Hormonal contraceptives alter amphetamine place preference and dopamine activity in the intact female rat.

Emily N. Hilz, Marcelle E. Olvera, Hongjoo J. Lee

The University of Texas at Austin, Dept. of Psychology, Austin, TX

The University of Texas at Austin
Waggoner Center for Alcohol & Addiction Research

INTRODUCTION

Hormonal contraceptives (HCs) are commonly used among women in the United States and act in part by reducing the release of ovarian hormones[1,2].

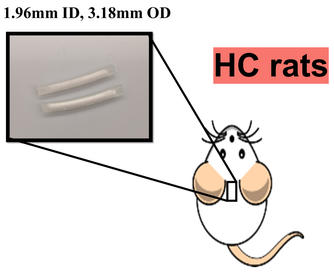
Stages of the rat estrous cycle associated with elevated ovarian hormones (i.e., estradiol and progesterone) increase drug-addictive behaviors in female rats[3,4].

Ovarian hormones enhance dopamine (DA) transmission[5], which is thought to contribute to motivation for rewarding stimuli such as that of drugs of abuse[6].

The following experiment characterizes amphetamine (AMP) place preference and dopamine (DA) activity in naturally cycling or HC-implanted female rats. Rats were conditioned and tested for AMP-preference with either an HC-implant or during estrous cycle stages associated with opposing ovarian hormone levels. Serum hormones were directly examined for any influence on AMP-preference and DA activity.

EXPERIMENTAL GROUPS

Subset of rats implanted with subcutaneous HC capsule.



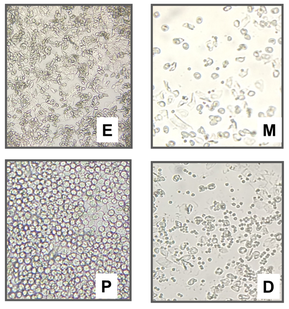
A subset of rats (n = 10) implanted in the scapular region with 2 capsules containing the progestin HC levonorgestrel (LNG, ~28mg each).

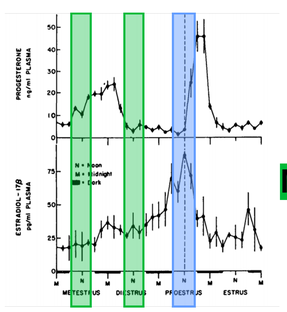
LNG binds to progesterone and androgen receptors[7].

- Delays or blunts LH and FSH peaks; impairs follicular development and ovulation[8].

Rats were anovulatory and showed persistent diestrus throughout procedures (~1 month).

Cycling rats were grouped in opposing hormonal states based on the estrous cycle.





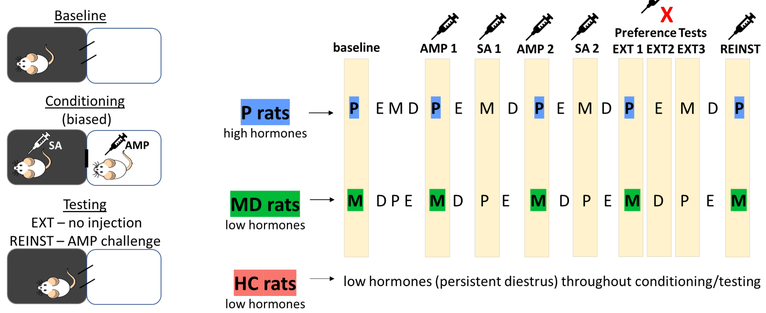
P rats
high hormones

MD rats
low hormones

Left. Cytology of cells collected from vaginal epithelium corresponding to estrous cycle stages. P = proestrus, M/D = metestrus/diestrus, E = estrus. **Right.** Estradiol and progesterone serum hormone levels associated with estrous cycle stages / experimental groups P (n = 10) and MD (n = 13).

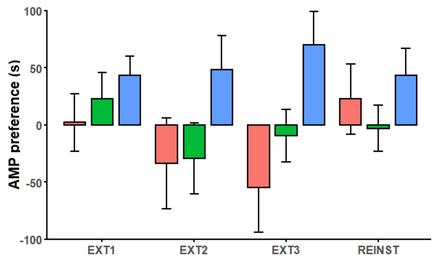
AMPHETAMINE PLACE PREFERENCE

Experimental timeline



P-rats have higher overall preference for AMP-associated context.

HC rats (n = 10) MD rats (n = 13) P rats (n = 10)

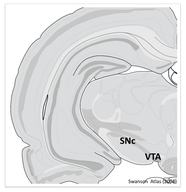


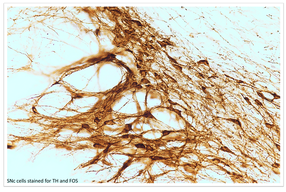
Time spent in the AMP-associated context (difference from baseline) +/- SEM.

