Behavioural Brain Research 395 (2020) 112843

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

# Sex differences in impulsivity in adult rats are mediated by organizational actions of neonatal gonadal hormones and not by hormones acting at puberty or in adulthood

Jeffrey S. Darling[a,b,*], Daniel W. Bayless[c,d], Lauren R. Dartez[a,b], Joshua J. Taylor[a,b], Arjun Mehrotra[a,b], William L. Smith[a,b], Jill M. Daniel[a,b,c]

[a] Neuroscience Program, United States
[b] Tulane Brain Institute, United States
[c] Psychology Department, Tulane University, New Orleans, LA, 70118, United States
[d] Stanford University School of Medicine, Stanford, CA, 94305 United States

ARTICLE INFO

*Keywords:*
sex difference
gonadal hormone
inhibitory control
impulsivity
impulsive action
impulsive choice

ABSTRACT

Males as compared to females display increased impulsivity and inefficient inhibitory control and are more frequently diagnosed with disorders characterized by impulsivity. We previously demonstrated male rats make more impulsive action responses (i.e. premature responding) than females on the 5-choice serial reaction time task (5-CSRTT). Furthermore, pre-pubertal male rats make more impulsive choice responses (i.e. choosing an immediate small reward over a delayed larger reward) than females on a delayed-based reward T-maze task. The goal of the current work was to determine if gonadal hormones impact sex differences in impulsivity in adult rats. In an initial experiment, male and female rats underwent sham surgeries or were gonadectomized either pre-pubertally or during adulthood and tested on the 5-CSRTT in adulthood. Males displayed more impulsive action responses than females regardless of hormone status. In a second experiment, females received testosterone or vehicle injections on postnatal days 1 and 2. Males received vehicle injections. All rats were gonadectomized prior to puberty and tested on the 5-CSRTT in adulthood. Females treated neonatally with testosterone and control males made more impulsive action responses than control females. In another set of experiments, manipulation of gonadal hormones led to no differences in performance on the delayed-based reward T-maze task in males and females. Results indicate that no sex difference is apparent in impulsive choice on a delayed-base reward task in adult rats. They also reveal that adult sex differences on a task of impulsive action is mediated by organizational effects of gonadal hormones acting during the neonatal period and not impacted by hormones acting during puberty or adulthood.

## 1. Introduction

Males as compared to females display increased impulsivity and inefficient inhibitory control across the lifespan (Bayless and Daniel, 2012; Bayless et al., 2013; Hyten et al., 1994; Whelan et al., 2012). Impulsivity can be defined as actions without proper forethought or deliberation and is categorized as either an impulsive action or impulsive choice (Evenden, 1999; Weafer and de Wit, 2014). Impulsive actions constitute a lack of behavioral control resulting in an inability to suppress premature or inappropriate actions and can be measured in both humans and rodents utilizing stop-signal and serial reaction time tasks that require subjects to suppress a response until properly cued (Eagle and Baunez, 2010). Impulsive choice represents an inability to properly deliberate over alternative options and is often tested via delayed-reward tasks (Dalley et al., 2008). Results of research in our lab reveal male rats as compared to female rats to exhibit greater levels of impulsive action in adulthood (Bayless and Daniel, 2012) and impulsive choice prior to puberty (Bayless et al., 2013).

Sex differences in brain morphology and behavior have been determined to be heavily influenced by gonadal hormone exposure, acting early during development through old age. It is well established that this hormonal influence first acts early during prenatal and neonatal periods of development to sexually differentiate brain and behavior (for review, see Wallen and Baum, 2002). Phoenix et al. (1959) first showed prenatal testosterone exposure to masculinize and defeminize behaviors in rodents, launching an organizational-activational hypothesis behind

* Corresponding author. Present address: University of Texas at Austin, Austin, TX, 78712, United States.
*E-mail address:* jeffrey.darling@austin.utexas.edu (J.S. Darling).

https://doi.org/10.1016/j.bbr.2020.112843
Received 11 March 2020; Received in revised form 24 July 2020; Accepted 28 July 2020
Available online 02 August 2020
0166-4328/ 

sexual differentiation. Exposure to steroid hormones during the perinatal window leads to permanent brain organizing effects from activation of androgen and estrogen receptors that can enhance male typical behaviors and suppress female typical behaviors later on (McCarthy, 2008).

Puberty has been determined as an additional period of organization of brain and behavior as a result of changes in levels of gonadal hormones (Schulz et al., 2009). Pubertal onset is marked by an increase in pulsatile release of gonadatrophin-releasing hormone (GnRH) which in turn leads to secretion of luteinizing hormone (LH) and follicle-stimulating hormone (FSH) from the pituitary (Watanabe and Terasawa, 1989). These hormones in turn lead to increased stimulation of the gonads to produce a surge in gonadal sex hormones. This gonadal steroid hormone surge has been directly linked to permanent organizational sex-specific changes in brain morphology including cell death and cell phenotype (Forger, 2006), levels of markers of white matter (Darling and Daniel, 2019) as well as adult behavior (Schulz et al., 2004, 2006).

In addition to organizational effects, sex differences in brain and behavior can be mediated by activational hormone effects. Activational effects are those exerted by circulating hormones in adult organisms, are transient in nature, and exist only in the presence of the hormones (Adkins-Regan and Leung, 2006). Activational effects of hormones are thought to result from actions on existing neural circuits, many of which were likely organized by hormones acting neonatally or at puberty. Adult sex behavior in rats, which can be eliminated and reinstated by the respective removal or administration of gonadal hormones, is a classic example of activational actions of gonadal hormones (Takahashi, 1990).

The goal of the present study is to test the hypothesis that gonadal hormone exposure mediates adult sex differences in impulsivity. We explored the ability of gonadal hormones to exert organizational effects either during the neonatal or pubertal periods or activational effects during adulthood to impact adult performance of male and female rats on measures of impulsive action and impulsive choice. To measure impulsive action behavior in adult rats we utilized the five-choice serial reaction time task (5-CSRTT) (Robbins, 2002). The task takes place in an operant chamber with five holes (apertures) in the rear of the chamber. A rat (or mouse, Sanchez-Roige et al., 2012) is trained to make nose pokes in a lit hole to receive a food reward. Impulsive action is measured by premature nose pokes (responses made after initializing a new trial but made before stimulus direction) (Bayless and Daniel, 2012; Diergaarde et al., 2008). Previous work in our lab has shown adult male rats to make more impulsive premature nose pokes than females on the 5-CSRRT (Bayless and Daniel, 2012).

To measure impulsive choice, we utilized a delay-based reward T-maze task. In this task, rats must differentiate and choose between an immediate small food reward and a larger but delayed food reward. Impulsivity is measured as the choosing of low, immediately gratifying rewards versus large but delayed rewards. We have previously demonstrated that before puberty, male rats make more impulsive choices than female rats and that difference was due to organizational effects of gonadal hormones acting during the neonatal period (Bayless et al., 2013). Given that we see this pre-pubertal sex difference in impulsive choice, it is of interest to see if this differences carries on into adulthood and to what degree gonadal hormone exposure may contribute to maintenance or loss of such a difference. Such a change would suggest that an organizational effect seen in neonatal rats may later be impacted by further organization during puberty, shown to be another window where organization takes place in the presence of changes in gonadal hormone level (reviewed by Schulz et al., 2009).

