Drug and Alcohol Dependence 174 (2017) 137–144

Contents lists available at ScienceDirect

Drug and Alcohol Dependence

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

Full length article

Sex differences and hormonal modulation of ethanol-enhanced risk taking in rats

Kathryn G. Wallin-Miller[a], Jordyn Chesley[b], Juliana Castrillon[b], Ruth I. Wood[b,*]

[a] Neuroscience Graduate Program, University of Southern California, Los Angeles, CA 90033, USA
[b] Department of Cell and Neurobiology, Keck School of Medicine of the University of Southern California, Los Angeles, CA 90033, USA

ARTICLE INFO

Article history:
Received 17 August 2016
Received in revised form 1 December 2016
Accepted 9 January 2017
Available online 7 March 2017

Keywords:
Sex characteristics
Gonadal hormones
Decision making
Choice behavior
Operant behavior
Reward

ABSTRACT

Background: Ethanol (EtOH) intake correlates with increased risk-taking, and sex differences exist in both EtOH use and risk-taking in humans and rats. However, the interaction of sex and gonadal hormones to affect risk-taking under the influence of EtOH has not been determined. This was the focus of the current study.

Methods: Adult Long-Evans rats (n = 18 males and females) were gonadectomized and received hormone replacement at physiologic levels or blank implants (n = 7-9/group). Risk-taking was assessed with probability discounting, requiring rats to choose between a small/certain reward and a large/uncertain reward delivered with decreasing probability throughout each daily session. Before testing, rats received saline or EtOH (0.5 or 1.0 g/kg) ip.

Results: In males, EtOH increased preference for the large/uncertain reward lever ($F_{2,28} = 10.462$, $p < 0.05$). However, there was no effect of EtOH on lever preference in females ($F_{1,30} = 0.914$, $p > 0.05$). At baseline, ORCHX+T males showed a greater preference for the large/uncertain reward lever then ORCHX males ($F_{1,14} = 13.805$, $p < 0.05$). In females only, EtOH decreased choice latency relative to baseline ($F_{1,10} = 7.25$, $p < 0.05$). EtOH decreased loss sensitivity in both sexes, with all rats exhibiting decreased lose-shift ratios (males: $F_{2,18} = 5.10$, $p < 0.05$; females $F_{2,10} = 4.37$, $p < 0.05$).

Conclusions: These results show that EtOH, sex, and hormones interact to influence decision making. EtOH increases risk taking in males, but not in females. However, EtOH selectively decreases choice latency in females, and decreases loss sensitivity in both sexes. These findings are relevant to understanding human behavior, particularly in adolescents who experience increased hormone levels and often drink EtOH and engage in risky behavior.

1. Introduction

Ethanol (EtOH) is the most-commonly used drug worldwide (Winstock, 2014), with 87.6% of American adults reporting EtOH consumption during their lifetime (NIAAA, 2016). According to The Independent Scientific Committee on Drugs, EtOH causes more harm than any other illicit substance, due to its prevalence and detriments to both the user and society (Nutt et al., 2010). National surveys show that drinking, impulsive behavior and injury are positively correlated (NIAAA, 2016; Cherpitel, 1993; Borges et al., 2004). Additionally, drinking is associated with gambling behavior, as research suggests that alcohol use positively correlates with both gambling frequency and gambling losses (Martens et al., 2009). However, in these large-scale human studies, it is difficult to establish cause and effect, and to examine underlying mechanisms. Furthermore, sex differences exist in both EtOH use and risk-taking, with men consuming more alcohol and engaging in more risky behavior such as criminal activity, drug use, biking or driving without a helmet or seatbelt, and problematic gambling (Johansson et al., 2009; Carroll et al., 2004; MacArthur et al., 2012). However, it is unknown if EtOH has differential effects on risk-taking in men and women, an issue which has important implications for public health and safety.

Gonadal steroids also stimulate risk-taking behavior. Under the influence of gonadal steroids, male and female adolescents engage in increased risk-taking behaviors (such as hazardous drinking, drug use, and unprotected sex) relative to pre-pubertal years (MacArthur et al., 2012; Kuntsche and Gmel, 2013).

* Corresponding author at: Department of Cell and Neurobiology, Keck School of Medicine of the University of Southern California, 1333 San Pablo St., BMT 401a, Los Angeles, CA 90033, USA.
E-mail addresses: kwallin@usc.edu (K.G. Wallin-Miller), chesley@usc.edu (J. Chesley), jcastrillon@college.harvard.edu (J. Castrillon), riw@usc.edu (R.I. Wood).

http://dx.doi.org/10.1016/j.drugalcdep.2017.01.023
0376-8716/

Additionally, endogenous testosterone levels in human men correlate with increased risk taking under uncertainty in both the Iowa Gambling Task (Stanton et al., 2011) and the stock market (Coates and Herbert, 2008). Furthermore, EtOH use in teen girls correlates with circulating estradiol levels (Martin et al., 1999) and higher androgen levels in pubertal boys correlate with greater probability of lifetime EtOH use (de Water et al., 2013). Therefore, the presence of circulating gonadal steroids may enhance EtOH effects on behavior, potentially increasing risk-taking in one or both sexes. To address this possibility, the present study compared gonadectomized male and female rats with and without hormone replacement to investigate the influence of EtOH and gonadal steroids on the response to uncertainty as a measure of risk-taking behavior.

