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Interaction between 1,4-butanediol and ethanol on operant responding and the cardiovascular system☆

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Abstract

The current studies characterized the rate-decreasing and cardiovascular responses produced by 1,4-butanediol administered alone and in combination with ethanol to test the hypothesis that these effects resulted from the degradation of 1,4-butanediol to γ-hydroxybutyrate. One group of rats responded under a fixed-ratio 20 schedule of food presentation; ethanol and 1,4-butanediol dose-dependently decreased response rates. Ethanol administered in combination with 1,4-butanediol attenuated the rate-decreasing effects of 1,4-butanediol without altering the potency of ethanol. In separate groups of conscious rats, radio telemetry was used to record mean arterial pressure and heart rate. In contrast to its depressant effects on schedule-controlled responding, 1,4-butanediol increased mean arterial pressure and heart rate; these increases were attenuated by ethanol. Thus, the behavioral and cardiovascular actions of 1,4-butanediol are similar to those elicited by γ-hydroxybutyrate. The ability of ethanol to attenuate the behavioral and cardiovascular effects of 1,4-butanediol indicates that these effects require the conversion of 1,4-butanediol to γ-hydroxybutyrate.

Keywords: 1,4-Butanediol; Schedule-controlled behavior; Mean arterial pressure; Heart rate; Radio telemetry; (Rat)

1. Introduction

γ-Hydroxybutyrate is a compound found endogenously in mammalian brains that has also been used recreationally for its ability to produce euphoria with a loss of social inhibition (Chin et al., 1998; Friedman et al., 1996). As a club drug, γ-hydroxybutyrate is commonly used in combination with ethanol, and it has also been used illicitly to facilitate sexual assault (Degenhardt et al., 2002; Nicholson and Balster, 2001). Aside from its recreational use, γ-hydroxybutyrate was approved by the United States Food and Drug Administration for the treatment of cataplexy associated with narcolepsy (Abramowicz, 2002; Fuller and Hornfeldt, 2003). γ-Hydroxybutyrate has also been used therapeutically for alcohol withdrawal in some European countries (Gallimberti et al., 1989; Poldrugo and Addolorato, 1999). Metabolically linked to the major inhibitory neurotransmitter γ-aminobutyric acid (GABA), γ-hydroxybutyrate is often compared to drugs that positively modulate $GABA_A$ receptors such as benzodiazepines and barbiturates (Bernasconi et al., 1999; Nicholson and Balster, 2001). Although both γ-hydroxybutyrate and positive $GABA_A$ receptor modulators can produce sedation and induce anesthesia, many behavioral effects of γ-hydroxybutyrate are different from those produced by positive $GABA_A$ receptor modulators and likely result from the interaction between γ-hydroxybutyrate and specific γ-hydroxybutyrate receptors or $GABA_B$ receptors (Nicholson and Balster, 2001). For example, $GABA_B$ receptors appear

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to be important in the discriminative stimulus effects of γ-hydroxybutyrate (Carter et al., 2003; Carter et al., 2004a; Lobina et al., 1999). In contrast to the behavioral effects of γ-hydroxybutyrate, which generally result from depression of the central nervous system, γ-hydroxybutyrate is a cardiovascular stimulant (Blumenfeld et al., 1962; Boyd et al., 1992; Gomes et al., 1976; Persson and Henning, 1980a). γ-Hydroxybutyrate elicits marked and prolonged increases in mean arterial pressure and heart rate involving the activation of central $GABA_B$ receptors (Hicks et al., 2004).

