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Environmental and immune stressors enhance alcohol-induced motor ataxia in rat

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Abstract

Infection is now accepted as a stressor, consequently we sought to compare the short- and longer-term consequences of several environmental stressors versus an endotoxin challenge on alcohol-induced motor ataxia. The present set of studies examined the impact of intermittent electric shock (SHOCK), intermittent cold water swim (ICWS), or lipopolysaccharide (LPS) administration on the motor ataxic effects of an intraperitoneal (i.p.) injection of alcohol (ETOH). In Experiment 1 SHOCK, but not ICWS, enhanced the motor ataxic effects of ethanol at both 2 and 24 h post-stress. In Experiment 2 administration of LPS did not affect the motor ataxic effects of ETOH 4 h later, but enhanced the ataxic potency of ETOH 24 h later. The results indicate that certain environmental and immune stressors have the potential to alter the long-term behavioral reactivity to alcohol. These examples of stress-induced enhancement of the motor ataxic effects of ETOH may have important implications for the development of alcohol dependence.

Keywords: Shock; Swim; Stress; Sickness; Endotoxin; Rotarod; Intoxication

1. Introduction

Alcohol abuse and addiction are very serious worldwide health problems. Alcoholism is precipitated by a variety of factors with key links between genetics and environment. Some individuals have a genetic predisposition to alcoholism and this combined with environmental triggers can lead to alcoholism (Charness et al., 1989). Environmental stress exposure is known to play a role in the initiation of alcohol abuse (Brady and Sonne, 1999).

In addition to stress influencing the initiation of alcohol consumption (i.e. self-medication), there is good reason to suspect that recent stress history may influence an organisms reaction to this central nervous system depressant. More specifically, environmental stress exposure changes both brain neurochemistry and endocrine systems which may alter the pharmacodynamic actions of alcohol (De Kloet, 2000; McEwen, 1994; Weiss et al., 1981). Alcohol produces its anxiolytic and motor ataxic effects by acting on several neurotransmitter receptors including gamma aminobutyric acid (GABAa), NMDA/glutamate and serotonin (5-HT) sites (Grant, 1994). Stress and alcohol share a final common pathway by affecting these similar neurotransmitter systems (Brady and Sonne, 1999). The convergence of these effects of stress and alcohol on the same neural systems may alter the behavioral response to alcohol in organisms with a recent stress history.

The relationship between stress and alcohol reactivity has been clearly demonstrated, particularly in animal models (see reviews by Pohorecky, 1981, 1990). For example, mild stressors such as handling can intensify the acute effects of ETOH on such indices as heart rate and body temperature (Peris and Cunningham, 1986). The psychosocial stress of subordination is also associated with an increased reactivity to the depressant

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effects of ETOH in rats (Blakely and Pohorecky, 2006). In humans, there is also a connection between stress and the use of alcohol or other drugs (AODs). The current explanation for stress enhancing AOD use is that it increases the motivational and/or reinforcing effects of various AODs at the neurochemical level (e.g., CRH and dopamine system — Brady and Sonne, 1999; Kreek and Koob, 1998; Piazza and Le Moal, 1996). However, there are reports in humans where chronic stress may have the opposite effect (i.e., sobering) when examining subsequent acute stress and alcohol reactivity (Breslin et al., 1995). Our lab has previously demonstrated that psychological dynamics of stress (i.e., controllability) increases the hypnotic and ataxic properties of alcohol (Drugan et al., 1992; Drugan et al., 1996), as well as B-carboline-induced stress (Austin et al., 1999). These effects were demonstrated either immediately or 2 h post-stress, but the longer-term effects of stress on alcohol reactivity are currently unknown.