In the current study, we conducted two sets of experiments to test our hypothesis that gonadal hormones impact adult levels of impulsivity. In the first set of experiments, we determined the ability of neonatal, pubertal, and adult gonadal hormones to impact the previously identified sex difference (Bayless and Daniel, 2012) in adult performance of impulsive action on the 5-CSRTT. In the second set of experiments, we determined if the sex difference in impulsive choice on the delay-based reward T-maze task we previously identified in pubertal rats (Bayless et al., 2013) would be present in adulthood and also if gonadal hormones, acting at puberty or in adulthood, impacts adult levels of impulsive choice.

## 2. Experimental Procedures

### 2.1. -Choice Serial Reaction Time Task (5-CSRTT)

The 5-CSRTT was used to measure impulsive action in Experiments 1A (Section 2.2.) and 1B (Section 2.3.).

#### 2.1.1. Apparatus

Animals were trained and tested in one of four separate 25 x 25 cm aluminum chambers (Lafayette Instrument Co., Lafayette, IN), each housed in sound attenuating cabinets. The rear wall of each chamber was convexly curved and contained five apertures, each 2.5 cm square, 4 cm deep, and set 2 cm above floor level. Each hole could be illuminated with a 3 W light bulb located at the rear of the hole and had an infrared photocell beam monitoring the entrance. Each chamber was illuminated by a house light. On the front wall, 25 cm from each nose-poke hole, there was a food magazine where 45 mg food pellets (Test Diet, Richmond, IN) could be automatically dispensed. Each animal received one session of training per day throughout the experiment. House lights were on unless stated otherwise.

#### 2.1.2. Behavioral Training

First, animals were successively shaped to retrieve a food reward from the food tray and to poke any of the holes to receive food rewards. Then, each animal was trained daily for 30 min on the 5-CSRTT by passing through several training stages of increasing difficulty. Each session was terminated after 100 trials had been completed or 30 min had expired. An animal was moved to the next training stage once it performed 100 trials at > 80 percent correct and < 20 percent omission for two consecutive days. Each rat was always trained in the same conditioning chamber. Females were always trained in the same two chambers while males were always trained at the same time as the females in the other two chambers. Animals were trained at approximately the same time of the light phase each day.

For the initial training stage, animals were placed in the chamber and could initiate the first trial by retrieving a single food pellet from the food tray. After a fixed 5 sec inter-trial interval (ITI), one of the five horizontal lights would illuminate for a maximum of 60 sec (cue duration) or until a response had been made. From the time the light first turned on, the animal had 60 sec (limited hold period) to respond by making a nose poke into the previously lit aperture. Correct responses were immediately rewarded with delivery of a food pellet into the food magazine, and retrieval of the food restarted the next trial after a 5-sec ITI. Several types of errors were recorded: i) Nose pokes during the ITI were recorded as premature responses, ii) Repeated nose pokes into the correct aperture were recorded as perseverative responses; iii) Responding into a non-lit aperture was recorded as an incorrect response; iv) Failure to respond within the limited hold period was recorded as an omission. All errors were punished by switching off the house light for a 5 sec time-out period, and no food was delivered. Responses to holes during this period would restart the time-out period.

For subsequent training stages all parameters remained the same, but the stimulus duration was successively decreased from 60 sec to 0.6 sec and the limited hold period was successively decreased from 60 sec to 5 sec. For the final baseline training stage, the cue duration was reduced to 0.5 sec. Training with this protocol continued until animals perform 100 trials at a baseline criterion of > 70 percent correct with < 20 percent omissions for five consecutive days.

*2.1.3. Behavioral Testing*

Following training, a series of manipulations to challenge performance, as described below, were introduced for one daily session each and when intact females were at the proestrous stage of the estrous cycle. Animals received three days of baseline training between each behavioral challenge.

*2.1.3.1. Baseline*. Light stimulus lasted for 0.5 sec. A 5 sec ITI was presented before onset of stimulus. Animal was given 5 sec to respond before an omission was counted. Each session consisted of 100 trials.

*2.1.3.2. Unpredictable Long Inter-Trial Interval Challenge*. As seen in previous results (Bayless and Daniel, 2012), this challenge is more likely to draw out impulsive responding than other challenges and was the measure of primary interest in the current work. Time before the onset of the light stimulus was pseudorandomly lengthened to 4.5, 5.5, 6.5, or 7.5 sec distributed across the 100 trials. This condition challenges impulsive action control because of the increase in time between trials and the decrease in predictability of the stimuli.

*2.1.3.3. Other Challenges*. Although our primary interest in the current study was to assess impulsive responding that would be most apparent under the *Unpredictable Long Inter-Trial Interval* condition, we did conduct other challenge conditions to assess impact of our manipulations on measures of attention. *Short Stimulus* challenges attentional performance by decreasing the duration of the stimuli. *Unpredictable Short Inter-Trial Interval* challenges attention by pseudorandomly shortening time between trials.

*2.1.4. Behavioral Measures*

Throughout testing, the following behavioral measures were recorded by automated computer software (ABET II, Lafayette Instruments) on a PC connected to conditioning chambers.

*2.1.4.1. Premature responses*. This measure is the primary measure of interest in the current study. The total number of trials in which a rat poked into an aperture during the ITI reflects deficits in inhibitory control processes of response preparation and is the primary measure of impulsive action.

*2.1.4.2. Perseverative responses*. The total number of additional nose pokes made into the apertures following either a correct or an incorrect response reflects deficits in inhibitory control processes of response control and is a secondary measure of impulsive action.

*2.1.4.3. Percent correct*. The total number of correct responses relative to the total number of trials completed indicates overall attentional performance.

*2.1.4.4. Percent omissions*. The percentage of trials in which a rat failed to respond during the limited hold period can reflect a failure to detect the stimulus due to inattentiveness or due to motivational and/or motor deficits.

*2.1.4.5. Speed*. Two measures of speed were collected. *Correct response latency* is the time between onset of stimulus and a correct nose poke. *Reward latency* is the time between a correct nose poke and retrieval of food from the magazine. Differences in response latency can indicate changes in decisional mechanisms, whereas differences in reward latency can indicate changes in motivational factors. If both measures are affected, motivational and/or motor functions could be affected (Muir et al., 1996; Robbins, 2002).

*2.2. Experiment 1A: The effect of gonadectomy prior to pubertal onset or in adulthood on adult levels of impulsive action*

*2.2.1. Subjects*

Pre-pubertal Long-Evans hooded rats (male n = 24, female n = 24, age approximately 21 days of age on arrival), were purchased via Harlan Sprague-Dawley, Inc. (Indianapolis, IN). Rats were pair housed in a temperature-controlled vivarium under a 12 -h light/dark cycle (lights on at 7:00am). Animals were weighed daily following behavioral training and food was provided in their home cages to maintain their weights at 85% of their free-feeding weights while allowing for growth of approximately 2% of their body weight each week. All procedures for this and subsequent experiments were in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals (1996) and approved by the Tulane University Institutional Animal Care and Use Committee.

*2.2.2. Gonadectomy*

Eight females and eight males were gonadectomized at 28 days of age and prior to puberty. An additional eight females and eight males were gonadectomized in adulthood, at 70 days of age. The remaining animals remained gonadally intact. For control purposes, half of the intact groups underwent sham surgery at 28 days of age and half at 70 days of age. Surgeries were performed while under anesthesia induced by injections of ketamine (100 mg/kg, ip; Bristole Laboratories; Syracuse, NY) and xylazine (7 mg/kg, ip; Miles Laboratories; Shawnee, KS). Females were ovariectomized or underwent sham ovariectomy. Males were castrated or underwent sham castration. At the time rats were killed, right uterine horns of females and ischiocavernosa muscle weight of males were extracted and weighed to confirm gonadectomy status.