The illicit use of γ-hydroxybutyrate has raised concern over its toxicity, leading the United States Food and Drug Administration to ban the nonprescription sale of γ-hydroxybutyrate, which greatly decreased its availability (Center for Disease Control, 1991; Food and Drug Administration, 1991). As such, many users have turned to a common industrial solvent and unscheduled metabolic precursor of γ-hydroxybutyrate, 1,4-butanediol. 1,4-Butanediol is metabolized to γ-hydroxybutyrate in vivo by the same enzymes responsible for the degradation of ethanol (Bessman and McCabe, 1972; Poldrugo and Snead, 1986). Moreover, ethanol and 1,4-butanediol are competitive substrates for alcohol dehydrogenase. Ethanol inhibits the conversion of 1,4-butanediol to γ-hydroxybutyrate in rat liver and increases mortality and tissue damage associated with 1,4-butanediol administration (Poldrugo et al., 1985; Poldrugo and Snead, 1986). The significance of the interaction between ethanol and 1,4-butanediol with regard to the rate-decreasing and cardiovascular effects of 1,4-butanediol has not been studied.

Some of the behavioral effects of 1,4-butanediol, such as the loss of righting reflex, have been attributed to its metabolism to γ-hydroxybutyrate (Carai et al., 2002; Quang et al., 2002; Roth and Giarman, 1968); however, the effects of 1,4-butanediol on the cardiovascular system are unknown. Furthermore, the role, if any, that γ-hydroxybutyrate has in the cardiovascular responses associated with the administration of 1,4-butanediol is unknown. Therefore, the purpose of this study was to (1) examine the effects of 1,4-butanediol and ethanol administered alone on schedule-controlled behavior, (2) characterize the cardiovascular responses elicited by 1,4-butanediol and ethanol administered alone and (3) evaluate the interaction of ethanol and 1,4-butanediol within the behavioral paradigm and cardiovascular system.

2. Methods

2.1. Subjects

Animals used in these studies were maintained in accordance with the Institutional Animal Care and Use Committee, Louisiana State University Health Sciences Center, and guidelines of the Committee on Care and Use of Laboratory Animal Resources, National Research Council [Department of Health, Education and Welfare, Publication No. (NIH) 85-23, revised 1996].

2.1.1. Behavioral studies

Ten male Long–Evans hooded rats were housed individually in a humidity- and temperature-controlled room with a 12-h light/dark cycle. In their home cages, rats had tap water available ad libitum. To maintain rats at 85% of the free-feeding weight, which was determined immediately before the start of the experiment, they received food pellets during experimental sessions (Research Diets, New Brunswick, NJ) and rodent chow (Lab Diets, Brentwood, MO) after experimental sessions in their home cage.

2.1.2. Cardiovascular studies

Experiments were performed using 22 male Sprague–Dawley rats (252–344 g; Harlan, Indianapolis, IN). Before implantation of radio telemetry probes and femoral venous cannulae, the rats were group housed in a humidity- and temperature-controlled room with a 12-h light/dark cycle. After surgery, the rats were individually housed with rodent chow (Lab Diets) and tap water available ad libitum. The rats were anesthetized using pentobarbital (60 mg/kg, i.p.) for all surgical procedures. Supplemental anesthesia was administered in response to spontaneous changes in respiration, or in response to tail or foot pinch.

2.2. Apparatus

2.2.1. Behavioral studies

During experimental sessions, rats were placed in six modular test chambers enclosed within sound-attenuating cubicles equipped with fans for ventilation. Each chamber was equipped with a houselight, speaker, pellet trough, pellet dispenser and two response levers with stimulus lights located directly above each lever. In the current study, one stimulus light was illuminated during response periods and the corresponding lever was active; the other lever was inactive. White noise was present in the chambers to mask extraneous noise. Each chamber was connected to a computer programmed in MED-PC/MEDSTATE NOTATION software (MED Associates, St. Albans, VT) and to cumulative recorders (Gerbrands, Arlington, MA).

2.3. Procedure

2.3.1. Behavioral studies

Rats responded under a fixed-ratio 20 schedule of food presentation. Experimental sessions consisted of three to eight discrete, 15-min cycles. Each cycle began with a 10-min timeout period during which the chamber was dark and responses had no programmed consequence. Illumination of the house light and the stimulus light above the active lever signaled the beginning of a response period that had a maximum duration of 5 min. During the response period, 20 responses on the active lever resulted in delivery of a 45-mg

food pellet. Responses on the inactive lever had no programmed consequence. The active lever was counterbalanced among rats. Stimulus and house lights were extinguished after 5 min or the delivery of 30 food pellets, whichever occurred first; any time remaining between the end of the response period and the beginning of the next cycle was a timeout period.

