Analysis of maternal behavior in oxycodone-exposed rats transitioned to methadone or abstinence

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Introduction

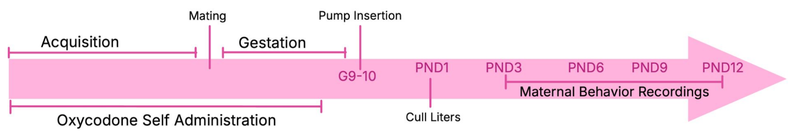
- Opioid use disorder **(OUD)** during pregnancy has increased substantially, raising concerns about maternal and infant health.
- Medications for opioid use disorder **(MOUD)**, including methadone and buprenorphine, stabilize opioid exposure and reduce withdrawal-related complications. However, their effects on maternal behavior remain unclear (Suarez et al., 2022; Lin et al., 2025).
- Maternal caregiving behaviors, including nursing, licking and grooming, and pup retrieval, are regulated by the Maternal Brain Network **(MBN)**, which includes the medial preoptic area (mPOA), ventral tegmental area (VTA), nucleus accumbens (NAc), and ventral pallidum (VP) (Lonstein et al., 2015; Swain et al., 2019).
- Oxytocin, dopamine, and endogenous opioids regulate maternal motivation and bonding. Dopamine signaling between the VTA and NAc promotes maternal care by rewarding interactions with offspring (Numan & Young, 2016; Swain et al., 2019).
- Opioid exposure during pregnancy may disrupt the MBN, altering nursing, pup-directed behaviors, and maternal responsiveness, potentially affecting mother–infant bonding (Wallin et al., 2021).
- Although methadone maintenance reduces withdrawal-related risks, its effects on postpartum maternal care compared with forced opioid withdrawal remain poorly understood (Wallin et al., 2021).

Aim: Investigate how gestational methadone maintenance versus forced opioid withdrawal affects maternal caregiving behaviors

Methods

Gestational MOUD exposure via osmotic minipumps





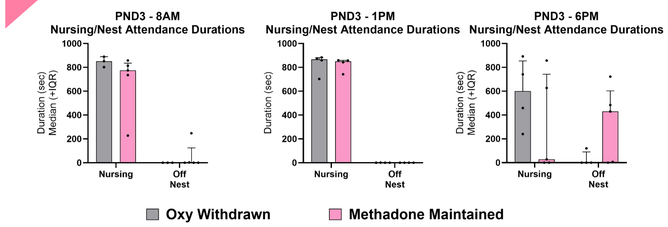
- Adult female Sprague Dawley rats (n-14) self-administer intravenous oxycodone (0.1 mg/kg/infusion) for 6 hours daily over ~2.5 weeks before mating.
- Gestational day (GD) 9 or 10, rats received osmotic minipumps delivering methadone (10 mg/kg/day; n = 7) or saline (n = 7).
- Methadone administration modeled opioid maintenance treatment, while saline administration modeled forced opioid withdrawal following oxycodone exposure.

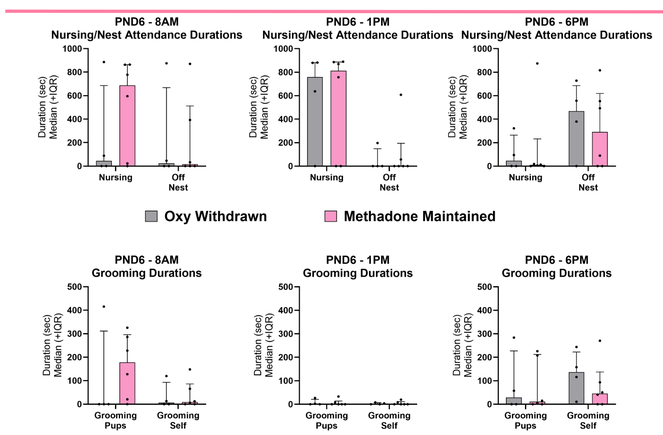
Maternal Behavior Video Analysis

- Home-cage maternal behavior recordings were collected on postnatal days **(PND)** 3, 6, 9, and 12.
- Videos were scored using Behavioral Observation Research Interactive Software **(BORIS)** to characterize maternal caregiving behaviors, including: Nursing posture, Licking and grooming of pups, Nest building, Pup retrieval, and Time spent off the nest
- Behavioral definitions were standardized before coding, and scoring reliability was assessed using a subset of recordings.
- Maternal behavior was evaluated across treatment groups and postnatal days to characterize differences in caregiving patterns throughout early postpartum development.