*2.2.3. Behavior*

Beginning when rats were approximately 80 days of age, behavioral training and then testing was completed on the 5-CSRTT (~6 months of age) as described under Section 2.1.

*2.2.4. Vaginal Cytology*

To control for effects of fluctuating ovarian hormones on performance, testing took place when intact females were at the proestrous stage of the estrous cycle (when levels of estrogen are at their highest). Vaginal smears of females were collected by lavage each morning and analyzed daily beginning two weeks prior to behavioral testing. Males underwent sham smears during which a small amount of water was placed on the genitals using a medicine dropper. Behavioral challenge conditions were administered when a female was at the proestrous stage of the estrous cycle. Each male and gonadectomized female was yoked to and assigned to be tested on the same day as an intact female.

*2.2.5. Statistical Analyses*

*2.2.5.1. Baseline*. To assess the stability of baseline performance at the end of training, performances across the last five days of training were analyzed for all dependent variables using repeated measures ANOVAs with day as the within-subjects factor and sex and surgery as between-subjects factors. To assess possible changes in baseline performance during behavioral challenge testing, performances across the initial baseline data and baseline data collected the day before each behavioral challenge condition were analyzed for all dependent variables using overall repeated measures ANOVAs with day as the within-subjects factor and sex and surgery as the between-subjects factors.

*2.2.5.2. Unpredictable Long Inter-Trial Interval*. Each measure was analyzed using repeated measures ANOVA with ITI duration as the within-subject factor and sex and surgery as between-subject factors. Significant main or interactive effects were probed by Fisher's LSD post

hoc test as appropriate.

*2.2.5.3. Other Challenges*. Each measure for the *Short Stimulus* condition was averaged across trials and analyzed by two-way ANOVA with sex and surgery as between-subject factors. Each measure for the *Unpredictable Short Inter-Trial Interval* condition was analyzed using repeated measures ANOVA with ITI duration as the within-subject factor and sex and surgery as between-subject factors.

### *2.3. Experiment 1B: The effect of neonatal gonadal hormone exposure on adult levels of impulsive action*

#### *2.3.1. Subjects*

Male and female Long Evans rats (male n = 10, female n = 10) were purchased via Harlan Sprague-Dawley, Inc. (Indianapolis, IN) for breeding purposes. Females were allowed to deliver normally 22-23 days after conception. Litters were culled to 5 males and 5 females when possible to reduce variability in maternal care. Rats were weaned at 21 days of age and group housed by sex and treatment condition.

#### *2.3.2. Neonatal Manipulation*

On day of birth and 24 h later, female pups received injections (s.c.) of either 150 μg of testosterone propionate in 0.1 ml of sesame oil vehicle (n = 8) or oil vehicle (n = 8). Male pups received vehicle injections (s.c.) (n = 8). Testosterone dose was based on previous studies examining organizational effects of testosterone exposure (Bayless et al., 2013) and its activation of androgen receptors in the brain (Zhang et al., 2008) to influence behavior. Pups were given India Ink foot paw tattoos to label groups.

#### *2.3.3. Gonadectomy*

All male and female rats were gonadectomized at 28 days of age (immediately prior to onset of puberty). Results from Experiment 1A showed neither pubertal nor adult gonadal hormone exposure influences adult levels of impulsive action (Section 3.*1.2*). Therefore, rats were gonadectomized prior to puberty to eliminate irregularities neonatal-manipulated intact rats may exhibit in pubertal development.

#### *2.3.4. Behavior*

Beginning when rats were approximately 80 days of age, behavioral training and then testing was completed on the 5-CSRTT (~6 months age) as described under Section 2.*1.*

#### *2.3.5. Statistical Analyses*

Analyses were as described in Experiment 1 except that there was one between-factor (treatment).

### *2.4. Delay-Based Reward T-Maze Task*

The delay-based reward T-maze task was used to measure impulsive choice in Experiments 2A (Section 2.5) and 2B (Section 2.6).

#### *2.4.1. Apparatus*

Behavioral testing was conducted in a plastic T-maze (arms: 10 cm wide × 40 cm long × 20 cm high) with a black floor and clear walls. The start arm (north) led to two goal arms (east and west). A clear plastic 25-cm-high sliding door, placed 5 cm into the entrance of each goal arm, confined the rat into a goal arm upon entrance. A second clear plastic 25-cm-high sliding door, placed 5 cm from the end wall of each goal arm, controlled access to the food reward.

#### *2.4.2. Behavior*

*2.4.2.1. No-Delay Trials*. Animals were habituated to the maze for two 15-minute periods across two days during which they had free access to Froot Loops placed in goal arms. Following habituation, rats were trained to choose between a low-reward arm that contained one piece of Froot Loop and a high-reward arm that contained five pieces of Froot Loop. Rats were trained and tested in assigned male/female pairs in which rats alternated trials each session. The location of the high-reward arm was counterbalanced across pairs and treatment groups but always in the same location for any given rat. The maze was cleaned with ethanol between trials. Each session started with a forced trial into each of the low-reward and high-reward arms during which a black plastic sliding door blocked access to the opposite arm. The order of these forced trials alternated each session. Following the forced trials, rats were given five choice trials in which they were free to choose either the low-reward or high-reward arm. When a rat entered an arm, the first sliding door was lowered to confine the rat in the arm. The second sliding door was then lifted to provide immediate access to the food reward. Sessions of this No-Delay condition continued until rats were choosing the high-reward arm on at least 80% of trials for two consecutive sessions. All rats achieved criterion performance within 3 days of training.

*2.4.2.2. Delay Trials*. After all rats reached criterion on the No-Delay sessions, rats were given three 15-sec Delay sessions. The goal of the first session at each delay was to habituate and expose rats to delay conditions. Performance during the final two sessions was used for analyses. During these sessions, a 15-sec delay was imposed when rats entered the high-reward arm. The 15-sec delay on the high-reward arm was imposed during two forced trials and five choice trials. When a rat entered the high-reward arm, the first sliding door was lowered to confine the rat in the arm. The rat then had to wait 15 sec before the second sliding door was lifted to provide access to the larger food reward. When a rat entered the low-reward arm, the first sliding door was lowered to confine the rat in the arm and the second sliding door was lifted to provide immediate access to the smaller food reward. After three 15-sec Delay sessions, rats were given three 30-sec Delay Sessions and then three 45-sec Delay sessions using identical procedures except the delay duration on the high-reward arm was increased.

### *2.5. Experiment 2A: The effect of sex on adult rat levels of impulsive choice*

#### *2.5.1. Subjects*

Ten female and ten male Long-Evans hooded rats, approximately 70 days of age, were purchased from Harlan Sprague-Dawley. Rats were housed in same-sex pairs. Animals were weighed daily following behavioral testing and food was provided in their home cages to maintain their weights at 85% of their free-feeding weights while allowing for growth of approximately 2% of their body weight each week.