Intraperitoneal injections were administered prior to sessions. For some training sessions, a single injection of saline was administered at various times before the session (0, 15 or 90 min); on other occasions, two injections of saline were administered: one 15 min and one immediately before sessions. For still other training sessions, rats did not receive injections. Test sessions were identical to training sessions except that drugs were administered before sessions which consisted of eight cycles. The time courses for ethanol (0.5–2.0 g/kg) and 1,4-butanediol (0.18–0.56 g/kg) were determined by administering a single dose of drug immediately before sessions; because marked rate-decreasing effects of 1,4-butanediol were still evident 2 h after administration, each dose of 1,4-butanediol was also administered 90 min before a session. The interaction between 1,4-butanediol and ethanol was evaluated by administering a single dose of 1,4-butanediol 15 min before a single dose of ethanol, which was administered immediately before sessions. The intervals between injections and sessions were selected based on the time to peak effect for each drug as determined in the initial time course studies. Several doses of ethanol were studied in combination with each dose of 1,4-butanediol; after the dose–effect curve for ethanol was completed in the presence of a dose of 1,4-butanediol, the time course for that dose of 1,4-butanediol was redetermined.

2.3.2. Cardiovascular studies

Mean arterial pressure and heart rate were recorded in conscious, unrestrained rats in their home cages using a radio telemetry system (Dataquest A.R.T. 2.3; Data Sciences International, St. Paul, MN) as described previously (Badon et al., 2002; Varner et al., 2002). Briefly, the pressure cannula of a radio telemetry probe was inserted into the descending abdominal aorta rostral to the iliac bifurcation and the probe was secured to the abdominal musculature. In rats receiving intravenous drug, a polyurethane cannula (Micro-renathane, 0.33 inch o.d.×0.014 inch i.d.; Braintree Scientific, Braintree, MA) was inserted into the femoral vein and the free end tunneled subcutaneously to the nape of the neck and exteriorized. After surgery, fluids and penicillin (60,000 units, i.m.) were administered. Buprenorphine (2.5 mg/kg, i.p.) was administered twice a day for 2 days. The rats were allowed to recover from the surgical procedure for 7–10 days before beginning any experimental protocol. Approximately 1 week after surgery, the rats' home cages were placed over telemetry receivers where they remained for the duration of the experiment. In all telemetry studies, the daily weight was measured and baseline mean arterial pressure and heart rate were recorded for a minimum of 20 min prior to the administration of any drug.

Six rats were instrumented with telemetry probes. On the first day of the study, the rats received a single intraperitoneal dose of saline (1.0 ml) or 1,4-butanediol (0.18, 0.32, 0.56 or 1.0 g/kg, 0.18–1.3 ml) and the cardiovascular responses were recorded for a minimum of 3 h. On subsequent days, doses of 1,4-butanediol or saline were administered in mixed order with only one dose administered per day. A second group of rats (n=7) was instrumented with telemetry probes and femoral venous cannulae. This group received a single intravenous dose of saline or 1,4-butanediol (0.18, 0.32, 0.56 or 1.0 g/kg) and the cardiovascular responses were recorded for at least 3 h. 1,4-Butanediol was injected (50 μl to 1.0 ml) over 1–2 min followed by a 0.5–0.7 ml saline flush. Again, on subsequent days, doses of 1,4-butanediol or saline were administered in mixed order with only one dose administered per day. In both groups, each animal received all doses of 1,4-butanediol.

