Hormones and Behavior 64 (2013) 579–588

Contents lists available at ScienceDirect

Hormones and Behavior

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

Voluntary exercise impairs initial delayed spatial alternation performance in estradiol treated ovariectomized middle-aged rats

Steven L. Neese [a,b,c,*], Donna L. Korol [a,d,e,f], Susan L. Schantz [a,b]

[a] Neuroscience Program, University of Illinois at Urbana-Champaign, USA
[b] Department of Comparative Biosciences, University of Illinois at Urbana-Champaign, USA
[c] Department of Psychology and Neuroscience, Baldwin Wallace University, USA
[d] Department of Psychology, University of Illinois at Urbana-Champaign, USA
[e] Institute for Genomic Biology, University of Illinois at Urbana-Champaign, USA
[f] Department of Biology, Syracuse University, USA

ARTICLE INFO

Article history:
Received 10 May 2013
Revised 22 July 2013
Accepted 22 August 2013
Available online 3 September 2013

Keywords:
Working memory
Voluntary wheel running
Estradiol
DSA
Middle-aged

ABSTRACT

Estrogens differentially modulate behavior in the adult female rodent. Voluntary exercise can also impact behavior, often reversing age associated decrements in memory processes. Our research group has published a series of papers reporting a deficit in the acquisition of an operant working memory task, delayed spatial alternation (DSA), following 17β-estradiol treatment to middle-aged ovariectomized (OVX) rats. The current study examined if voluntary exercise could attenuate the 17β-estradiol induced deficits on DSA performance. OVX 12-month old Long–Evans rats were implanted with a Silastic capsule containing 17β-estradiol (10% in cholesterol: low physiological range) or with a blank capsule. A subset of the 17β-estradiol and OVX untreated rats were given free access to a running wheel in their home cage. All rats were tested for 40 sessions on the DSA task. Surprisingly, we found running wheel access to impair initial acquisition of the DSA task in 17β-estradiol treated rats, an effect not seen in OVX untreated rats given running wheel access. This deficit was driven by an increase in perseverative responding on a lever no longer associated with reinforcement. We also report for the first time a 17β-estradiol induced impairment on the DSA task following a long intertrial delay (18-sec), an effect revealed following more extended testing than in our previous studies (15 additional sessions). Overall, running wheel access increased initial error rate on the DSA task in 17β-estradiol treated middle-aged OVX rats, and failed to prevent the 17β-estradiol induced deficits in performance of the operant DSA task in later testing sessions.

Introduction

Specific cognitive processes appear to decline more rapidly in the aging female after menopause, including memory processes regulated by the prefrontal cortical system (Sherwin and Henry, 2008), but findings from research assessing the effects of hormone replacement therapies (HRT) on cognitive decline have been inconsistent (reviewed in Coker et al., 2010; Hogervorst and Bandelow, 2010; Lethaby et al., 2008; Sherwin and Henry, 2008). The use of HRT by menopausal women declined dramatically after the Women's Health Initiative's study found that these therapies can increase the risk of stroke, breast cancer and heart attack (Chlebowski et al., 2003; Rossouw et al., 2002; Wassertheil-Smoller et al., 2003). However, the number of women using HRT remains significant, with over 35 million women filling prescriptions for HRT in 2010 (North American Menopause Association). Therefore, determining the impact of HRT on cognitive processes in aging postmenopausal women remains important.

Studies in animal models have revealed that 17β-estradiol differentially modulates performance on several distinct types of cognitive tasks (see Dohanich et al, 2009; Frick, 2009; Korol, 2004; Luine, 2008). In particular, 17β-estradiol restores the performance of ovariectomized (OVX) female rats to pre-OVX levels on tasks that engage the hippocampus (Daniel and Dohanich, 2001; Daniel et al., 1997; Davis et al., 2005; Korol and Kolo, 2002; Zurkovsky et al., 2006, 2007), but can impair the performance of OVX female rats on tasks that engage other brain structures, including prefrontal cortical and striatal memory systems (Davis et al., 2005; Korol and Kolo, 2002; Neese et al., 2010a, 2013; Wang et al., 2008, 2009; Zurkovsky et al., 2007). These findings suggest that 17β-estradiol can have task and brain region-specific effects on cognition. Of particular importance are tasks that tap the prefrontal cortex as chronic 17β-estradiol treatment to OVX rodents can alter dopamine signaling, resulting in reduced prefrontal tissue dopamine content (Dupont et al., 1981; Luine et al., 1998; Pandaranandaka et al., 2006). Further, rodent research has found that alterations in prefrontal dopamine result in performance deficits on a variety of delayed response tasks (Bubser and Schmidt, 1990; Kozlov et al., 2001;

* Corresponding author at: Department of Psychology and Neuroscience, 275 Eastland Road, Baldwin Wallace University, Berea, OH 44017, USA.
E-mail address: sneese@bw.edu (S.L. Neese).

