Alcohol 49 (2015) 121–126

Contents lists available at ScienceDirect

Alcohol

journal homepage: http://www.alcoholjournal.org/

The effects of acute alcohol on motor impairments in adolescent, adult, and aged rats

Laura C. Ornelas [a], Adelle Novier [a], Candice E. Van Skike [a], Jaime L. Diaz-Granados [a], Douglas B. Matthews [a,b,*]

[a] Baylor University, One Bear Place #97334, Waco, TX 76798, USA
[b] University of Wisconsin–Eau Claire, HHH273, Eau Claire, WI 54702, USA

ARTICLE INFO

Article history:
Received 10 October 2014
Received in revised form
1 December 2014
Accepted 5 December 2014

Keywords:
Alcohol
Aerial righting reflex
Rotarod
Aging
Loss of righting reflex

ABSTRACT

Acute alcohol exposure has been shown to produce differential motor impairments between aged and adult rats and between adolescent and adult rats. However, the effects of acute alcohol exposure among adolescent, adult, and aged rats have yet to be systematically investigated within the same project using a dose-dependent analysis. We sought to determine the age- and dose-dependent effects of acute alcohol exposure on gross and coordinated motor performance across the rodent lifespan. Adolescent (PD 30), adult (PD 70), and aged (approximately 18 months) male Sprague–Dawley rats were tested on 3 separate motor tasks: aerial righting reflex (ARR), accelerating rotarod (RR), and loss of righting reflex (LORR). In a separate group of animals, blood ethanol concentrations (BEC) were determined at multiple time points following a 3.0 g/kg ethanol injection. Behavioral tests were conducted with a Latin square repeated-measures design in which all animals received the following doses: 1.0 g/kg or 2.0 g/kg alcohol or saline over 3 separate sessions via intraperitoneal (i.p.) injection. During testing, motor impairments were assessed on the RR 10 min post-injection and on ARR 20 min post-injection. Aged animals spent significantly less time on the RR when administered 1.0 g/kg alcohol compared to adult rats. In addition, motor performance impairments significantly increased with age after 2.0 g/kg alcohol administration. On the ARR test, aged rats were more sensitive to the effects of 1.0 g/kg and 2.0 g/kg alcohol compared to adolescents and adults. Seven days after the last testing session, animals were given 3.0 g/kg alcohol and LORR was examined. During LORR, aged animals slept longer compared to adult and adolescent rats. This effect cannot be explained solely by BEC levels in aged rats. The present study suggests that acute alcohol exposure produces greater motor impairments in older rats when compared to adolescent and adult rats and begins to establish a procedure to determine motor effects by alcohol across the lifespan.

The average age of the world's population is rapidly rising (Lutz, Sanderson, & Scherbov, 2008). According to the United States Census Bureau, individuals aged 65 and older are projected to total approximately 72 million people and represent nearly 20% of the total U.S. population in the year 2030 (He, Sengupta, Velkoff, & DeBarros, 2005). Importantly, approximately 50% of the elderly population (aged over 65) and almost 25% of individuals over 85 years old currently drink alcohol (Caputo et al., 2012). In addition, nearly 13% of men and 8% of women over the age of 65 consume alcohol in a binge-drinking manner (Blazer & Wu, 2009). In the United States, it is estimated that the prevalence of alcohol-use disorders among the elderly population is approximately 1–3% (Caputo et al., 2012). As the global population continues to increase, substantial alcohol consumption and its consequences in the elderly are an important, but understudied, public health concern (Babor, 2010).