The fact that the acute phase response stimulates the major stress axes has been appreciated since the initial work of Besodovsky and colleagues showing an increase of circulating ACTH and glucocorticoids in response to sheep red blood cells (Besodovsky et al., 1981). Recent evidence has replicated and extended this initial finding by showing that infection and sickness behavior stimulates the identical endocrine stress response systems as do environmental stressors (Deak et al., 1997; Dunn, 1995; Maier and Watkins, 1998). The mechanism of action for these effects has been elucidated in that proteins released by activated immune cells, cytokines, alter various neurotransmitter systems such as the biogenic amines in a way that resembles an organism's stress response (Dunn and Wang, 1996). This has led to the belief that infection by itself is a stressor. The present study aimed to test the short- and long-term effects of either environmental or endotoxin stress on subsequent alcohol-induced motor ataxia in an effort to evaluate the time course and external validity of our previous findings.

There is a bidirectional relationship between LPS administration and alcohol. Alcohol changes the immune response to LPS exposure, but LPS administration also alters the endocrine response to an alcohol challenge (Rivier, 1993, 1999). In their study, LPS was administered to intact and adrenalectomized rats to determine the effects of LPS on alcohol reactivity. ACTH, and the release of the cytokines IL-6 and TNF-alpha was determined following LPS and alcohol administration together. As expected, LPS, alcohol, and cytokine administration increased ACTH release when given alone. However, prior exposure to LPS attenuated subsequent alcohol-induced ACTH release (Rivier, 1993). Conversely, alcohol pretreatment potentiated LPS-induced corticosterone release, while reducing the IL-6 concentration in plasma. However, alcohol was without effect on the LPS potentiation of TNF and IL-6 in pituitary gland, adrenal gland or spleen (Rivier, 1999). Although this demonstrates a clear influence of bacterial infection on endocrine reactivity to alcohol, the consequences for alcohol-induced behavioral changes have not been evaluated. Therefore, if prior LPS administration blunts subsequent alcohol-induced ACTH release, then the motor ataxic effects of alcohol should also be reduced. To our knowledge the current study is the first to investigate the behavioral reactivity to alcohol following an LPS-induced immune challenge. In addition, we monitored body weight and food and water intake to determine if alcohol exposure altered the trajectory of sickness behavior (Kent et al., 1992a).

2. Methods

2.1. Subjects

112 male Sprague–Dawley rats were obtained from Charles River Laboratories (Boston, MA). Rats weighed 180–200 g upon arrival and were housed 4/cage with free access to food and water in a vivarium that maintained a 12/12 h light/dark cycle with lights on at 0600 h. Rats were allowed to acclimate to the vivarium for at least 7 days before treatment. All behavioral procedures were reviewed and approved by the University of New Hampshire Institutional Animal Care and Use Committee (IACUC).

2.2. Apparatus

Intermittent tailshocks were administered in acrylic boxes (15.5 × 12 × 17 cm) similar to those used by other investigators with a locked wheel or no wheel (Drugan et al., 1989; Maier et al., 1973; Weiss et al., 1970). Copper electrodes were attached to the tail with cloth tape and augmented with electrode paste (Redux, HP). The tail was then taped to an acrylic post extending from the rear of the chamber. 1 mA shocks were delivered by Lafayette Instrument shock generators (Model 82400) and controlled by a computer. Restrained rats were placed in the same acrylic chambers but were not attached to a shock generator.

Intermittent swim stress was administered in acrylic cylinders (21 × 42 cm — dia. × *H*) with 37, 1.2 cm holes in the bottom, suspended over a tank of 15 °C water. On a swim trial, the cylinders would be lowered into the water to a depth of 20 cm. Space heaters blew warm air (36 °C) into the cylinders during the inter-trial intervals (ITI). Confined rats were placed in a shorter cylinder (21 × 15 cm — dia. × *H*) divided into two equal hemispheres with ventilating holes on the floor and lid. Confined rats moved with the swim rats on a swim trial, but never entered the water (Brown et al., 2001).

Motor ataxia was assessed on a Rotarod treadmill (6 × 35 cm — dia. × *L*) that has been used previously in our laboratory (Drugan et al., 1996). The Rotarod was divided into 4 equal sections by opaque plastic discs, and rotated at a constant speed of 10 rpm (Ugo Basile Biological Research Apparatus, 21025 Comerio, Varese, Italy, Model 7700).