#### *2.5.2. Statistical Analyses*

The percentages of high-reward arm choices during the choice trials from the final two sessions of the No-Delay condition were averaged and analyzed using a one-way ANOVA with sex as the factor. Percentage of high-reward arm choices during the choice trials from the final two sessions of each Delay condition were averaged and analyzed using a repeated measures ANOVA with delay as the within-subjects factor and sex as the between-subjects factor. To examine female performance across the estrous cycle, the percentages of high-reward arm choices from each delay session were analyzed using a repeated measures ANOVA with delay session as the within-subjects factor and estrous cycle stage as the between-subjects factor.

### *2.6. Experiment 2B: The effect of gonadectomy in adulthood or prior to pubertal onset on adult levels of impulsive choice*

#### *2.6.1. Subjects*

Subjects were previously used in Experiment 1A (Section 2.*2.*). Following testing in Experiment 1A, rats began training and then testing (~7 months of age) on the Delay-Based Reward Task.

Figure 1.

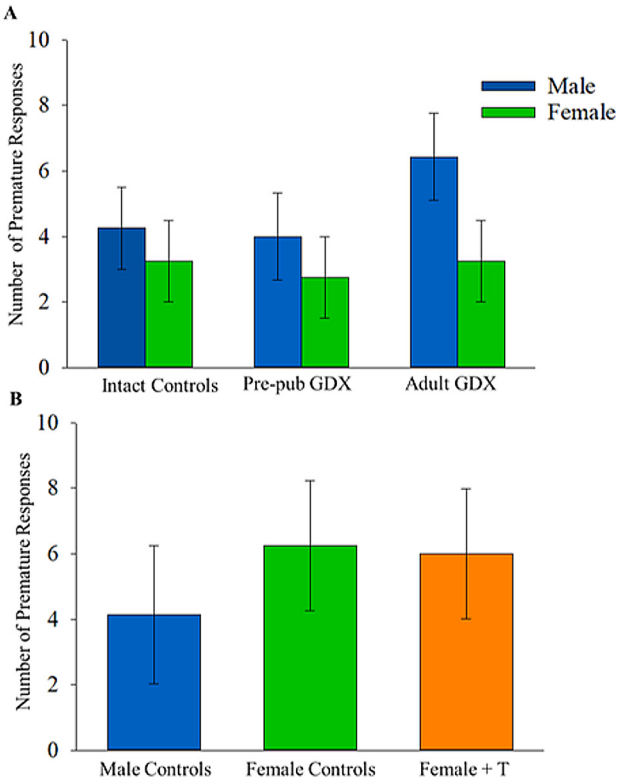


**Fig. 1.** Impact of sex and gonadal hormones on impulsive action responding on the 5-CSRTT under baseline conditions. (A) Mean total number of premature responses in adult male and female rats that were gonadally intact (Intact Controls, male n = 8, female n = 8), gonadectomized pre-pubertally (Pre-pub GDX, male n = 8, female n = 8), and gonadectomized in adulthood (Adult GDX, male n = 8, female n = 8). No main or interactive effects were revealed at baseline conditions. (B) Mean total number of premature responses in adult male (Male Controls, n = 8) and female rats (Female Controls, n = 8) treated with vehicle neonatally and adult female rats treated with testosterone propionate neonatally (Female + T, n = 8). No main or interactive effects were revealed at baseline conditions.

### 2.6.2. Statistical Analyses

Analyses for No Delay and Delay conditions were as described under Experiment 2A (Section 2.5.) except two between subjects factors, sex and surgery, were applied.

## 3. Results

### 3.1. Experiment 1A: The effect of gonadectomy prior to pubertal onset or in adulthood on adult levels of impulsive action

#### 3.1.1. Baseline Session

One male gonadectomized prior to puberty and one male gonadectomized in adulthood failed to reach criterion level performance during training and were excluded from all analyses. Other animals successfully acquired the task, as indicated by criterion performance, within 100 training sessions. There were no effects on any measure of sex or surgery during the last five days of training, indicating stable performance. No significant change in baseline performance occurred across baseline sessions conducted between behavioral challenge testing days. There was no main effect of sex or surgery or treatment on levels of impulsive action at Baseline condition (Fig. 1A) nor on two measures of speed, correct response latency and reward collection latency (Table 1), indicating that there was no sex difference in motor or sensory function, motivational factors, or the overall ability of the animals to perform the task (Robbins, 2002).

**Table 1**
Experiment 1A Speed Measures at Baseline and Under Long Inter-Trial Interval (ITI) Conditions.

| | Correct Response Latency | | Reward Collection Latency | |
|---|---|---|---|---|
| | Baseline (M ± SEM) | Long ITI (M ± SEM) | Baseline (M ± SEM) | Long ITI (M ± SEM) |
| Female Intact | 0.98 ± 0.04 | 0.92 ± 0.07 | 1.01 ± 0.04 | 1.22 ± 0.05 |
| Male Intact | 0.91 ± 0.04 | 0.93 ± 0.08 | 1.03 ± 0.04 | 1.03 ± 0.04 |
| Female Pre-pub GDX | 1.01 ± 0.04 | 1.03 ± 0.07 | 1.07 ± 0.04 | 1.12 ± 0.05 |
| Male Pre-pub GDX | 0.96 ± 0.05 | 1.06 ± 0.08 | 1.01 ± 0.03 | 1.13 ± 0.06 |
| Female Adult GDX | 0.97 ± 0.04 | 0.95 ± 0.07 | 1.11 ± 0.04 | 1.02 ± 0.05 |
| Male Adult GDX | 0.98 ± 0.05 | 0.94 ± 0.08 | 0.99 ± 0.03 | 1.03 ± 0.05 |

Pre-pub, Pre-pubertal; GDX, gonadectomy.

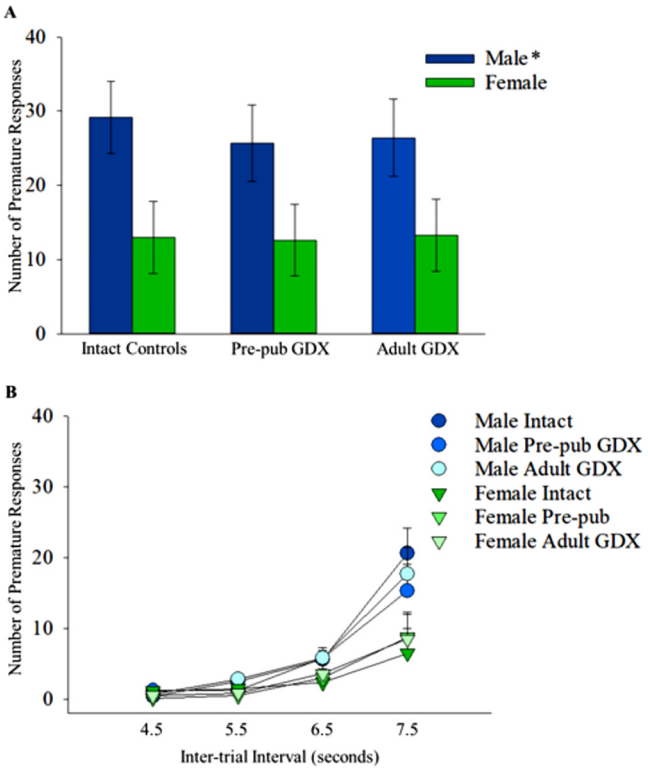


**Fig. 2.** Impact of sex and gonadal hormones acting pre-pubertally or in adulthood on impulsive action responding on the 5-CSRTT under long inter-trial interval conditions. Mean total number of premature responses in adult male and female rats that were gonadally intact (Intact Controls, male n = 8, female n = 8), gonadectomized pre-pubertally (Pre-pub GDX, male n = 8, female n = 8), and gonadectomized in adulthood (Adult GDX, male n = 8, female n = 8) (A) averaged over all inter-trial intervals or (B) across inter-trial intervals. There was a significant main effect of sex (* $p < 0.001$) and significant effect of inter-trial intervals duration ($p < 0.001$).

