Pharmacology, Biochemistry and Behavior 150–151 (2016) 124–133

Contents lists available at ScienceDirect

Pharmacology, Biochemistry and Behavior

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

Appetite and weight gain suppression effects of alcohol depend on the route and pattern of administration in Long Evans rats

Nnamdi G Nelson [a], Faten A Suhaidi [b], Ross S DeAngelis [c], Nu-Chu Liang [a,b,*]

[a] Neuroscience Program, University of Illinois at Urbana-Champaign, USA
[b] Department of Psychology, University of Illinois at Urbana-Champaign, USA
[c] Program for Ecology, Evolution and Conservation Biology, University of Illinois at Urbana-Champaign, USA

ARTICLE INFO

Article history:
Received 28 June 2016
Received in revised form 14 October 2016
Accepted 18 October 2016
Available online 20 October 2016

Keywords:
Appetite
Food intake
Diet preference
Body weight
Ethanol injection
Voluntary ethanol drinking
Blood ethanol concentration

ABSTRACT

Ethanol can be a food source but its effects on energy balance and contribution to obesity remain inconclusive. In this study, we hypothesized that the effects of ethanol on energy intake and body weight would depend on the administration dose, pattern and the blood ethanol concentration (BEC) time-course. Experiment 1 examined changes in food intake, diet preference, and body weight after saline or ethanol (1 and 3 g/kg) injection (IP). Experiment 2 compared the effects in rats that received either 3 g/kg/day ethanol administered all at once (EtOH_S) or 2 1.5 g/kg injections spaced by 3 h (EtOH_D). Experiment 3 examined the effects of 7.5 h/day, Mon through Fri for 8 weeks, voluntary ethanol drinking (5% and 10% ethanol) on food intake and body weight. Results of Experiments 1 and 2 indicate that acute ethanol administrations dose-dependently reduced energy intake, high fat diet preference and weight gain. Acute 3 g/kg ethanol injection in the EtOH_S group decreased energy intake, weight gain and visceral fat to a greater extent than in the EtOH_D group. Results of Experiment 3 show that male and female rats voluntarily drank 1.65–2.31 g/kg ethanol within 3.5 h with reduced chow intake but unchanged total energy intake and weight gain. Furthermore, 3 g/kg ethanol injection resulted in BEC that remained at intoxicating levels e.g. >120 mg/dL for several hours post-administration and was higher in the EtOH_S than in the EtOH_D group. In contrast, BEC in voluntarily drinking was ~67 mg/dL and decreased to below 10 mg/dL 5 h after termination of ethanol access. Taken together, these data suggest that 3 g/kg ethanol injection robustly suppresses appetite and weight gain due to the higher BECs attained. Furthermore, BEC attained and maintained is a determining factor for how ethanol administration affects appetite and long-term energy balance.

1. Introduction

Biochemical analyses have shown that the caloric value of ethyl alcohol (7 kcal/g) is intermediate between that of carbohydrate (4 kcal/g) and fat (9 kcal/g) (reviewed in Suter, 2005; Yeomans and Phillips, 2002). Most social alcohol drinkers in the United States and in other developed countries are often not cognizant of the calorie content of ethanol when they consume alcoholic beverages with or without meals (Suter, 2005; Yeomans, 2010). On social drinking occasions, calories derived from alcoholic beverages can comprise 1–27% of the recommended daily calorie intake in men and women (Shelton and Knott, 2014; Bebb et al., 1971; Jones et al., 1982; Ferro-Luzzi et al., 1988). Research has shown that ethanol can have stimulatory effects on appetite (Caton et al., 2004; Yeomans et al., 1999). Because most drinkers do not reduce their food intake accordingly to compensate for the extra calories supplied by ethanol, these extra calories tend to be additive to their regular daily energy intake and can favor positive energy balance (Yeomans and Phillips, 2002; Bebb et al., 1971; Jones et al., 1982; Ferro-Luzzi et al., 1988; Caton et al., 2004; Yeomans et al., 1999; Colditz et al., 1991; Fisher and Gordon, 1985; Hetherington et al., 2001; Levine et al., 2000; Poppitt et al., 1996; Tremblay et al., 1995; Westerterp-Plantenga and Verwegen, 1999; de Castro and Orozco, 1990; Caton et al., 2007; Mattes, 1996). In contrast, ethanol can reduce rated hunger or appetite (Yeomans et al., 1999; Westerterp-Plantenga and Verwegen, 1999), and some human drinkers can substitute portions of their daily calorie needs with ethanol (Ferro-Luzzi et al., 1988; Caton et al., 2004; Hetherington et al., 2001; Foltin et al., 1993) without increasing total daily caloric intake (Clevidence et al., 1995).