2.2.1. Drugs

LPS (*Escherichia coli*, serotype: 0111:B4) was obtained from Sigma Chemical Co., (St. Louis, MO). LPS 25 μg/kg; ETOH 1 g/kg (Experiment # 1) and 1.5 g/kg (Experiment # 2), or equal volume of physiological saline were administered intraperitoneally (i.p.).

2.3. Procedure

2.3.1. Experiment # 1

On the first day, 80 male rats were weighed and randomly assigned to one of 8 treatment groups (10 rats/group): either 2 or 24 h post-stress and SHOCK, RESTRAINED, SWIM (ICWS), or CONFINED treatment groups (see below) in a 2×4 (time×stress) design. All rats were given a criterion test on the Rotarod. The criterion test required that each rat walk on the Rotarod continuously for 2 min. A naïve home cage control group was not included in the current experimental design because prior work indicated no difference in alcohol-induced motor ataxia between restrained versus home cage control groups at either 3½ or 24 h post-treatment (Austin et al., 1999).

Stress treatment began after the criterion test. Rats in the SHOCK group received 80, 5 s tailshocks administered on a variable time 60 s schedule (VT-60). Rats in the RESTRAINED group were placed in the shock apparatus for equal time but did not receive shock. ICWS stress consisted of 80, 5 s exposures to 15 °C water. Swim trials were also administered on a VT-60 schedule (Christianson and Drugan, 2005). CONFINED rats were placed in the shorter cylinder; which moved on the swim trial but never entered the water. After 80 swim trials ICWS and CONFINED rats were gently wrapped in a towel and then placed under incandescent warming lamps for 30 min.

Rats in the 2 h group were given a second 2 min Rotarod criterion test 105 min after stress treatment. After the test all rats were injected with 1 g/kg ETOH (i.p.). 12 min later rats were given the Rotarod test. This time interval was used to match our previous papers evaluating the impact of stress on motor ataxia (Austin et al., 1999; Drugan et al., 1996). Rats in the 24 h group were given a criterion test at 23 h post-stress and then administered ETOH or saline in a counterbalanced order at a time as close to the 24 h time point as possible. One at a time, a rat was placed in a lane on the Rotarod. An observer, blind to group membership, recorded the time the rat spent walking on the Rotarod. Rats received 3 test trials; the maximum trial duration was set at 300 s. If a rat remained on the Rotarod for more than 180 s on the first trial, and 300 s on the second trial, no third trial was given. Two trials were conducted only in the saline groups because of the effective ataxic doses of ethanol employed. This criterion was in place to insure that all rats were tested exactly 12 min after ETOH injections.

2.3.2. Experiment # 2

After the acclimation period of 7 days, all 32 male rats were individually housed. On this day, all rats were weighed, and given 30 g of rat chow pellets, and 100 ml of water in graduated cylinders. Four days prior to the experiment, body weight, food and water intake were measured to obtain baseline scores. Each day, food was weighed and rats were given 30 g again and the graduated cylinder tubes were refilled to 100 ml. On experiment day 1, all animals were weighed and trained to a criterion of 2 min of continuous walking on the Rotarod without falling off, prior to the commencement of any experimental procedures. During the criterion testing if the rat fell off before the 2 min period had elapsed, it was immediately placed back on the Rotarod and the timer was restarted. This criterion testing procedure has been used in past studies to ensure that the rats are capable of running on the Rotarod (Drugan et al., 1996; Austin et al., 1999).

Rats were then randomly assigned to one of four groups (8 rats/group): LPS–ETOH, LPS–Saline, Saline–ETOH and Saline–Saline. Rats were given an injection of LPS (25 μg/kg) or equal volume saline. This dose of LPS as well as the time frame (4 h post-injection for Rotarod testing) was chosen based on previous reports indicating that measurable cytokines (e.g. IL-1 and IL-6) and stimulation of the hypothalamic–pituitary–adrenal (HPA) axis were observed at that time (Bristow et al., 1991; Rivier, 1993; Shalaby et al., 1989). All rats were then returned to their home cages. Four hours later, all groups were given a post-injection criterion test on the Rotarod before ethanol or saline injections. Rats were then injected with 1.5 g/kg of ETOH, an effective dose to induce ataxia in animals handled for 4 days prior to experimentation (R.C. Drugan, unpublished observations) or equal volume saline solution. This dose was also the amount used in the study evaluating the impact of LPS on ethanol-induced activation of the HPA axis (Rivier, 1993). In addition, the time frame of 4 h was chosen based on previous time course data indicating that LPS altered alcohol's actions on the HPA axis at this point in time (Rivier, 1993). Also, the immune response to LPS injection is typically observed to peak at 2–6 h post-injection (Kent et al., 1992b; Luheshi and Rothwell, 1996; Romanovsky et al., 1998).