#### 3.1.2. Unpredictable Long Inter-Trial Interval

For premature responding, the primary measure of impulsivity, a significant main effect of sex was revealed $F(1,40) = 13.01$, $p = .001$), with males displaying increased premature responses as compared to females (see Fig. 2A). There was no main effect of surgery and no interaction between sex and surgery, indicating that neither pubertal nor adult gonadectomy impacted performance. There was a significant main effect of ITI duration ($F(3(120) = 60.28$, $p < .0001$) indicating that, as expected, animals displayed greater premature responding as ITI duration increased (see Fig. 2B). A significant interaction of sex and ITI duration was found ($F(3,120) =$, $p < .0001$) with males making significantly more premature responses than females at the longest ITI

of 7.5 sec ($p < .005$). No interactions were revealed between surgery and ITI duration nor sex and surgery and ITI.

On measures of speed (correct response latency, reward collection latency), there were no significant main or interactive effects (Table 1). On other measures (perseverative responses, percentage correct, percentage omissions), there were no significant main or interactive effects (data not shown).

#### 3.1.3. Other Challenges

There were no main or interactive effects on any measure during the *Short Stimulus* and *Unpredictable Short Inter-Trial Interval* challenge conditions (data not shown). These results are in contrast to our previous results in which females as compared to males displayed increased omissions under these challenge conditions, results suggestive of a sex difference in vigilance (Bayless and Daniel, 2012). Investigation of potential explanation of result discrepancy is ongoing.

### 3.2. Experiment 1B: The effect of neonatal gonadal hormone exposure on adult levels of impulsive action

#### 3.2.1. Baseline Sessions

Animals successfully acquired the task, as indicated by criterion performance, within 100 training sessions. Performance remained stable across the last five days of baseline training, and there were no significant treatment effects on any measure during the last five days of training. In addition, no significant change in baseline performance occurred across baseline sessions conducted between behavioral challenge testing days. There was no main effect of treatment on levels of impulsive action at Baseline condition (Fig. 1B) nor on the two measures of speed, correct response latency and reward collection latency (Table 2), indicating that there was no effect of treatments in motor function, sensory function, motivational factors, or the overall ability of the animals to perform the task (Robbins, 2002).

#### 3.2.2. Behavior Testing

*3.2.2.1. Unpredictable Long Inter-Trial Interval.* For premature responding, the primary measure of impulsivity, a significant main effect of treatment was revealed ($F(2,20) = 5.67$, $p = .011$) (see Fig. 3A). Posthoc analyses revealed that females given testosterone exhibited significantly increased premature responses ($p < .003$) and control males exhibited a trend towards increased premature responses ($p = .093$) as compared to control females. There was a significant main effect of ITI duration ($F(3,60) = 32.98$, $p < .0001$) indicating that as expected, animals displayed greater premature responding as ITI durations increased (see Fig. 3B). A significant interaction of treatment and ITI duration was found ($F(6,60) = 27.43$, $p < .0001$) with both control males ($p < .05$) and females receiving testosterone ($p < .001$) making significantly more premature responses than control females at the longest ITI of 7.5 sec.

On measures of speed (correct response latency, reward collection latency), there were no significant main or interactive effects (Table 2). On other measures (perseverative responses, percentage correct, percentage omissions), there were no significant main or interactive effects

**Table 2**
Experiment 1B Speed Measures at Baseline and Under Long Inter-Trial Interval (ITI) Conditions.

| | Correct Response Latency | | Reward Collection Latency | |
|---|---|---|---|---|
| | Baseline (M ± SEM) | Long ITI (M ± SEM) | Baseline (M ± SEM) | Long ITI (M ± SEM) |
| Female Control | 0.78 ± 0.04 | 0.89 ± 0.05 | 1.00 ± 0.04 | 1.12 ± 0.04 |
| Male Control | 0.82 ± 0.05 | 0.83 ± 0.06 | 1.08 ± 0.04 | 1.13 ± 0.04 |
| Female + T | 0.81 ± 0.04 | 0.88 ± 0.05 | 1.07 ± 0.04 | 1.02 ± 0.04 |

T, testosterone propionate.

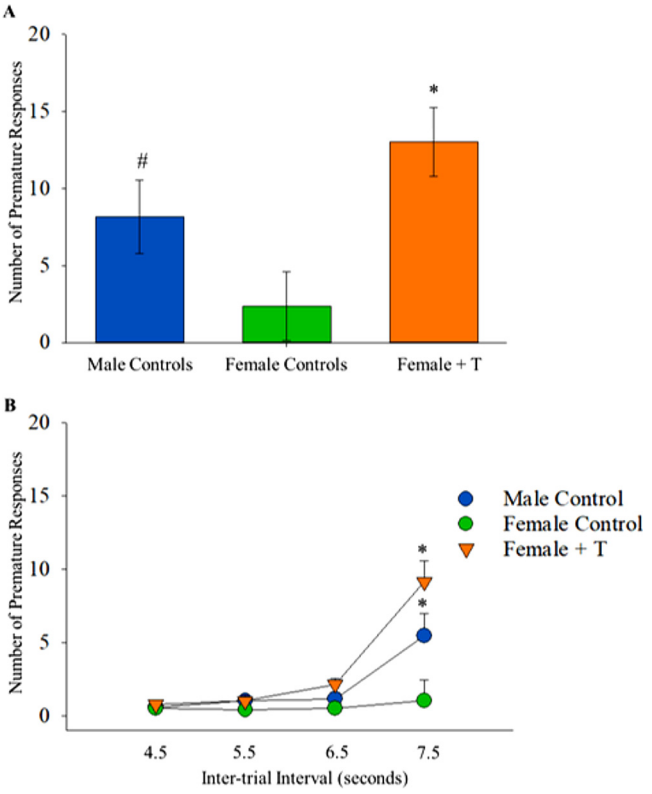


**Fig. 3.** Impact of sex and testosterone acting during the neonatal period on impulsive action responding on the 5-CSRTT under long inter-trial interval conditions. Total number of premature responses in adult male (Male Controls, n = 8) and female (Female Controls, n = 8) rats treated with vehicle neonatally and adult female rats treated with testosterone propionate neonatally (Female + T, n = 8) (A) averaged over all inter-trial intervals (Main effect of treatment, $p < 0.05$; * $p < 0.05$ and # $p = .093$ as compared to Female Controls); and (B) across inter-trial intervals (Main effect of inter-trial interval, $p < 0.001$, and interactive effect of treatment and inter-trial interval, $p < 0.001$; * = $p < 0.05$ as compared to Female Controls at the 7.5 second inter-trial interval).

(data not shown).

#### 3.2.3. Other Challenges

There were no main or interactive effects on any measure during the *Short Stimulus* and *Unpredictable Short Inter-Trial Interval* challenge conditions (data not shown).