Four rats were instrumented with telemetry probes and allowed to recover for 7–10 days. On the morning of the experiment, the rats were weighed and baseline mean arterial pressure and heart rate were recorded for a minimum of 20 min. Ethanol (2.0 g/kg, i.p.) was then administered and the cardiovascular responses recorded for a minimum of 3 h. A separate group of rats (n=5) was instrumented with telemetry probes and femoral venous cannulae and allowed to recover. Approximately 1 week later, the rats received an intraperitoneal injection of ethanol (2.0 g/kg) 10 min prior to the administration of 1,4-butanediol (0.56 g/kg, i.v.) and the cardiovascular responses were continuously recorded for a minimum of 3 h.

2.4. Drugs

1,4-Butanediol (Aldrich Chemical, Milwaukee, WI) was diluted with saline to obtain a 31.5% (v/v) solution that was used for all injections. In the behavioral studies, 1,4-butanediol was administered intraperitoneally, and in the cardiovascular studies, 1,4-butanediol was administered either intraperitoneally or intravenously. Ethanol was diluted with saline to obtain a 20% (v/v) solution that was administered intraperitoneally. Doses are expressed in terms of the forms listed above in g/kg body weight. Pentobarbital (Sigma-Aldrich, St. Louis, MO) was dissolved in a solution consisting of propylene glycol (40%), ethyl alcohol (10%) and sterile water (50%) at 60 mg/ml.

2.5. Data analyses

2.5.1. Behavioral studies

Control response rates were determined by averaging response rates across cycles to obtain mean response rates for each session. Mean response rates for 10 training sessions were averaged to obtain the control response rate (±1

S.E.M.) for each rat. Response rates for each subject were expressed as a percentage of the control rate and averaged across subjects. Mean response rates were plotted as a function of time (min) from the beginning of the session. In order to determine whether sensitivity to ethanol changed as a result of treatment with 1,4-butanediol, response rates obtained 15 min after ethanol administration were replotted as a function of ethanol dose. Doses of ethanol required to decrease responding to 50% of control (ED_{50}) were estimated for each subject using linear regression when three data points were available, otherwise by interpolation. In one subject, the ED_{50} value could not be determined for ethanol in the presence of 0.56 g/kg of 1,4-butanediol because response rates were not decreased to <50% of control on the descending limb of the ethanol dose–effect curve; larger doses of ethanol were not studied to avoid toxicity. For that subject, a value of 1.5 g/kg was used for the ED_{50} value; this dose of ethanol was the largest administered in the presence of 0.56 g/kg of 1,4-butanediol.

2.5.2. Cardiovascular studies

The output from the telemetry probes was recorded (250 Hz) using receivers placed under the animals' home cages. The data was sent to a consolidation matrix before being stored on a personal computer. Data acquisition was controlled using Data Sciences Dataquest acquisition software. The data were averaged into 2-s bins and displayed. The magnitude of the peak changes in mean arterial pressure and heart rate elicited by the administered drugs were calculated off-line using Dataquest analysis software and are reported as means ± S.E.M. The magnitudes of the peak changes in mean arterial pressure and heart rate elicited by the intraperitoneal administration of saline were measured 98 and 130 min, respectively, after injection. These time points correspond to the time to peak of the increases in mean arterial pressure (98±10 min) and heart rate (130±14 min) elicited by the intraperitoneal administration of 1.0 g/kg of 1,4-butanediol. The time to peak for the 1.0 g/kg dose of 1,4-butanediol was chosen as the volumes of these injections were similar to the volume of saline administered. Similarly, changes in mean arterial pressure and heart rate elicited by intraperitoneal administration of ethanol alone and in combination with 1,4-butanediol (0.56 g/kg, i.v.) were measured at time points that corresponded to the average time to peak for the intravenous administration of 0.56 g/kg of 1,4-butanediol (65±3 and 92±8 min, respectively). Response durations were also calculated off-line as the time interval between drug administration and the point at which the mean arterial pressure and heart rate returned to within 7 mm Hg or 7 bpm of baseline, respectively. Within groups, baseline and peak mean arterial pressure and heart rate responses were compared using one-way repeated-measures analysis of variance (ANOVA). The peak mean arterial pressure and heart rate responses recorded across subject groups and the durations of the mean arterial pressure and heart rate responses elicited by intraperitoneal and intravenous administrations were compared using one-way ANOVA. After the ANOVAs, the pairwise comparisons of the groups were conducted using the Tukey test. Statistical significance was assigned at $P<0.05$.