Individuals aged 65 years and older are especially susceptible to the risk factors associated with alcohol consumption. Increased alcohol consumption in aged individuals could be a contributing factor to cognitive deficits including dementia (Thomas & Rockwood, 2001), and to impaired motor coordination and increased falls in older individuals (Høidrup, Grønbæk, Gottschau, Lauritzen, & Schroll, 1999; Jones, Cyr, & Patil, 1994; Mukamal et al., 2004; Mukamal, Robbins, Cauley, Kern, & Siscovick, 2007; Weafer & Fillmore, 2012). Deficits in motor coordination in this age group may be related to age-dependent changes in cerebellar function (Piguet et al., 2006). Alcohol has been shown to decrease cell density and size of the cerebellar vermis, resulting in gait ataxia

* Corresponding author. University of Wisconsin–Eau Claire, Department of Psychology, Eau Claire, WI 54702, USA. Tel.: +1 715 836 2119; fax: +1 715 836 2124.
E-mail address: matthedb@uwec.edu (D.B. Matthews).

http://dx.doi.org/10.1016/j.alcohol.2014.12.002
0741-8329/

(Johnson-Greene et al., 1997; Phillips, Harper, & Kril, 1990; Piguet et al., 2006). Therefore, the aging cerebellum paired with heavy alcohol consumption could lead to increased motor impairments in the elderly.

In concordance with age-related effects of alcohol in humans, research using animal models has shown that many of the effects of alcohol are age-dependent (Chin, Van Skike, & Matthews, 2010 for review). Adolescent rats, compared to adult rats, have been shown to be less sensitive to the sedative (Little, Kuhn, Wilson, & Swartzwelder, 1996), anxiogenic (Doremus, Brunell, Varlinskaya, & Spear, 2003), hypnotic (Matthews, Tinsley, Diaz-Granados, Tokunaga, & Silvers, 2008; Silveri & Spear, 1998), and motor impairing effects of acute alcohol (Ramirez & Spear, 2010; Van Skike et al., 2010; White et al., 2002). Furthermore, adolescent rodents exhibit significantly more acute tolerance compared to adult rodents (Draski, Bice, & Deitrich, 2001; Grieve & Littleton, 1979; Silveri & Spear, 1998, 2002; Varlinskaya & Spear, 2006) and are more sensitive to alcohol-induced hypothermia (Ristuccia & Spear, 2008).

Research indicates that older rodents are more sensitive to acute alcohol exposure compared to adolescent and adult rats (Novier, Van Skike, Diaz-Granados, Mittleman, & Matthews, 2013; Van Skike et al., 2010). Older rodents are more sensitive to the hypnotic (Ott, Hunter, & Walker, 1985) and hypothermic effects of acute alcohol (Wood & Armbrecht, 1982), as well as the severity of alcohol withdrawal (Wood, Armbrecht, & Wise, 1982) compared to younger rodents. To explore the effects of alcohol across the lifespan, Van Skike et al. (2010) examined the impact of a single acute alcohol dose on motor impairments in 4 rodent age groups. Aged rats showed significantly more impairment in motor performance compared to periadolescent and adolescent rats. Furthermore, Novier et al. (2013) investigated the differences in the motor and memory-impairing effects of acute alcohol between adult and aged rats. Similarly, results indicate that aged animals performed significantly worse in all behavioral measures compared to adult rats.

Although current research has recognized differential motor impairments between adolescent and aged rats (Van Skike et al., 2010; White et al., 2002) and adult and aged rats (Novier et al., 2013), research has yet to systematically investigate the effect of acute ethanol on motor impairments across the lifespan using a dose-dependent analysis. In addition, the effect of age on a high dose of alcohol-induced hypnosis has yet to be investigated. Therefore, we sought to determine the age- and dose-dependent effects of acute alcohol exposure on gross and coordinated motor performance in adolescent, adult, and aged rats using the accelerated rotarod (RR) and aerial righting reflex (ARR). In addition, the effect of 3.0 g/kg on ethanol-induced loss of righting reflex (LORR) was determined. Finally, blood ethanol concentrations (BEC) were determined in a separate group of animals at 7 different time points following a 3.0 g/kg ethanol injection to better understand how BEC affects LORR at 3 different ages. We present evidence that alcohol produces greater motor impairments in older rats when compared to adolescent and adult rats and these motor impairments are not completely explained by blood ethanol concentrations.