### 3.3. Experiment 2A: The effect of sex on adult rat levels of impulsive choice

#### 3.3.1. Performance on the Delay-Based Reward Task

As illustrated in Fig. 4A, male and female adult rats displayed similar levels of impulsive choice behavior. Analysis of the percentages of high-reward arm choices from the final two sessions of the no delay condition revealed no significant main effect of sex, indicating that male and female rats equally preferred the larger reward over the smaller reward when rewards were immediately available. Analysis of the percentages of high-reward arm choices across delay condition revealed no significant main effect of sex, indicating that adult male and female rats did not differ in terms of number of high-reward arm choices. There was a significant main effect of delay, $F(2, 20) = 30.61$, $p < .001$, indicating that choice of the high-reward arm decreased with the increasing delay for each sex. There was no significant interaction between sex and delay. In addition, there was no main effect of estrous cycle stage, delay session, or interaction between estrous cycle stage and delay session in females, indicating that impulsive choice behavior did not vary across the stages of the estrous cycle. Overall, results

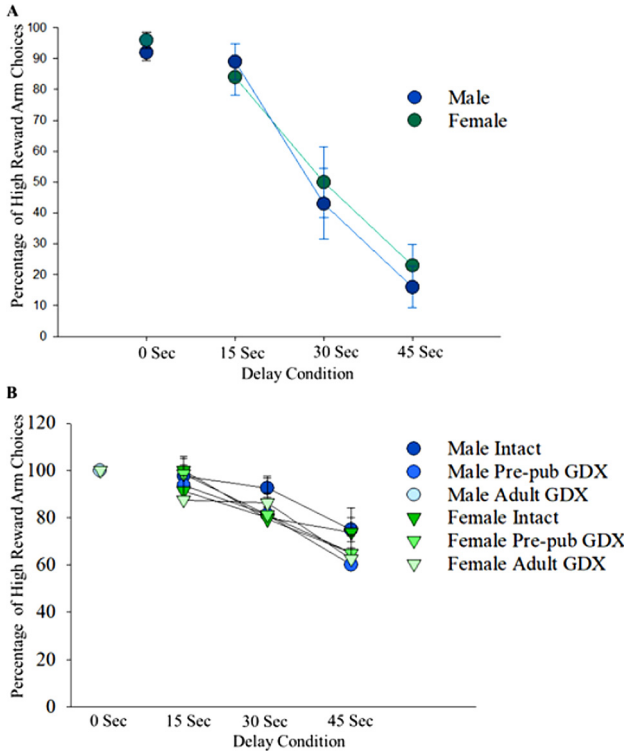


**Fig. 4.** Impact of sex and gonadal hormones acting pre-pubertally or in adulthood on impulsive choice responding on a delay-based reward T-maze task. (A) Effect of increasing delay for high reward on percentage of high reward arm choices in adult male and female gonadally intact adult rats (male n = 10, female n = 10). No main or interactive effects were revealed. (B) Effect of increasing delay for high reward on percentage of high reward arm choices in adult male and female rats that were gonadally intact (Intact, male n = 8, female n = 8), gonadectomized pre-pubertally (Pre-pub GDX, male n = 8, female n = 8), and gonadectomized in adulthood (Adult GDX, male n = 8, female n = 8). No main or interactive effects were revealed.

indicate that the sex difference in impulsive choice seen prior to puberty (Bayless et al., 2013) does not persist into adulthood.

### 3.4. Experiment 2B: The effect of gonadectomy in adulthood or prior to pubertal onset on adult levels of impulsive choice

Because we see a sex difference in impulsive choice prior to puberty (and before the pubertal hormone surge and in the absence of adult circulating hormones) (Bayless et al., 2013) and not in adulthood (Section 3.3), we determined if we could reinstate the sex difference seen prior to puberty by removal of pubertal or adult gonadal hormones.

#### 3.4.1. Performance on the Delay-Based Reward Task

As illustrated in Fig. 4B, male and female adult rats, regardless of hormone status, displayed similar levels of impulsive choice. Analysis of the percentages of high-reward arm choices from the final two sessions of the No-Delay condition revealed no significant main effects of sex or surgery, indicating that all groups preferred the larger reward over the smaller reward when rewards were immediately available. Analysis of the percentages of high-reward arm choices across Delay conditions revealed no significant main effect of sex or surgery, indicating that neither sex or hormone status impacted choice of high- and low-reward arm choices. There was a significant main effect of delay, $F(3, 48) = 41.572$, $p < .001$, indicating that choice of the high reward arm decreased with the increasing delay. There were no significant interactions. Results confirm the lack of sex difference in adulthood seen on this impulsive choice task in Experiment 2A on the Delay-Based Reward Task and indicate that neither pubertal nor adult levels of gonadal hormones impact impulsive choice in adults.

## 4. Discussion

### 4.1. Gonadal hormone influence on mediating sex differences in adult impulsive action

Results of the present experiments indicate that exposure to gonadal hormones during the neonatal period, and not during puberty or adulthood, leads to observed sex difference in levels of impulsive action in adult rats. In Experiment 1A, we compared performance of adult gonadally intact male and female rats as well as those gonadectomized immediately prior to puberty and in adulthood on an impulsive action task. A sex difference in impulsive action was maintained under all conditions, with males committing more impulsive responses than females, regardless of hormone status. In Experiment 1B, we compared performance of adult females that had been treated with testosterone during the neonatal period to adult male and female gonadally intact adult controls on the same impulsive action task. Females treated with testosterone neonatally made significantly more impulsive responses than female controls and were not different from male controls. Results demonstrate that gonadal hormone exposure during the neonatal performance exerts organizational effects on adult behavior resulting in increased impulsive action in males as compared to females. They also demonstrate that pubertal and adult levels of hormones do not impact sex-specific performance on this measure of impulsivity. Of note, our control females exhibited reduced levels of impulsive action in Experiment 3A versus 1B. While these females still exhibited elevated impulsive action under the longest delay of 7.5 s, their levels of impulsive action were reduced at shorter delays. While the direct cause of this reduced impulsive behavior is unknown, one possible difference may be rearing practices with rats in Experiment 1B being shipped in at a peri-pubertal time point versus self-rearing in Experiment 3A. Future investigation should examine to what degree the stress of shipping during this age frame impacts adult levels of impulsive action.

Our findings of an adult sex difference in impulsive action in rats match those previously reported in our lab and others in rodent models (Bayless and Daniel, 2012; Jentsch and Taylor, 2003) and in go/no-go tasks in humans with men making more inhibitory errors than women (Sjoberg and Cole, 2018). No sex difference was found in rats in a lever press go-no go task (Swalve et al., 2018). The mixed results (reviewed by Fattore and Melis, 2016) may be due to different methodologies in testing for impulsive action in rodents, particularly as related to the challenge level of the task used. In the current work, only under challenging conditions of a long inter-trial interval, in which the stimulus is unpredictably lengthened, do males make more impulsive responses than females. This effect is strengthened when examining premature responses during the longest delay of 7.5 seconds, showing males to have greater difficulty in inhibiting premature responding under periods of increased challenge. Additionally, to ensure that differences in responding were not due to differences in motivation for reward nor speed to answer, reward response latency and reward collection latency were measured. No sex differences were seen in either reward response latency nor reward collection latency, demonstrating that the sex difference in impulsive action is not due to motivation or appetitive differences.