3. Results

3.1. Behavioral studies

Under control conditions, response rates were similar throughout the experiment; for example, mean (±1 S.E.M.) response rates for the 10 rats that participated in these studies was 2.51±0.17 responses/s prior to establishing the ethanol dose–effect curve and 2.39±0.24 responses/s at the end of the experiment. Ethanol dose-dependently decreased response rates (Fig. 1). The onset of action of ethanol was rapid with the maximum effects evident 15 min after administration. Although response rates increased over the 2-h experimental session, rate-decreasing effects of 1.5 and 2.0 g/kg of ethanol were still evident at the end of the session.

1,4-Butanediol also dose-dependently decreased response rates (Fig. 2). A dose of 0.18 g/kg of 1,4-butanediol had no effect on response rates, and larger doses eliminated responding, although these marked rate-decreasing effects were not evident until 30 min after administration. Response rates were markedly decreased for 2 h after administration of a dose of 0.32 g/kg and for at least 3.5 h after a dose of 0.56 g/kg of 1,4-butanediol.

Interactions between 1,4-butanediol and ethanol were examined by administering both drugs prior to the same experimental sessions so that the maximum rate-decreasing effects of each drug occurred during the first cycle. Small doses of ethanol attenuated the rate-decreasing effects of 1,4-butanediol (Fig. 3). For example, the rate-decreasing effects

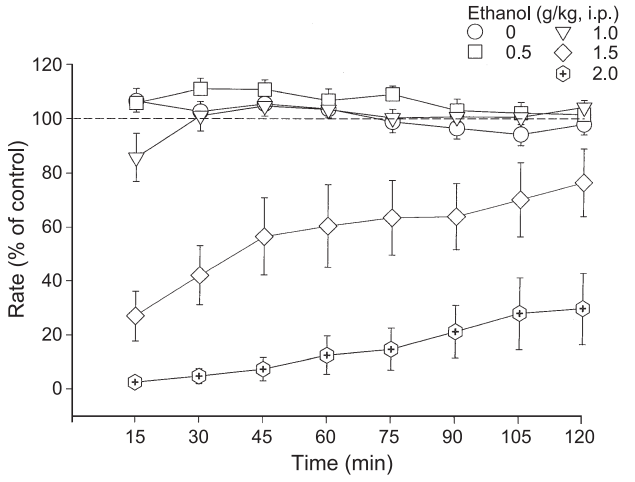

Fig. 1. Rate-decreasing effects of ethanol in 10 rats responding under a fixed-ratio 20 schedule of food presentation. Abscissa: time after ethanol administration in min. Ordinate: average rate expressed as a percentage of control response rate±1 S.E.M.

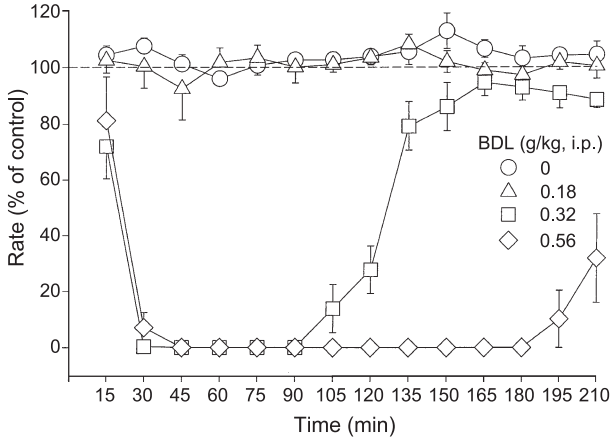

Fig. 2. Rate-decreasing effects of 1,4-butanediol administered either immediately or 90 min before experimental sessions in six rats responding under a fixed-ratio 20 schedule of food presentation. Abscissa: time after 1,4-butanediol administration in min. Ordinate: average rate expressed as a percentage of control response rate±1 S.E.M.