Results

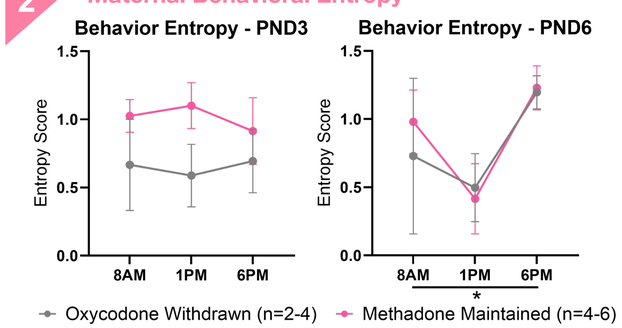
1 Maternal Behavior Durations





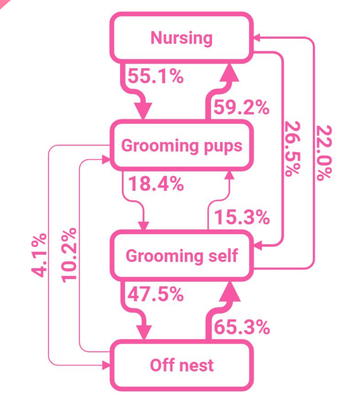
METH (pink) and VEH (gray) dams exhibited differences in maternal and non-maternal behavioral durations across observation times (8 AM, 1 PM, and 6 PM), but no statistically significant treatment effects were observed (all $p > 0.05$).

2 Maternal Behavioral Entropy

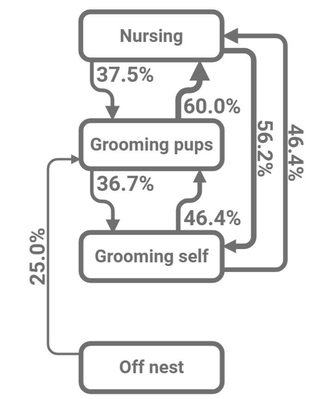


On PND3, METH dams exhibited higher behavioral entropy than VEH dams across all observation timepoints. The treatment-group difference was not statistically significant ($p = .0603$). On PND6, entropy scores were more similar between groups, but a significant effect of observation time was observed ($p = .0291$), with lower entropy at 1 PM and higher entropy at 6 PM.

3 Maternal Behavior Transitions



Methadone Maintained - PND3



Oxycodone Withdrawn - PND3

Methadone Maintained - PND6

Oxycodone Withdrawn - PND6

On PND3, METH dams exhibited more frequent transitions between behaviors, including non-maternal behaviors (self-grooming and off-nest activity). By PND6, behavioral transition patterns appeared more similar between treatment groups.

Conclusions

- No statistically significant differences in individual maternal behavioral durations were observed between METH and VEH dams on PND3 or PND6 (all p> .05).
- On PND3, METH dams exhibited more frequent behavioral transitions and higher entropy, suggesting a more disrupted maternal behavioral profile compared with VEH dams.
- By PND6, behavioral patterns became more similar between treatment groups, but entropy varied significantly across observation timepoints ($p = .0291$), potentially reflecting daily changes in maternal and nonmaternal activities.
- All seven METH dams successfully delivered litters, whereas only four of seven VEH dams gave birth, suggesting a possible association between forced opioid withdrawal and unsuccessful pregnancy outcomes.
- Future studies should investigate larger sample sizes, additional postnatal days, and potential neurobiological mechanisms, including Maternal Brain Network activity and HPA-axis regulation.

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