Whereas sex differences in impulsive action have been widely observed, the influence of gonadal hormones on their underlying decision-making processes has been less so. To our knowledge, we show for the first time the neonatal critical window to be a period of organization for this adult sex difference in behavior. Previous results in our lab demonstrated a neonatal organizational effect on pre-pubertal impulsive choice (Bayless et al., 2013) adding evidentiary support for gonadal hormone exposure during this period of major brain development to be key in the development of brain structures involved in decision making

specifically. Both in that experiment and in our results, females exposed to testosterone saw a masculinization of impulsive behavior. The mechanism by which this testosterone exposure organizes impulsive behavior is not yet known. Hormones can promote cell survival or death in a sexually dimorphic manner (Forger, 2006) which has implications for behavioral output. Perinatal testosterone exposure has also been shown to lead to lateralization of the brain and increased cell death in the right hemisphere (Geschwind and Galaburda, 1985). This increased lateralization is seen in ADHD subjects, a disorder with higher prevalence in males than females (Seidman et al., 2005). Additionally, perinatal gonadal hormones have been shown to alter dopamine projecting cortical circuitry (Kritzer, 1998). Any or all of these changes in morphology and neurotransmitter function may contribute to a sex difference in impulsive action. These differences are especially of importance when considering therapeutic avenues of approach for treating disorders of impulsivity. For example, modern pharmacotherapies have utilized rodent models to target different neurotransmitter systems for the treatment of impulse control disorders (reviewed by Winstanley, 2011). Sex differences in these systems may impact to what degree such therapies are efficacious. Further investigation needs to be done as to what structural changes occur due to exposure of gonadal hormones during development, changes made to structures involved in impulsive behavior.

### 4.2. Gonadal hormone influence on adult impulsive choice

Previous work in our lab revealed that across two experiments that pre-pubertal males displayed increased impulsive choice as compared to pre-pubertal females on a delayed-reward based task. (Bayless et al., 2013). Interestingly, the present results indicate that this sex difference apparent before puberty does not persist into adulthood. Results of Experiment 2A revealed that males and females displayed similar levels of impulsive choice on a delayed-reward based task. Therefore, we hypothesized that gonadal hormones acting either during puberty or in adulthood would mediate this age-related loss of sex-specific impulsive-choice behavior. However, results of Experiment 2B were not supportive of this hypothesis. Neither gonadectomy prior to puberty nor during adulthood impacted adult levels of impulsive choice in males and female rats. Our findings suggest that age rather than gonadal hormones mediates the transition from male-increased levels of impulsive choice as compared to females that occurs pre-pubertally (see Bayless et al., 2013) to similar levels of impulsive choice across sexes as occurs in adulthood seen in the present study.

In the current work, no sex difference in impulsive choice in adult rats was displayed across two experiments. In both experiments, male and female rats became more impulsive as delay increased choosing the immediate reward over delayed high reward more often. However, the overall impact on impulsivity of increasing the delay differed across the two experiments in our current study. Rats used in Experiment 2B, which displayed overall decreased impulsive choices as compared to rats used in Experiment 2A, were older at the time of testing and had been previously used in an impulsive action study. It remains to be determined if either increased age and/or previous operant conditioning training leads to enhanced ability to inhibit impulsive choice behavior. Interestingly, in a dual behavior and brain imaging study using fMRI, neither behavior correlates nor neutral substrate correlates were found between subjects on impulsive action and impulsive choice tasks (Wang et al., 2016). This supports our findings that impulsive choice and action are dissociated behaviors from one another.

Previous research has shown mixed results with regard to sex differences in impulsive choice, with females sometimes exhibiting greater levels of impulsive choice vs. males (Koot et al., 2009; van Haaren et al., 1988) and some studies finding no sex difference at all (Perry et al., 2007, 2009; Smethells et al., 2016). Impulsive choice, usually measured in delayed discounting tasks, has seen mixed results when studying sex differences in human subjects as well. In a review by Weafer and de Wit (2014) human sex differences in impulsive choice have been shown to be question and task specific with women making more impulsive choices than males for hypothetical rewards and males making more impulsive choices for real rewards. It could be that the lack of sex difference in adulthood in impulsive choice displayed revealed in the current work is task specific.

## 5. Conclusion

The current investigation extends previous findings showing neonatal gonadal hormone exposure is critical for early sexual differentiation of brain and behavior. Our findings determine this window is of particular importance for the development of an adult sex difference in impulsive action, a behavior not affected by loss of pubertal or adult gonadal hormones. These data illustrate the importance of an early time frame for hormonal contribution to sex differences in behavior. Future studies should examine the impact on brain mechanisms implicated in impulsive action of neonatal gonadal hormone exposure.

## CRediT authorship contribution statement

**Jeffrey S. Darling:** Conceptualization, Methodology, Software, Writing - original draft, Software. **Daniel W. Bayless:** Conceptualization, Methodology. **Lauren R. Dartez:** Data curation, Investigation. **Joshua J. Taylor:** Data curation, Investigation. **Arjun Mehrotra:** Data curation, Investigation. **William L. Smith:** Data curation, Investigation. **Jill M. Daniel:** Investigation, Supervision, Validation, Writing - review & editing.

## Declarations of Competing Interest

None.

## Acknowledgements

We thank Amy Theriot Pierce, Associate Director Uptown Campus of the Tulane Department of Comparative Medicine and the Tulane vivarium staff for their expert animal care. This work was supported by the National Institute of Health Grant R21DA043072 to JMD, the Carol Lavin Bernick Tulane Faculty Grant to JMD, and the State of Louisiana Board of Regents Graduate Fellowship LEQSF(2013-18) GF-17 to JSD.

## References

Adkins-Regan and Leung, 2006 E. Adkins-Regan, C.H. Leung, Sex steroids modulate changes in social and sexual preference during juvenile development in zebra finches, Horm Behav. 50 (2006) 772–778.

Bayless and Daniel, 2012 D.W. Bayless, J.M. Daniel, Sex differences in attentional processes in adult rats as measured by performance on the 5-choice serial reaction time task, Behav. Brain Res. 235 (2012) 48–54.

Bayless et al., 2013 D.W. Bayless, J.S. Darling, J.M. Daniel, Mechanisms by which neonatal testosterone exposure mediates sex differences in impulsivity in pre-pubertal rats, Horm. Behav. 64 (2013) 764–769.

Dalley et al., 2008 J.W. Dalley, A.C. Mar, D. Economidou, T.W. Robbins, Neurobehavioral mechanisms of impulsivity: fronto-striatal systems and functional neurochemistry, Pharmacol. Biochem. Behav. 90 (2008) 250–260 PMID: 18272211.

Darling and Daniel, 2019 J.S. Darling, J.M. Daniel, Pubertal hormones mediate sex differences in levels of myelin basic protein in the orbitofrontal cortex of adult rats, Neuroscience 16 (406) (2019) 487–495.

Diergaarde et al., 2008 L. Diergaarde, T. Pattij, I. Poortvliet, F. Hogenboom, W. de Vries, A.N. Schoffelmeer, T.J. De Vries, Impulsive choice and impulsive action predict vulnerability to distinct stages of nicotine seeking in rats, Biol. Psychiatry. 63 (3) (2008) 301–308.

Eagle and Baunez, 2010 D.M. Eagle, C. Baunez, Is there an inhibitory-response-control system in the rat? Evidence from anatomical and pharmacological studies of behavioral inhibition, Neurosci Biobehav Rev. 34 (2010) 50–72.