The popularity of ethanol coupled with the high global incidence of overweight and obesity (Gearhardt and Corbin, 2009; Finkelstein et al., 2012) sparked interest in understanding the contribution of ethanol to energy homeostasis and weight maintenance (Suter, 2005; Yeomans, 2010; Shelton and Knott, 2014). Previous studies in healthy human subjects have arrived at equivocal conclusions concerning this link (reviewed in Yeomans, 2010; Traversy and Chaput, 2015). Moderate

* Corresponding author at: Department of Psychology, University of Illinois at Urbana Champaign, 513 Psychology Bldg., 603 E. Daniel Street, M/C 716, Champaign, IL 61820, USA.
E-mail address: ncliang8@illinois.edu (N.-C. Liang).

http://dx.doi.org/10.1016/j.pbb.2016.10.006
0091-3057/

ethanol use can correlate with decreased (Jones et al., 1982; Colditz et al., 1991; Fisher and Gordon, 1985; Gearhardt and Corbin, 2009; Liu et al., 1994) or increased (Bebb et al., 1971; Ferro-Luzzi et al., 1988; Arif and Rohrer, 2005; Breslow and Smothers, 2005; Kromhout, 1983; Wannamethee et al., 2005) weight gain or BMI in adult females and males. A clinical study showed that isocaloric substitution of ethanol for carbohydrate in the diet of male and female adult subjects resulted in a significant weight loss whereas long-term addition of ethanol to daily diet did not result in significant weight gain (Pirola and Lieber, 1972). On the other hand, ethanol intake by obese, but not lean, men and women was shown to contribute to additional weight gain (Crouse and Grundy, 1984). Notwithstanding, there exist reports of no association between ethanol use and BMI in either gender (Yeomans, 2010; Jones et al., 1982; Colditz et al., 1991; Levine et al., 2000; Liu et al., 1994; Cordain et al., 1997). It is noteworthy that most findings from human studies are mere correlation and do not imply a cause-effect relationship. It is difficult to manipulate some important variables in human research (Suter, 2005). In light of the potential confounds associated with human studies coupled with the possibility of both intentional and unintentional biases in self-reports (Jones et al., 1982; Colditz et al., 1991; Fisher and Gordon, 1985; Mattes, 1996; Foltin et al., 1993; Gearhardt and Corbin, 2009; Caetano, 1998), more work needs to be conducted to precisely identify the contribution of alcoholic beverages to appetite and body weight control (Yeomans, 2010; Shelton and Knott, 2014; Cordain et al., 1997).

Limitations with human studies warrant careful investigations in rodent models. The rodent literature is also replete with equivocal reports. Some studies have documented that ethanol consumption suppresses food intake (Giner and Meguid, 1993; Strbak et al., 1998; Benicky et al., 2000; Richardson et al., 1990; Richter, 1926; Wolffgramm, 1990) and total energy intake (Reidelberger et al., 1996) or does not change food intake (Gill et al., 1996) and total energy intake (Strbak et al., 1998; Benicky et al., 2000; Richardson et al., 1990; Richter, 1926, 1941; McCoy et al., 1981). Others have observed that rats did not show compensatory reduction in food intake to account for ethanol-derived calories and increase in total energy intake as a result (Giner and Meguid, 1993). Additionally, there are also findings of decrease (Strbak et al., 1998; Benicky et al., 2000; Reidelberger et al., 1996; McCoy et al., 1981; Broadwater et al., 2014; Sherrill et al., 2011; Mendenhall et al., 1993) or no change (Richardson et al., 1990; Richter, 1926, 1941; Wolffgramm, 1990; Mendenhall et al., 1993; Kuzmin et al., 2012; Schulteis et al., 1996; Froehlich et al., 1988) in body weight as a result of ethanol administration. In those studies, dose dependent effects and the pattern of ethanol administration were not examined. In most studies, animals were forced to consume ethanol via ethanol-containing liquid diet (Reidelberger et al., 1996; Mendenhall et al., 1993; Schulteis et al., 1996) or ethanol solution (Strbak et al., 1998; Benicky et al., 2000; Richardson et al., 1990; Richter, 1926, 1941; McCoy et al., 1981) respectively as the sole food or water source. Ethanol also was administered involuntarily to the rodents by intraperitoneal injection (Sherrill et al., 2011) (IP), intragastric infusion (Giner and Meguid, 1993; Broadwater et al., 2014; Kuzmin et al., 2012) or intravenous infusion (Giner and Meguid, 1993). Furthermore, intragastric infusion of ethanol differently reduces food intake in rats to a greater extent than intravenous infusion did (Giner and Meguid, 1993). In that study, however, the routes of administration compared were both involuntary modes and blood ethanol concentration (BEC) was not measured. Taken together, it is possible that dose, pattern, and route of ethanol administration differently affect appetite and energy balance in rodents. Accordingly, we hypothesized that ethanol will lead to dose-dependent changes of food intake and body weight, and the extent of changes will rely on the administration pattern and the BEC time-course. In Experiment 1, dose-dependent effects were examined by IP injections of 1 and 3 g/kg ethanol in male rats. In previous studies, only one diet was available and ethanol's effects on diet choice have not been examined. Thus, rats were maintained on a chow and a high fat (HF) diet to examine ethanol's effects on HF diet preference in this experiment. To determine whether the effect of IP ethanol on body weight is a direct effect of ethanol or secondary to changes in food intake, we included a pair-fed group whose daily calorie intake was equal to that of those receiving the IP ethanol treatments. Experiment 2 assessed the effects of two different patterns of 3 g/kg ethanol injections, all at once (EtOH_S) or in 2 separate 1.5 g/kg injections that were spaced 3 h apart (EtOH_D), in rats maintained on a chow diet. Finally, to compare with the effects of involuntary IP ethanol injection, Experiment 3 examined changes in food intake and body weight in rats voluntarily drinking sweetened ethanol solutions (Roberts et al., 1999). BEC was measured in Experiments 2 and 3 to determine the roles of BEC attained on ethanol associated changes in appetite and body weight.