Rats were then tested for motor ataxia 10 and 60 min after the injection by an experimenter blind to group membership. This time course was chosen based on previous work that tested alcohol-induced blood and brain changes (Rivier, 1993). On experimental day 2, 24 h post-LPS or saline injections, all groups were weighed, and food and water intake was recorded. All rats were given a third criterion test on the Rotarod. On this day, all groups received an i.p. injection of 1.5 g/kg of ETOH. They were then tested 10 and 60 min post-ethanol injection for motor ataxia. For 4 days following the initial experimental procedure, daily body weight, food and water intake were examined to evaluate sickness behaviors.

2.4. Statistical analysis

In Experiment # 1, mean time on the Rotarod was analyzed by a 2 (2 h or 24 h)×4 (SHOCK, ICWS, RESTRAINED or CONFINED) between subjects Analysis of Variance (ANOVA). For Experiment # 2, mean time on Rotarod (on day 1) was analyzed with a 2 (LPS or Saline)×2 (ETOH or Saline)×2 (10 min and 60 min) ANOVA with LPS and ETOH as between-subject factors and time as a within-subjects factor. Mean time on Rotarod on day 2 was analyzed with a 4 (Day One Treatment: LPS–ETOH, LPS–Saline, Saline–ETOH, or Saline–Saline)×2 (10 and 60 min) ANOVA with day one treatment as a between-subjects factor and time as a within-subjects factor. Data were

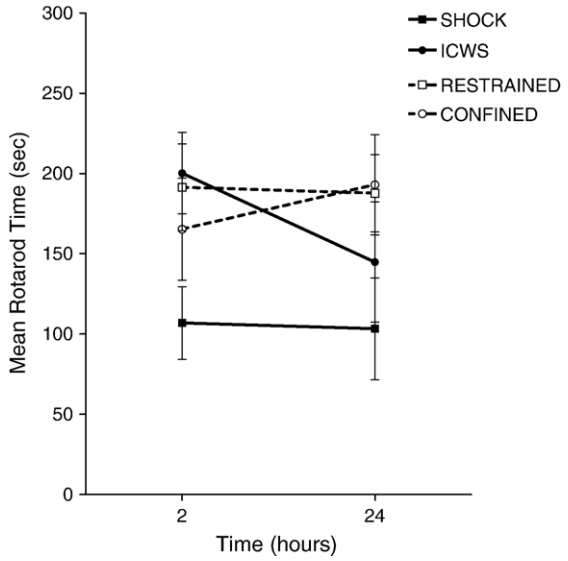

Fig. 1. Mean (+/−SEM) time on Rotarod 2 or 24 h after stress exposure. Rotarod performance was assessed 2 min after administration of an ataxic dose of ETOH. IS significantly reduced time on Rotarod compared to all other groups at both 2 and 24 h (Newman–Keuls, $p<0.05$).

