxycodone Exposure Enhances Call Frequency Above 20kHz in Male and Female Mice

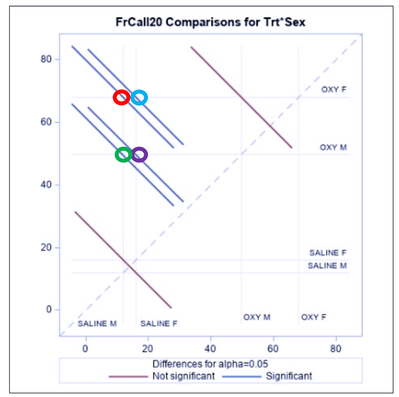

Figure 4: Frequency (in kHz) of calls comparing male and female gestational oxycodone exposure to the control. Significant differences are circled: Saline M vs Oxy F (red), Saline F vs Oxy F (blue), Saline M vs Oxy M (green), and Saline M vs Oxy F (purple). Oxycodone males and females exhibited vocalizations significantly above 20kHz. P < 0.05.

CONCLUSIONS & FUTURE AIMS

- Current results show gestational exposure to oxycodone impairs socio-communicative behaviors in male and female mice at PND21.
- Future studies may measure socio-communicative behaviors after gestationally exposed oxycodone mice have reach sexual maturity.
- As shown in the Davis et al. study[7] rats prenatally exposed to oxycodone show deficits in spatial learning and/or memory. Thus, gestationally exposed oxycodone mice may also show similar behavioral changes. To further understand the effects of this exposure, a Barnes maze may be used to measure spatial learning and memory in mice.[8]
- Future studies will perform histological analyses of the brain and gonads to correlate the effects of gestational oxycodone exposure and its effects on the central nervous system, reproductive system and other organs.

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