Sex differences exist in animal models of both risk-taking and EtOH use. Female rats, like female humans, are more risk-averse than males in an experimental model of decision-making (Orsini et al., 2016). Unlike humans, female rats exhibit greater preference for and higher voluntary intake of EtOH (McGlacken et al., 1995). However, this difference may be due to cultural constraints on women in regard to EtOH use. Animal models allow the study of sex and hormone effects on behavior and eliminate the potential confound of gender expectations. To test decision making in rats, discounting paradigms require subjects to choose between two rewards: a small "safe" reward, and a large reward that is "discounted" or made less desirable by pairing with a cost. Probability discounting tests risk taking in the context of reward uncertainty, a paradigm related to human risk taking in the form of gambling. Rats choose between a small/certain reward of 1 pellet, and a large/uncertain reward of 3–4 pellets, delivered with decreasing probability in successive blocks of trials. Probability discounting has been used to show effects of drugs such as amphetamine on risk taking (St. Onge and Floresco, 2009). However, EtOH effects on risky behavior in probability discounting have not been determined.

Because drinking, risk taking, and gambling are correlated in human studies, we hypothesized that EtOH would increase tolerance to uncertainty in rats performing probability discounting. Like EtOH, gonadal steroids influence decision-making, with increased hormone levels facilitating risk taking in human males (Stanton et al., 2011; Coates and Herbert, 2008). Therefore, we expected that rats receiving gonadal steroid replacement would exhibit greater risk taking than their counterparts without steroids. Finally, EtOH may interact with sex and/or hormonal milieu. Because drinking and risk taking increase during human adolescence, EtOH may have a stronger effect on risky decision making in steroid-treated rats.

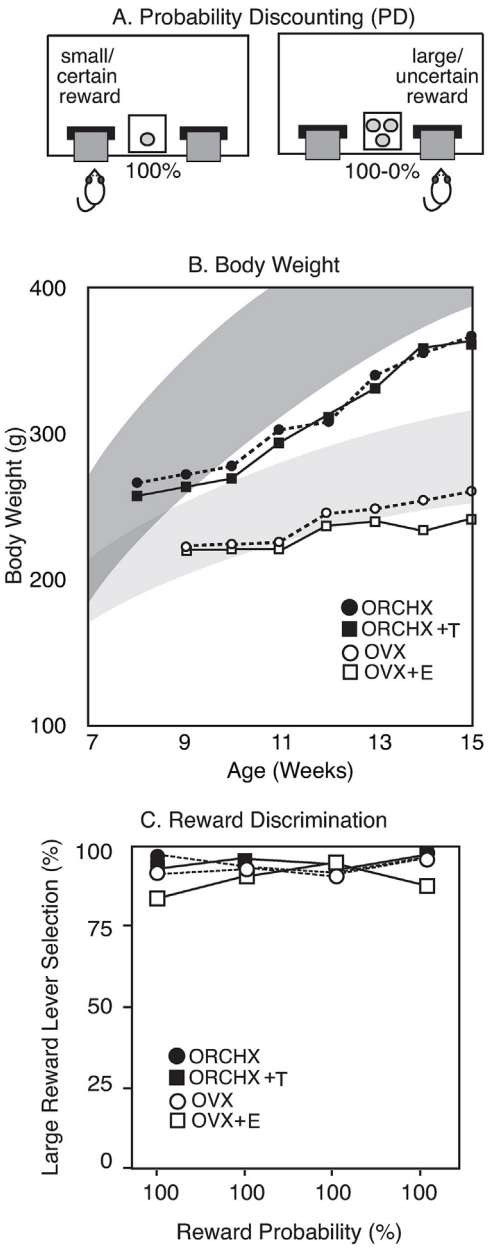

Fig. 1. A) Probability Discounting to measure risk taking with operant behavior. Rats choose between a small/certain reward (1 pellet delivered with 100% probability) and a large/uncertain reward (3 pellets delivered with decreasing probability). B) Average body weights of each group throughout the experiment, relative to the range of normal body weights for male and female Long-Evans rats fed *ad libitum* (Charles River, 2016) C) Large reward lever selection in free-choice trials by each of the 4 groups during reward discrimination training.

2. Methods

2.1. Animals

Young adult male and female Long-Evans rats (8–10 weeks of age at the start, Charles River Laboratories, MA) were pair-housed under a reversed 14L:10D photoperiod. As in our previous studies (Cooper et al., 2014; Wallin and Wood, 2015), rats were food-restricted to maintain a slow rate of growth (Fig. 1B) and facilitate operant responding. Experimental procedures were approved by USC's Institutional Animal Care and Use Committee and were conducted in accordance with the Guide for the Care and Use of Laboratory Animals, 8th Ed (National Research Council, National Academies Press, Washington DC, 2011).

2.2. Gonadectomy

All rats (16 male, 18 female) were gonadectomized under ketamine/xylazine anesthesia, and half received hormone replacement (n = 7–9/group) as in our previous studies (Antzoulatos et al., 2011; Kent et al., 2013). Hormone implants were made from Silastic tubing (id: 1.57 mm, od: 3.18 mm, Dow Corning, Midland, MI) filled with crystalline steroid to provide chronic replacement of testosterone or estradiol at physiologic levels (Moger, 1976; Bridges, 1984). Males were castrated via a mid-line scrotal incision, and received a 10 mm Silastic implant s.c. either filled with testosterone (ORCHX+T) or blank (ORCHX). These testosterone implants maintain serum testosterone at the level of intact adult male rats (~2.5 ng/ml) (Moger, 1976; Damassa et al., 1977). Females were ovariectomized via bilateral dorsal flank incisions and received a 5 mm implant s.c. filled with 17β-estradiol (OVX+E) or blank