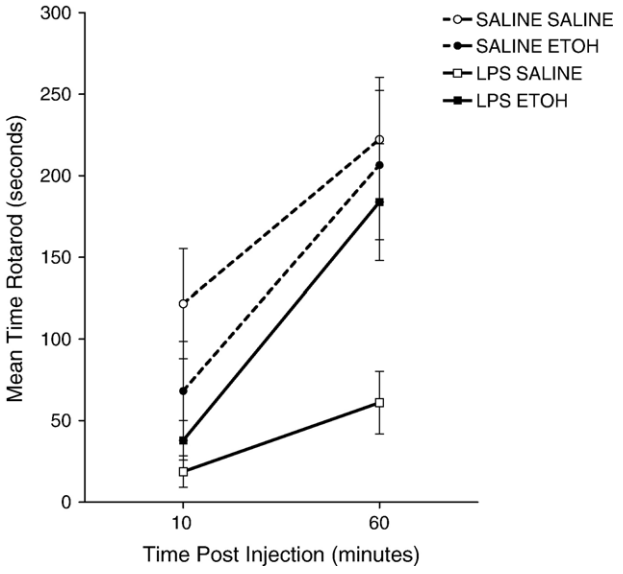

Fig. 3. Mean (+/−SEM) time on Rotarod 10 and 60 min after an ataxic dose of ETOH for all groups. ETOH was administered 24 h after LPS or Saline treatment (labels indicate day 1 treatments prior to Rotarod test on day 1). ETOH significantly reduced Rotarod time in rats previously exposed to LPS without ETOH (LPS–Saline) at the 60 min delay.

analyzed this way because in this case, all subjects received an ataxic dose of ETOH regardless of day one treatment. Mean body weight was analyzed as a 2 (LPS or Saline)×2 (ETOH or Saline)×5 (Days 1–5) ANOVA with LPS and ETOH as between-subjects factors and Days as a within-subjects factor. Mean water intake was analyzed as a 2 (LPS or Saline)×2 (ETOH or Saline)×5 (Days 1–5) ANOVA with LPS and ETOH as between-subjects factors and Days as a within-subjects factor. Mean food intake was analyzed as a 2 (LPS or Saline)×2 (ETOH or Saline)×4 (Days 1–4) ANOVA with LPS and ETOH as between-subjects factors and Days as a within-subjects factor.

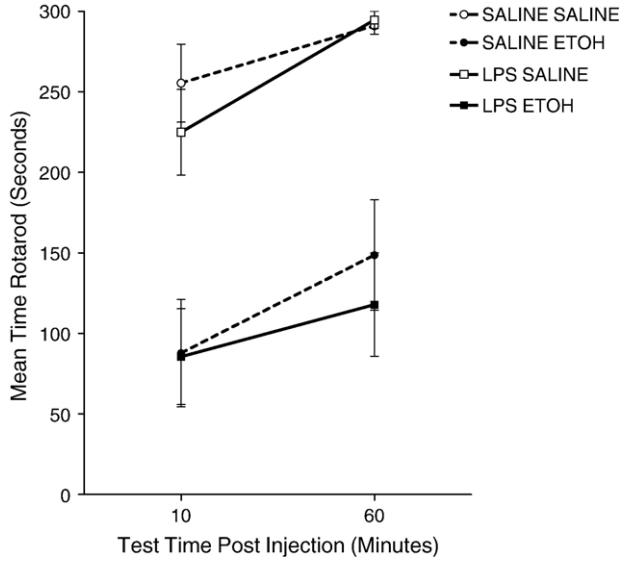

Fig. 2. Mean (+/−SEM) time on Rotarod 10 and 60 min after ataxic dose of ETOH or Saline. ETOH or Saline was administered 4 h after LPS or Saline pretreatment. ETOH significantly reduced Rotarod time in both LPS and Saline pretreated rats ($p<0.05$) at both 10 and 60 min delays.

3. Results

3.1. Experiment # 1

A significant main effect on Rotarod time was found for stressor, $F(3, 72)=3.42$, $p=0.022$, but not time, $F(1, 72)=0.11$, $p=0.74$ or the stress×time interaction, $F(3, 72)=0.72$, $p=0.56$. Student–Newman–Keuls post hoc analysis revealed that SHOCK reduced Rotarod time when compared to all other stress conditions, ($p<0.05$); the remaining groups did not differ from each other (Fig. 1).

3.2. Experiment # 2

No main effect on Rotarod time was observed for LPS treatment, $F(1, 28)=0.41$, $p=0.54$. However, ETOH significantly reduced time on the Rotarod, $F(1, 28)=45.10$, $p<0.001$. There was also a significant main effect for time ($F(1, 28)=16.23$, $p<0.001$) and post hoc analysis determined that Rotarod time was increased at 60 min ($p<0.05$). There were no significant interactions, $p>0.20$ (Fig. 2).