0018-506X/$ – see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.yhbeh.2013.08.008

Mizoguchi et al., 2009; Vijayraghavan et al., 2007; Zahrt et al., 1997), providing one mechanism through which chronic 17β-estradiol treatment can negatively alter behaviors dependent on this brain region.

Our research group has published a series of studies showing that 17β-estradiol treatment impairs the performance of young, middle-aged, and old OVX rats on an operant test of working memory, the delayed spatial alternation (DSA) task (Neese et al., 2010a, 2013; Wang et al., 2008, 2009). 17β-stradiol typically improves the performance of OVX rodents on maze-based working memory tasks, but these tasks often use long intertrial delays (see Aggleton et al., 1995; Pontecorvo et al., 1996; Van Hest and Steckler, 1996), and have been shown to rely heavily on the hippocampus for accurate performance (D'Hooge and DeDeyn, 2001; Floresco et al., 1997; Goldman-Rakic, 1995; Jones, 2002). Conversely, by utilizing short intertrial delays with automated operant testing the prefrontal cortex is selectively engaged. Specifically, lesioning or pharmacological inactivation of the prefrontal cortex results in clear deficits on operant working memory tasks with intertrial delays as short as 1–3 s, whereas hippocampal inactivation does not alter performance at these short delays (Chudasama and Muir, 1997; Harrison and Mair, 1996; Mair et al., 1998; Sloan et al., 2006; Van Haaren et al., 1985, 1988; Young et al., 1996). With longer intertrial delays (>10–15 s), hippocampal disruption begins to impair performance accuracy (Chudasama and Muir, 1997; Dunnett, 1985; Izaki et al., 2008; Kirkby and Higgins, 1998; Maruki et al., 2001). Therefore, operant working memory tasks that include short intertrial delays can specifically tap the prefrontal cortical system.

Voluntary exercise (i.e. running wheel access) has been studied as an intervention to reduce age associated cognitive decline in rodents (see van Praag, 2008). Voluntary exercise can improve the performance of gonadally-intact young, middle-aged and old male and female rodents on a variety of cognitive tasks, including maze-based reference and working memory tasks (Leggio et al., 2005; Marlatt et al., 2012; Van der Borght et al., 2007; van Praag et al., 1999, 2005), tests of recognition memory (Hopkins et al., 2011), and passive avoidance tasks (Samorajski et al., 1985). Of note, treadmill exercise for 20 min three times per week reverses deficits in reference and working memory in the Morris water maze in OVX female rats but fails to improve memory in gonadally intact female rats (Ben et al., 2010). However, little is known about the effects of exercise on memory processes in aging (>12 month old) OVX rats.

Human research also suggests a link between exercise and cognitive health during aging (see Plassman et al., 2010; van Uffelen et al., 2008), and one study found that fitness levels measured at the time of testing and duration of HRT taken previously can interact to differentially affect cognition in aging women (Erickson et al., 2007). Specifically, this study found that long term HRT (over 11 years) resulted in more perseverative errors on the Wisconsin Card Sorting Task in women with a lower fitness level, relative to women on long term HRT that were more physically fit. These study results suggest that long term HRT may impair some aspects of cognition mediated by the prefrontal cortex (see also Coker et al., 2010; Hogervorst and Bandelow, 2010; Lethaby et al., 2008), and that physical activity may protect against these effects in aging women undergoing HRT.

The current study was conducted to determine if the detrimental effects of 17β-estradiol treatment on a prefrontal-cortex sensitive operant DSA task could be prevented by voluntary daily exercise. Middle-aged rats were OVX, and treated with 17β-estradiol or left untreated. A subset of the 17β-estradiol-treated and untreated OVX rats were given free access to a running wheel in their home cage. Thus, the goal of this study was to determine if the 17β-estradiol-induced deficit in operant DSA performance could be attenuated by voluntary running wheel activity. All rats were subject to 40 sessions of DSA testing.