Materials and methods

Subjects

Twelve adolescent (postnatal day (PD) 28), 12 young adult (approximately PD 70), and 12 aged (approximately 18 months) male Sprague–Dawley rats were obtained from Harlan Laboratories (Indianapolis, IN). These ages were selected because PD 28–30 is a developmental period of early adolescence based on evidence that mature sperm is not yet found, while all sperm are mature at PD 70, indicating this age is the beginning of adulthood (Odell, 1990). Aged animals were 18 months, to be consistent with our previous work (Novier et al., 2013). Animal care procedures were approved by the Institutional Animal Care and Use Committee of Baylor University. Animals were individually housed and given *ad libitum* access to food and water throughout the experiment. Following previously published methods, all rats acclimated to the colony room for 2 days before any experimental procedures (Chin et al., 2011; Novier, Van Skike, Chin, Diaz-Granados, & Matthews, 2012; Novier et al., 2013; Silvers et al., 2006; Tokunaga, Silvers, & Matthews, 2006; Van Skike, Novier, Diaz-Granados, & Matthews, 2012).

To investigate the dose-dependent effects of acute ethanol exposure in the same animals and therefore reduce subject number, animals were involved in a Latin Square repeated-measures design with experimental doses of 1.0 g/kg or 2.0 g/kg (10% w/v) alcohol, or a saline dose equivalent to the volume of a 1.0 g/kg dose of alcohol. Animals were first tested on the RR at 10 min post ethanol or saline injection, and ARR was assessed 20 min post ethanol or saline injection. Three separate trials were conducted 3 days apart to minimize carryover effects. The first test session occurred 24 h after the last RR training trial (see below). Animals were randomly assigned to each of the 3 different drug orders and counterbalanced by age and dose for all 3 trials. Seven days after the last testing trial, a subset of animals ($n = 4$ per age group) were given an acute injection of 3.0 g/kg alcohol and loss of righting (LORR) was determined. Finally, a separate group of adolescent, adult, and aged animals ($n = 6$ per age) were injected i.p. with 3.0 g/kg ethanol. The tail was nicked and blood was collected for BEC analysis via the Analox AM1 protocol 30 min, 60 min, 120 min, 180 min, 240 min, 300 min, and 360 min post-injection.

Aerial righting reflex (ARR)

The effect of acute alcohol exposure on gross motor impairment was assessed 20 min after saline or alcohol administration by ARR as previously described (Novier et al., 2013; Van Skike et al., 2010). The 3 test trials occurred on PD 31, PD 35, and PD 39 in adolescent rats, and on PD 73, PD 77, and PD 81 in adult rats. Aged animals are classified as approximately 18 months old by the supplier and therefore exact ages are unknown. A ruler was vertically taped above a 10-inch foam pad. Animals were initially released 5 inches (12.7 cm) above the foam pad in a supine position. An animal's righting reflex was considered successful if 3 out of 4 paws made direct contact with the foam pad on 2 out of 3 releases. If righting was not successful, the height of release was increased in 5-inch (12.7 cm) increments up to a maximum height of 25 inches (63.5 cm). Subjects who failed to achieve successful righting reflex at 25 inches (63.5 cm) were given a score of 30 inches (76.2 cm) for statistical analysis.

Accelerating rotarod (RR)

Apparatus

RR was used to investigate the effects of alcohol exposure on motor coordination. Motor activity was tested on a 4-station Rota-Rod treadmill (Model ENV 575, Med Associates, St. Albans, VT). The apparatus was located in a behavioral room isolated from animal caging and housing.

Training

Subjects received 5 consecutive training trials on the RR, as previously described (Novier et al., 2013). Adolescents received RR training on PD 30 and adults on PD 72. The rod accelerated from 4 rpm to 40 rpm over a 5-min period. The rotarod is covered with