2. Materials and methods

2.1. Experiment 1: dose dependent effects of intraperitoneal ethanol injections

2.1.1. Subjects and housing

A total of 26 male Long Evans rats (weighing 190–212 g) were obtained from Harlan (Indianapolis, IN, USA). Animals for a pair-fed control group ($n = 7$) were received a week after the arrival of the first cohort ($n = 19$). Rats were individually housed in conventional laboratory cages under a 12-h light, 12-h dark cycle, with lights on at 10:30 AM. The colony was maintained in a room with controlled temperature (22–25 °C) and humidity. Animals had ad libitum access to tap water and standard rodent chow diet (3.1 kcal/g, 24% protein, 58% carbohydrate, 18% fat from soybean oil; Harlan 2018, Madison, WI). All procedures were approved by the IACUC at the University of Illinois at Urbana-Champaign and conformed to the guidelines stipulated by the National Institute of Health in the *Guide for the Care and Use of Laboratory Animals*.

2.1.2. Saline and ethanol for IP injection

Ethanol solutions were prepared (v/v) by diluting 200 proof (100%) ethyl alcohol (Decon™ Labs, Inc., King of Prussia, PA) with saline. To minimize injection volume, doses of 1 g/kg and 3 g/kg ethanol were respectively administered using room-temperature 20% and 30% ethanol solutions. In an effort to model the timing of human alcohol consumption, which typically occurs around dinner time or before the inactive circadian cycle (Clevidence et al., 1995), all injections occurred between 1.5 and 2 h before light onset (i.e., the last couple hours of the dark cycle, between 8:30 and 9:00 AM).

2.1.3. Procedures

Following habituation to the housing facility, the rats were divided into 3 groups by matching average body weights: saline control (SAL, $n = 9$), ethanol-treated (EtOH, $n = 10$), and pair-fed control (PF, $n = 7$). During the first week (Mon–Fri), all animals in the EtOH, PF and SAL groups received isovolumetric injections of saline every other day to acclimate them to being handled and injected. Subsequently, the EtOH group received intermittent injections of 1 g/kg ethanol every other day for 2 weeks (for a total of 6 injections) followed by a single 3 g/kg ethanol injection that was administered 3 days after the last 1 g/kg injection. The SAL and PF rats received saline injections at the time the EtOH rats were injected. Initially, animals had unrestricted access to chow and tap water, and at the commencement of the second week of 1 g/kg ethanol injections a 45% high-fat diet (4.73 kcal/g, D12451, 20% protein, 35% carbohydrate, 45% fat from soybean oil; Research Diets Inc., New Brunswick, NJ) was introduced. During diet choice, the positions of the food hoppers were alternated daily to prevent the development of side preference. After the saline habitation week, rats in the PF group were fed the daily average chow and HF diet intake by the EtOH group, with calories from ethanol substituted