of 0.56 g/kg of 1,4-butanediol administered in combination with ethanol were similar to those produced by the same dose of ethanol administered alone. Despite the attenuation of the rate-decreasing effects of 1,4-butanediol by ethanol, 1,4-butanediol did not markedly alter the ethanol dose–effect curves. The mean ED_{50} value (±1 S.E.M.) for ethanol was 1.23±0.09 g/kg in the absence of 1,4-butanediol and 1.06±0.11 g/kg in the presence of 0.56 g/kg of 1,4-butanediol.

The complex interaction between ethanol and 1,4-butanediol is reflected in time course data for the combination. Doses of 0.25 and 0.5 g/kg of ethanol completely attenuated the rate-decreasing effects of 1,4-butanediol for 30 or 75 min, respectively (Fig. 4). Although the rate-decreasing effects of 1 and 1.5 g/kg of ethanol were

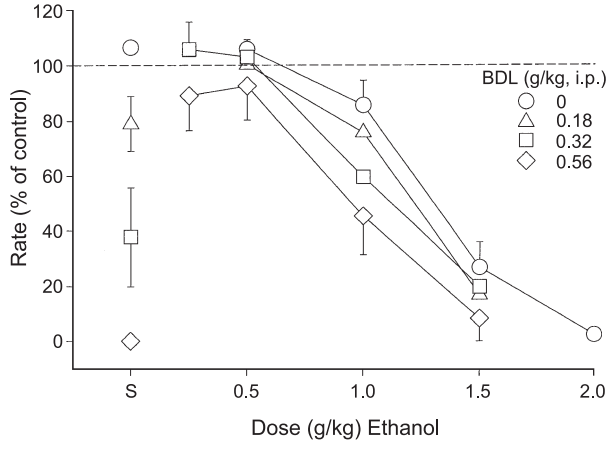

Fig. 3. Rate-decreasing effects of ethanol determined in the absence and presence of 1,4-butanediol (n=6). Dose–effect curves for ethanol administered alone and in combination with 1,4-butanediol were constructed using data from the first cycle of the session. 1,4-Butanediol was administered 15 min before ethanol, which was administered immediately before sessions. Points above S represent the effects of 1,4-butanediol administered alone. Abscissa: dose of ethanol in g/kg. Ordinate: Average rate expressed as a percentage of control response rate±1 S.E.M.

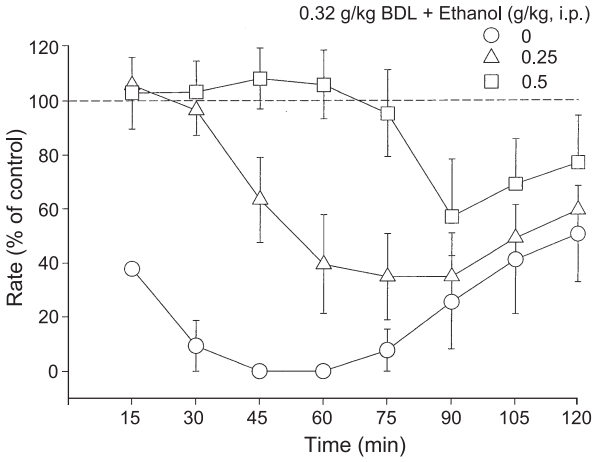

Fig. 4. Time course of the rate-decreasing effects of 0.32 g/kg of 1,4-butanediol administered in combination with doses of ethanol that did not decrease response rates (0.25 and 0.5 g/kg). 1,4-Butanediol was administered 15 min before ethanol, which was administered immediately before sessions. Abscissa: time after ethanol administration in min. Ordinate: Average rate expressed as a percentage of control response rate±1 S.E.M.

evident early in experimental sessions, these large doses also attenuated the rate-decreasing effects of 1,4-butanediol, and this attenuation was evident for the duration of the experimental sessions (Fig. 5).