A significant main effect on Rotarod time was found for treatment on day one, $F(3, 28)=4.72$, $p=0.009$. Student–Newman–Keuls post hoc comparisons revealed that LPS–Saline produced significantly lower Rotarod times than Saline–ETOH and Saline–Saline, $p<0.05$, but did not differ from LPS–ETOH. There was also a significant main effect for time ($F(1, 28)=41.62$, $p<0.001$) with post hoc tests determining that subjects spent more time on the Rotarod at the 60 min time point

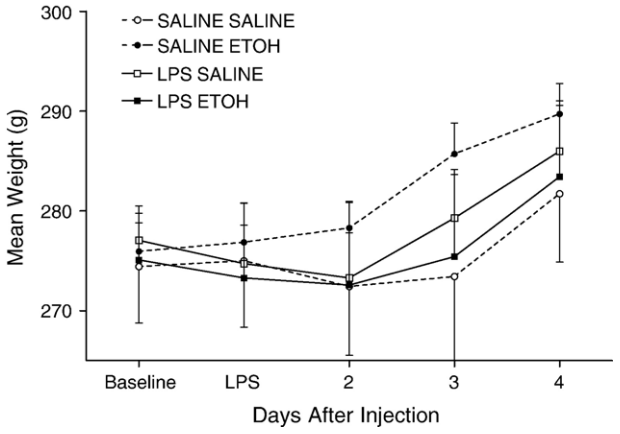

Fig. 4. Mean (+/−SEM) body weight for rats several days prior to the experiment (baseline), during, and following one of the 4 treatment conditions (Saline–Saline, Saline–ETOH, LPS–Saline or LPS–ETOH on day 1 of experimentation) followed by an ETOH injection 24 h later.

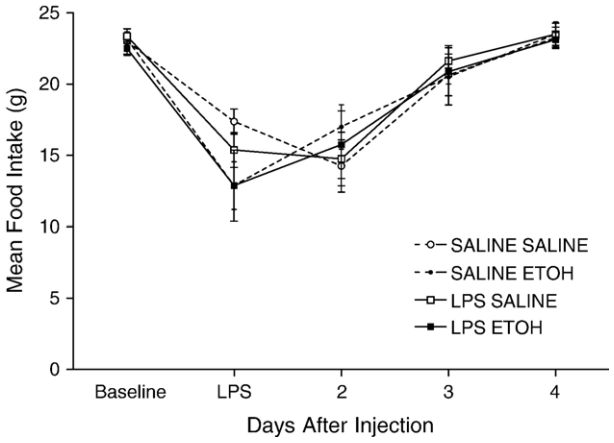

Fig. 6. Mean (+/−SEM) food intake for rats several days prior to the experiment (baseline), during and following one of the 4 treatment conditions (Saline–Saline, Saline–ETOH, LPS–Saline or LPS–ETOH on day 1 of experimentation) followed by an ETOH injection 24 h later.

($p<0.05$). No significant interaction was found, $F(3, 28)=2.05$, $p=0.13$ (Fig. 3).

The only significant main effect on body weight was for days, $F(4, 96)=19.21$, $p<0.001$. Serial post hoc contrasts found that body weight increased significantly from day 2 to 3 and from day 3 to 4, $p<0.01$. No significant effects were found for LPS, ETOH, or their interactions, $p>0.10$ (Fig. 4).

ANOVA on water intake data indicated that the only main effect was due to days, $F(4, 112)=45.91$, $p<0.001$. Serial post hoc comparisons found that water intake reduced from Baseline to LPS ($p<0.001$) then increased from day 2 to 3 and from 3 to 4 ($p<0.01$). No significant treatment effects were found for LPS or ETOH and nor were there any significant interactions, $p>0.05$ (Fig. 5).

