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## Abstract

Corticosterone (CORT) is a stress hormone that has been known to be responsible for balancing and controlling psychosocial and physical stressors during certain developmental stages. Within the neonatal time period (postnatal days 1-21), there are unique stressors, such as isolation, where the role of CORT could be essential. However, the hormone has not yet been assessed when maternal bonding and attachment takes place between the dam and her pups. Two behavioral paradigms were conducted to examine the role of CORT on mother-infant interaction: 1) Distress calls during isolation 2) Duration of contact with the anesthetized dam. CORT was manipulated with an injection of CORT or a CORT inhibitor called Metyrapone (Met). Metyrapone showed a trend for decreasing distress calls and decreasing amount of time spent with the anesthetized dam. In addition, CORT injections did not affect distress calls, but it nonsignificantly decreased the amount of time spent with the anesthetized dam. These preliminary results suggest that social bonding may be partially mediated by corticosterone.

## Methods

### Isolation Calls

The isolation testing chamber was located in a testing room separate from the housing room. It contained a 500 mL glass beaker with an ultrasonic microphone suspended approximately 12 cm above the base of the beaker. Using a high frequency bat detector, Pettersson D230 ultrasound detector (Uppsala, Sweden) that digitally records at 196 kHz. USVs were recorded and then analyzed offline via sonogram (Avisoft Bioacoustics).

On postnatal day nine, rat pups were habituated to the testing chamber for 1 minute. On postnatal day ten, the pups were removed from the colony room and placed individually in the isolation apparatus for 2 minutes while USVs are recorded. No other animals were present in the testing room during the session. Animals were transported back to the colony room after testing and were placed in a holding chamber until all testing is finished. All animals were returned to their home cage and dam upon testing completion. They were tested during the light cycle of a 12:12 light/dark cycle (lights on at 07:00). Data were scored manually offline for total number of 40 kHz isolation distress vocalizations.

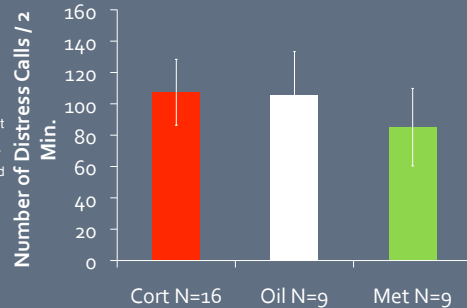
### Anesthetized Dam Approach

On postnatal day 12, all pups were subjected to 3 hours of maternal deprivation. After 3 hours, pups were tested for their approach latency and interaction with their anesthetized dam. The dam will be anesthetized with an i.p. injection of 50 mg/kg of sodium pentobarbital approximately 20 minutes prior to pup exposure. The dam was arranged such that she was lying on her ventrum and the perimeter of her body was wrapped with a mesh sock and gauze to prevent pups from attaching to the nipples. The pup was then introduced to the testing chamber at the "start" end. For a 5 minute time period, frequency and duration of physical contacts the pup initiates with the dam was recorded. Any physical contact between the pup and the dam, including her tail, was recorded. The pup was returned to the home cage at the end of the 5 minute session.

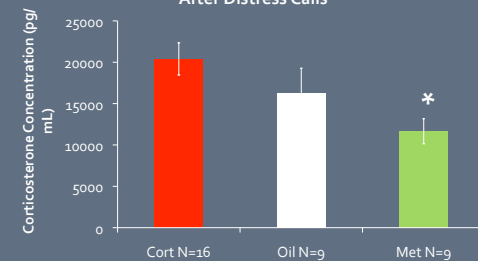
### Hormone Assay

After animals are tested in each paradigm, hormone samples were immediately assessed via decapitation. Blood was collected in centrifuge tubes, allowed to clot and centrifuged for 15 min. to separate the serum. Serum was stored at -20 C until analysis by commercially available EIA kit.

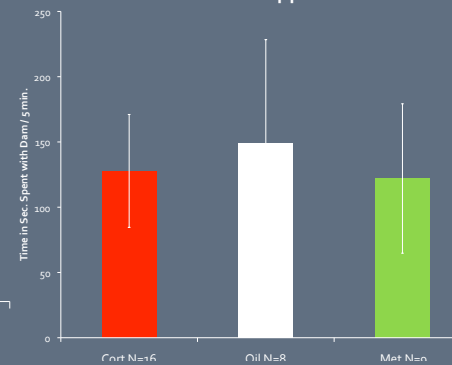
## Distress Calls



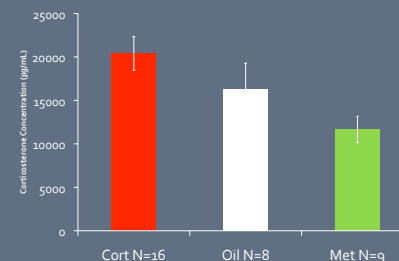
## Corticosterone Concentration Immediately After Distress Calls



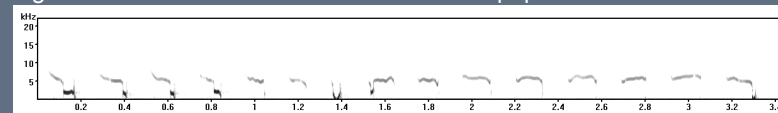
## Anesthetized Dam Approach Task



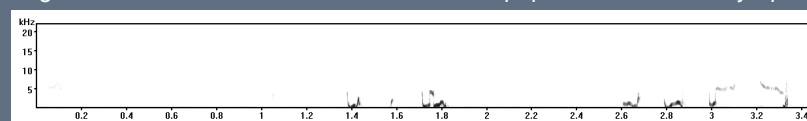
## Corticosterone after Anesthetized Dam Task



## Sonogram of Ultrasonic Vocalizations from male pup administered Corticosterone:



## Sonogram of Ultrasonic Vocalizations from male pup administered Metyrapone:



## Results

Metyrapone significantly reduced CORT concentrations compared to animals receiving CORT injections after distress calls.

MET lowered the number of distress calls.

CORT injections did not significantly affect distress calls nor amount of time spent with dam.

Future studies should examine more social behaviors such as alternative measures of attachment.

## Acknowledgements

Amanda Stewart  
David Mankin  
Emily Webber  
Molly Henry  
Hope for Depression Research Foundation

## References

- Harvey, A.T. & Hennessy, (1995). Corticotrophin-releasing factor modulation of the ultrasonic vocalization rate of isolated rat pups. *Developmental Brain Research*, 87, 125-134.
- Hennessy, M.B., O'Neill, D.R., Becker, L.A. et al. (1992). Effects of centrally administered corticotrophin-releasing factor (CRF) and  $\alpha$ -helical CRF on the vocalizations of isolated guinea pig pups. *Pharmacology Biochemistry and Behavior*, 43, 37-43.
- Smotherman, W.P. (1983). Mother-infant interaction and the modulation of pituitary-adrenal activity in rat pups after early stimulation. *Developmental Psychobiology*, 16, 169-176.
- Takahashi, L.K. (1994). Organizing action of corticosterone on the development of behavioral inhibition in the preweaning rat. *Developmental Brain Research*, 81, 121-127.
- Takahashi, L.K., Turner, J.G., & Kalin, N.H. (1991). Development of stress-induced responses in preweaning rats. *Developmental Psychobiology*, 24, 341-360.