Methods

Animals and exposure

Twenty-nine 12-month old female Long–Evans retired breeder rats were obtained from Harlan (Indianapolis, IN) in 2 cohorts (see below) and maintained in facilities fully accredited by the Association for the Assessment and Accreditation of Laboratory Animal Care (AAALAC). Rats were housed in a temperature and humidity controlled room (22 °C, 40–55% humidity) on a 12-hour reverse light–dark cycle (lights off at 8:30 am). All procedures were approved by the Institutional Animal Care and Use Committee (IACUC) at the University of Illinois at Urbana-Champaign and were in accordance with the guidelines of the *Public Health Service Policy on Humane Care and Use of Laboratory Animals* (National Institutes of Health, 2002) and the *Guidelines for the Care and Use of Mammals in Neuroscience and Behavioral Research* (National Research Council Institute for Laboratory Animals Research, 2003).

Rats were single-housed in plastic cages (22.9 × 21.6 × 20.3 in.) with woodchip bedding. All rats were subject to isoflurane gas anesthesia prior to OVX and the implant of a Silastic capsule that was either left blank (n = 13) or contained 17β-estradiol (n = 16). The Silastic capsules were 1 cm in length (1.5 mm i.d., 1.96 mm o.d.) and were plugged with silicone and dried overnight before packing with either a 10% 17β-estradiol/cholesterol mixture (Sigma, St. Louis, MO) or left blank, after which the other end was plugged with silicone. Capsules were soaked in sterile saline at 37 °C overnight before insertion during surgery. Previous research in this lab (Neese et al., 2010a) has shown that these 17β-estradiol implants produce stable serum estradiol concentrations of about 20–30 pg/ml for at least 10 weeks.

Beginning one week after OVX surgery, rats were weighed daily and food was restricted to maintain them at 85% of their free-feeding body weights. A subset of the 17β-estradiol implanted rats (estradiol, n = 8) and OVX rats given a blank implant (OVX, n = 5) were placed into cages with a running wheel freely available. The running wheel was 34.3 in. in diameter with a 10.2 inch lane width. Wheel rotations were recorded once daily. Final experimental groups were as follows: OVX with 17β-estradiol implant and running wheel access (estradiol-wheel, n = 8), OVX with 17β-estradiol implant and no running wheel access (estradiol-no wheel, n = 8), OVX with a blank implant and running wheel access (OVX-wheel, n = 5), and OVX with blank implant and no running wheel access (OVX-no wheel, n = 8). Due to a limited number of wheels, the OVX-wheel group was included in a separate testing cohort.

Soy phytoestrogens are present in standard rodent diets and can influence behavior (see Brown and Setchell, 2001; Lee et al., 2009; Lund et al., 2001; Neese et al., 2010b, 2012; Pisani et al., 2012; Thigpen et al., 2004, 2007). Therefore, to avoid exposure to additional estrogens, the rats in this study were maintained on a low-soy diet (Harlan diet 2016, Madison, WI). Water was available ad libitum. During behavioral testing, rats were fed 1 h after the daily test session was completed. Operant training began two weeks following OVX and occurred once daily, six days/week during the dark phase of the light cycle.

Operant testing

Behavioral testing was conducted in standard automated operant chambers (Med Associates Inc., St. Albans, VT) housed in sound-attenuated wooden boxes. All of the test chambers had the same features and dimensions: 21.6 cm high with a 29.2 cm × 24.8 cm stainless-steel grid floor that rested just above a tray filled with woodchip bedding. Soy-free 45-mg food pellets (5TUL, Test diet, Richmond, IN) were dispensed through a pellet dispenser centered 2.5 cm above the floor on the operant panel. A pair of retractable response levers and a pair of stimulus cue lamps, one above each lever, were positioned symmetrically on both sides of the pellet dispenser. The levers were 5.7 cm from midline and 7.0 cm above the floor and the cue lights were located 5.7 cm above the levers. Each chamber also contained a Sonalert tone generator, a white noise generator, and a house light located on the back wall. Experimental contingencies were programmed using the Med-State behavioral programming language (Med-Associates, Vermont).