Only a significant main effect on food intake was found for days, $F(4, 112)=45.91$, $p<0.001$. Serial post hoc contrasts found significant increases from LPS to day 1, day 1 to 2, and day 2 to 3, $p<0.01$. No LPS or ETOH main effects were found and no interactions were significant, $p>0.05$ (Fig. 6).

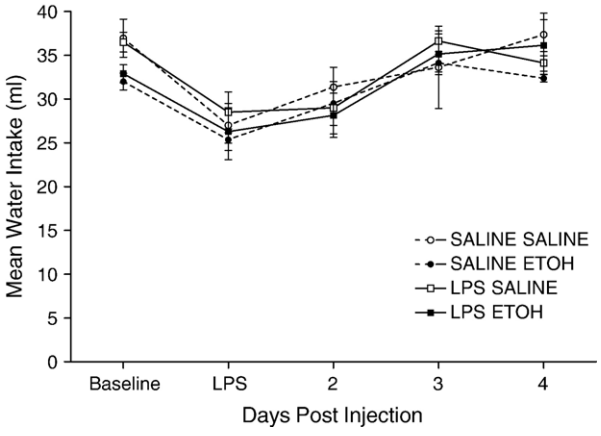

Fig. 5. Mean (+/−SEM) water intake for rats several days prior to the experiment (baseline), during, and following one of the 4 treatment conditions (Saline–Saline, Saline–ETOH, LPS–Saline, or LPS–ETOH on day 1 of experimentation) followed by an ETOH injection 24 h later.

4. Discussion

The results from the studies are clear. Intermittent inescapable tailshock exposure causes a significant increase in the ataxic potency of alcohol at both 2 and 24 h post-stress. This replicates and extends our previous observations of a short-term enhancement of alcohol's hypnotic and ataxic potency 2 h following inescapable but not escapable shock stress (Drugan et al., 1992; Drugan et al., 1996). Shock exposure causes a long-term enhancement of motor ataxic effects that is observed 24 h later, while intermittent cold water swim stress does not impact the behavioral reactivity to alcohol at either 2 or 24 h post-stress. Therefore, although ICWS does produce certain similar behavioral (i.e. increased immobility in the FST, Christianson and Drugan, 2005; Prince and Anisman, 1984), and endocrinological (i.e. significant increase in CORT, Drugan et al., 2005; Maier et al., 1986) effects to that seen with IS exposure, it diverges from shock stress concerning it's influence on the behavioral reactivity to alcohol. The restrained and confined controls employed in the current study were chosen because we thought that similar handling without swim or shock stress was the optimal control condition. Importantly, we have shown that there is no difference in the motor ataxic properties of alcohol between restrained versus naïve home cage controls either 3½ or 24 h following treatment (Austin et al., 1999).

LPS administration at the dose employed has no significant effect on alcohol reactivity when evaluated 4 h post-injection. However, when observed 24 h later, it shows a similar effect to that of IS by increasing the motor ataxic properties of alcohol. The dose of LPS and alcohol used in the current study were based on previous work indicating that 25 μg/kg was sufficient to alter the HPA response to 1.5 g/kg of alcohol 4 h later (Rivier, 1993). We did not observe any changes in the motor ataxic effects of alcohol by prior LPS exposure at this point in time in

comparison to saline-treated controls. However, when all groups were given an alcohol injection 24 h later, animals treated with LPS on day one, but not given alcohol (i.e. LPS–Saline), displayed a greater ataxic response than any of the other treatment groups 24 h later. The LPS–ETOH group does not show such a marked alteration which may be due to a tolerance to the ataxic effects of alcohol due to the prior exposure on day 1.

These data extend the observations of others suggesting that shock stress may activate the acute phase response in a similar way to sickness behavior (Deak et al., 1997). We observe the same long-term enhancement of alcohol's effects following both IS and LPS exposure. To our knowledge, this is the first report of a potentiation of alcohol's behavioral actions by endotoxin. It is unclear whether the same final common pathway is responsible for this effect.