Evenden, 1999 J. Evenden, Impulsivity: a discussion of clinical and experimental

findings, J. Psychopharmacol. 13 (1999) 180–192.
Fattore and Melis, 2016 L. Fattore, M. Melis, Sex differences in impulsive and compulsive behaviors: a focus on drug addiction, Addict Biol. 21 (2016) 1043–1051.
Forger, 2006 N.G. Forger, Cell death and sexual differentiation of the nervous system, Neuroscience 138 (2006) 929–938.
Geschwind and Galaburda, 1985 N. Geschwind, A.M. Galaburda, Cerebral lateralization. Biological mechanisms, associations, and pathology: I. A hypothesis and a program for research, Arch Neurol. 42 (1985) 428–459.
Hyten et al., 1994 C. Hyten, G.J. Madden, D.P. Field, Exchange delays and impulsive choice in adult humans, J Exp Anal Behav. 62 (2) (1994) 225–233.
Jentsch and Taylor, 2003 J.D. Jentsch, J.R. Taylor, Sex-related differences in spatial divided attention and motor impulsivity in rats, Behavioral Neuroscience. 117 (2003) 76–83.
Koot et al., 2009 S. Koot, R. van den Bos, W. Adriani, G. Laviola, Gender differences in delay-discounting under mild food restriction, Behav Brain Res. 200 (1) (2009) 134–143.
Kritzer, 1998 M.F. Kritzer, Perinatal gonadectomy exerts regionally selective, lateralized effects on the density of axons immunoreactive for tyrosine hydroxylase in the cerebral cortex of adult male rats, J Neurosci 18 (1998) 10735–10748.
McCarthy, 2008 M.M. McCarthy, Estradiol and the developing brain, Physiology Rev. 88 (1) (2008) 91–240.
Muir et al., 1996 J.L. Muir, B.J. Everitt, T.W. Robbins, The cerebral cortex of the rat and visual attentional function: Dissociable effects of mediofrontal, cingulate, anterior dorsolateral, and parietal cortex lesions on a five-choice serial reaction time task, Cerebral Cortex 6 (3) (1996) 470–481, https://doi.org/10.1093/cercor/6.3.470 In press.
Perry et al., 2009 J.L. Perry, S.E. Nelson, M.E. Carroll, Impulsive choice as a predictor of acquisition of IV cocaine self- administration and reinstatement of cocaine-seeking behavior in male and female rats, Exp Clin Psychopharmacology. 16 (2) (2009) 165–177.
Perry et al., 2007 J.L. Perry, S.E. Nelson, M.M. Anderson, A.D. Morgan, M.E. Carroll, Impulsivity (delay discounting) for food and cocaine in male and female rats selectively bred for high and low saccharin intake, Pharmacol. Biochem Behav. 86 (4) (2007) 822–837.
Phoenix et al., 1959 C.H. Phoenix, R.W. Goy, A.A. Gerall, W.C. Young, Organizing action of prenatally administered testosterone propionate on the tissues mediating mating behavior in the female guinea pig, Endocrinology 65 (1959) 369–382.
Robbins, 2002 T.W. Robbins, The 5-choice serial reaction time task: behavioural pharmacology and functional neurochemistry, Psychopharmacology (Berl) 163 (2002) 362–380.
Sanchez-Roige et al., 2012 S. Sanchez-Roige, Y. Pena-Oliver, D.N. Stephens, Measuring impulsivity in mice: the five-choice serial reaction time task, Psychopharmacology 219 (2) (2012) 253–270.
Schulz et al., 2009 K.M. Schulz, H.A. Molenda-Figueira, C.L. Sisk, Back to the future: the organizational-activational hypothesis adapted to puberty and adolescence, Hormones and Behavior 55 (5) (2009) 597–604.
Schulz et al., 2004 K.M. Schulz, H.N. Richardson, J.L. Zehr, A.J. Osetek, T.A. Menard, C.L. Sisk, Gonadal hormones masculinize and defeminize reproductive behaviors during puberty in the male Syrian hamster, Horm Behav 45 (2004) 242–249.
Schulz et al., 2006 K.M. Schulz, T.A. Menard, D.A. Smith, H.E. Albers, C.L. Sisk, Testicular hormone exposure during adolescence organizes flank-marking behavior and vasopressin receptor binding in the lateral septum, Horm Behav. 50 (2006) 477–483.
Seidman et al., 2005 L.J. Seidman, E.M. Valera, N. Makris, Structural brain imaging of attention-deficit/hyperactivity disorder, Biol Psychiatry 57 (11) (2005) 1263–1272, https://doi.org/10.1016/j.biopsych.2004.11.019 In press.
Sjoberg and Cole, 2018 E.A. Sjoberg, G.G. Cole, Sex differences on the Go/No-Go test of inhibition, Archives of Sexual Behavior 47 (2018) 537–542.
Smethells et al., 2016 J.R. Smethells, N.L. Swalve, L.E. Eberly, M.E. Carroll, Sex differences in the reduction of impulsive choice (delay discounting) for cocaine in rats with atomoxetine and progesterone, Psychopharmacology. 233 (2016) 2999–3008.
Swalve et al., 2018 N. Swalve, J.R. Smethells, R. Younk, J. Mitchell, B. Dougen, M.E. Carroll, Sex-specific attenuation of impulsive action by progesterone in a Go/No-Go task for cocaine in rats, Psychopharmacology. 235 (2018) 135–143.
Takahashi, 1990 L.K. Takahashi, Hormonal regulation of sociosexual behavior in female mammals, Neurosci. Biobehav. Rev. 14 (1990) 403–413 PMID: 24415819.
Van Haaren et al., 1988 F. Van Haaren, A. Van Hest, N. Van De Poll, Self-control in male and female rats, Journal of Exp Anal Behav. 49 (2) (1988) 201–211.
Wallen and Baum, 2002 K. Wallen, M.J. Baum, Masculinization and defeminization in altricial and precocial mammals: comparative aspects of steroid hormone action, Hormones, Brain, and Behavior (2002) 385–423 Elsevier.
Wang et al., 2016 Q. Wang, C. Chen, Y. Cai, et al., Dissociated neural substrates underlying impulsive choice and impulsive action, NeuroImage. 134 (2016) 540–549.
Watanabe and Terasawa, 1989 G. Watanabe, E. Terasawa, In vivorelease of luteinizing hormone releasing hormone increases with puberty in the female rhesus monkey, Endocrinology. 125 (1989) 92–99 PMID: 2661213.
Weafer and de Wit, 2014 J.W. Weafer, H. de Wit, Sex differences in impulsive action and impulsive choice, Addict Behav 39 (2014) 1573–1579.
Whelan et al., 2012 R. Whelan, P.J. Conrod, J.B. Poline, A. Lourdusamy, T. Banaschewski, G.J. Barker, H. Garavan, Adolescent impulsivity phenotypes characterized by distinct brainnetworks, Nature Neuroscience 15 (6) (2012) 920–925.
Winstanley, 2011 C.A. Winstanley, The utility of rat models of impulsivity in developing pharmacotherapies for impulse control disorders, British journal of pharmacology 164 (4) (2011) 1301–1321.
Zhang et al., 2008 J.M. Zhang, A.T. Konkle, S.L. Zup, M.M. McCarthy, Impact of sex and hormones on new cells in the developing rat hippocampus: a novel source of sex dimorphism? European Journal of Neuroscience 27 (4) (2008), https://doi.org/10.1111/j.1460-9568.2008.06073.x In press.