P-rats spent significantly more time in the AMP-associated context throughout the 3 preference/extinction tests (EXT 1-3) compared to MD and HC rats, $F(2,86) = 5.49$, $p < .01$.

Groups did not significantly differ at reinstatement (REINST).

DOPAMINE ACTIVATION





Left. Brains collected 90 min. after REINST were immunohistochemically processed with tyrosine hydroxylase (TH, precursor to DA) and FOS.

- Regions included substantia nigra (SNc) and ventral tegmental area (VTA) using Swanson's Atlas (2004).

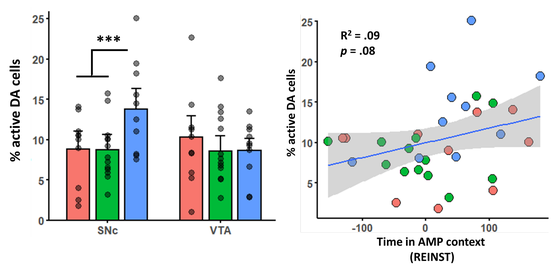
Right. Example image of TH+FOS labelling in SNc.

P-rats have higher percentage of active DA cells in SNc; DA activity non-significantly predicts AMP-preference at REINST.

HC rats (n = 10) MD rats (n = 13) P rats (n = 10)

Left. Percent total TH cells co-labelled with FOS (activated DA cells) +/- SEM. P-rats had significantly higher percent of TH+FOS cells compared to MD and HC rats in SNc but not VTA ($p < .001$).

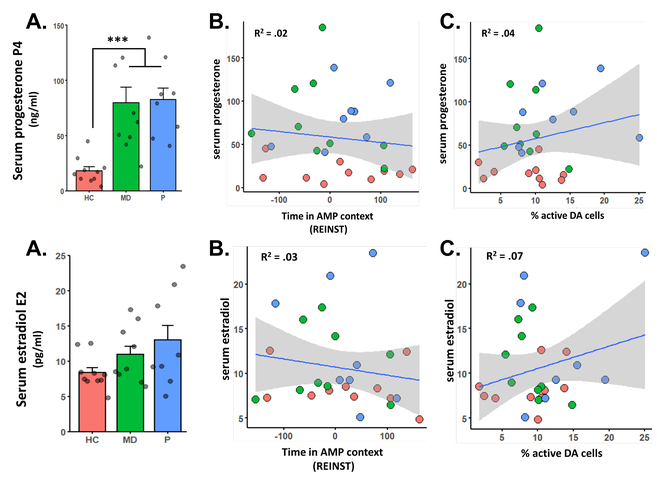
Right. Percentage of active SNc DA cells non-significantly predicts time spent in the AMP-associated context using linear regression ($\beta = 5.023$, $p = .08$).



SERUM ESTRADIOL AND PROGESTERONE

HC rats had lower P4 but not E2 compared to MD and HC rats; ovarian hormones did not predict AMP-preference or DA activity.

HC rats (n = 10) MD rats (n = 13) P rats (n = 10)



Top row. Serum progesterone (P4) was lower in HC rats compared to MD and P rats ($p < .001$; **A**). P4 level did not predict AMP-preference (**B**) or DA activity (**C**). **Bottom row.** Serum estradiol (E2) did not significantly differ between groups (**A**). E2 level did not predict AMP-preference (**B**) or DA activity (**C**).

DISCUSSION

This experiment is the first to show estrous cycle effects in AMP place preference; rats conditioned and tested when ovarian hormones are high (P rats) show increased preference for AMP-associated context.

Rats with HC implants, which reduce available ovarian hormones and suppress ovulation, showed low AMP-preference.

Results were reflected in pattern of DA activation in SNc: P-rats had a higher percentage of active DA cells after REINST and DA activity was a non-significant predictor of AMP-preference.

Although estrous cycle stage influenced AMP-preference and DA activity, serum hormones did not predict AMP-preference or DA activity.

- This may be due to timing of serum collection (~90 min after REINST).

REFERENCES

[1]Rivera et al., 1999
[2]Graham & Milad, 2013
[3]Carroll & Anker, 2010
[4]Becker & Hu, 2008
[5]Becker, 1990
[6]Di Chiara & Bassareo, 2007
[7]Lemus et al., 1992
[8]Gemzell-Danielsson et al., 2013

Bruce/Jones Fellowship for Addiction Biology, WCAAR – Emily N Hilz