An alternative explanation for both shock- and LPS-induced change in alcohol reactivity is a change in the pharmacokinetics or metabolism of alcohol. Both tailshock and LPS treatment activate an acute phase response, which includes a shift in liver metabolism characterized by changes in plasma acute phase proteins (Deak et al., 1997; Kushner, 1991). It may be that either of these manipulations slows the first pass metabolism of alcohol in the liver due to enzymatic priority changes induced by the acute phase response. Although this cannot be ruled out as an explanation for the effects observed at 2 h post-stress, it is highly unlikely for the significant effects observed 24 h later since these alterations would normally be back to baseline at that point. We did not measure blood alcohol concentrations (BACs) and so we do not know if first pass metabolism is reduced at the 2 h time point. However, four sets of inconsistencies make this possibility rather unlikely. First, since LPS and shock stress alter acute phase proteins similarly, one would expect similar changes in motor ataxia at 2 h post-stress. Yet, only shock stress results in a potentiated motor ataxia at this time, while LPS pretreatment is without effect. Second, we have previously shown that swim stress results in a profound hypothermia and body temperature does not return to baseline until approximately 3 h post-swim stress (Levay et al., 2006). Presumably, there is a concomitant decrease in metabolic function, yet this stressor does not alter the motor ataxic potency of alcohol. Third, since both shock and LPS result in a hyperthermic state that lasts for more than 2 h (Deak et al., 1997; Romanovsky et al., 1998), one would expect increased metabolic function of the liver causing a greater metabolic breakdown of alcohol. This enhanced degradation of alcohol would result in a diminished motor ataxic profile of alcohol rather than the increased effect as we observed. Finally, high levels of blood endotoxin do not affect alcohol elimination in rat (Nosova et al., 1998). Therefore, we suggest that tailshock and LPS administration are having an impact on the behavioral reactivity to alcohol due to pharmacodynamic actions on central nervous system receptors.

One possible candidate receptor is the benzodiazepine/GABAa receptor complex based on several observations: 1) In our original observation of shock stress controllability and alcohol effects (Drugan et al., 1996), we observed a potentiation of the motor ataxic effects of both alcohol and the benzodiazepine, midazolam, 2) 24 h following exposure to IS changes in the BDZ/GABAa receptor complex are associated with stress-induced behavioral depression (Drugan et al., 1989), and 3) exposure to the anxiogenic partial inverse agonist at the benzodiazepine receptor, FG-7142, causes a long-term potentiation of the motor ataxic effects of alcohol 24 h later (Austin et al., 1999). LPS administration has similar actions at the benzodiazepine/GABAa receptor that also suggest that this may be a final common pathway for both effects: 1) LPS exposure is known to increase blood and brain levels of IL-1B (Layé et al., 1995) and IL-1 has been shown to up regulate GABAa receptors (Serates et al., 2006), 2) experimentally-induced septic shock changes the binding kinetics of the benzodiazepine/GABAa receptor complex (Kadoi and Saito, 1996), and 3) alters the sensitivity to benzodiazepines when tested 24 h later (Komatsu-bara et al., 1995). The present results extend these previous findings by showing that more modest immune challenges can exert long-term effects on the behavioral reactivity to CNS depressants such as alcohol. It may be that the benzodiazepine/GABAa receptor is the final common pathway concerning the mechanism responsible for these disparate stressors enhancing the behavioral reactivity to ETOH.

Although LPS had a significant effect on alcohol's ataxic actions 24 h later, it was not sufficient to produce overt behavioral correlates of sickness behavior. More specifically, we evaluated body weight, food and water intake and did not observe a difference between LPS- and saline-treated rats. Perhaps if a more fine-tuned method of analysis were utilized such as biotelemetry devices, we might have seen a significant change in core body temperature or metabolic rate. Most studies that evaluate the effects of LPS on sickness behavior employ a much higher dose of this same serotype (e.g., 50 μg/kg or more; Castanon et al., 2001).

In sum, the results indicate that exposure to environmental or immune challenges can cause long-term alterations in the response to drugs of abuse such as alcohol. This points to the importance of evaluating stress and illness history in addition to prior alcohol use when assessing the behavioral reactivity to alcohol. This stress-induced exaggeration of the behavioral responses to alcohol may have important implications for the progression from alcohol use to abuse to addiction.

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