3.2. Cardiovascular studies

Fig. 6 shows a typical experimental recording of the mean arterial pressure and heart rate responses elicited by the intraperitoneal administration of 1,4-butanediol (0.56 g/kg) in a conscious telemetered rat. 1,4-Butanediol elicited a large and prolonged increase in mean arterial pressure. This dose of 1,4-butanediol also elicited a pronounced and prolonged increase in heart rate (Fig. 6). The intravenous administration

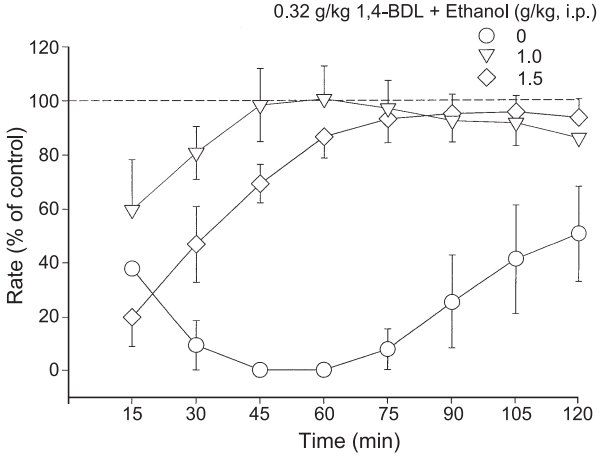

Fig. 5. Time course of the rate-decreasing effects of 0.32 g/kg of 1,4-butanediol administered in combination with doses of ethanol that decreased response rates (1.0 and 1.5 g/kg). 1,4-Butanediol was administered 15 min before ethanol, which was administered immediately before sessions. Abscissa: time after ethanol administration in min. Ordinate: Average rate expressed as a percentage of control response rate±1 S.E.M.

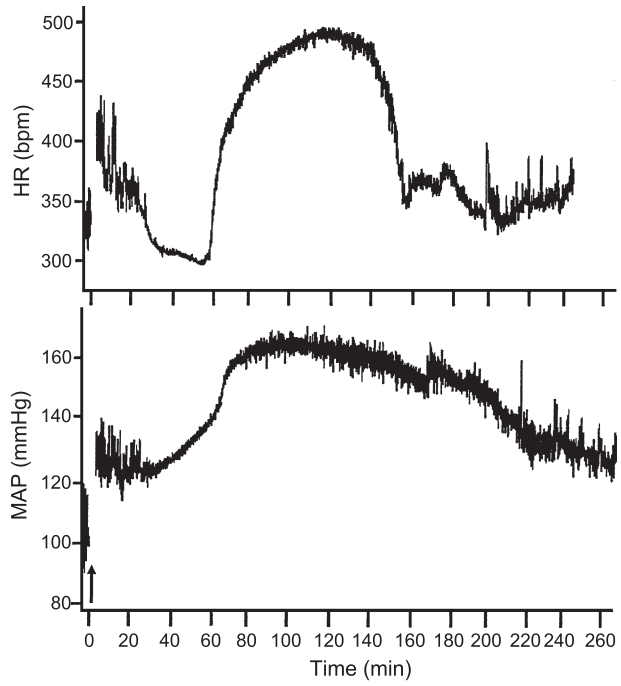

Fig. 6. Typical experimental records of the mean arterial pressure (MAP) and heart rate (HR) responses elicited by the intraperitoneal administration of 1,4-butanediol (0.56 g/kg) in a conscious, telemetered rat. Arrow denotes time of injection.

of 1,4-butanediol elicited increases in mean arterial pressure and heart rate whose magnitudes were nearly identical to those elicited by intraperitoneal administration. Fig. 7 summarizes the peak mean arterial pressure and heart rate responses elicited by the intraperitoneal (panel A, n=6) and intravenous (panel B, n=7) administration of 1,4-butanediol or saline in conscious rats. The intraperitoneal administration of 1,4-butanediol elicited prolonged increases in mean arterial pressure and heart rate lasting from 67±8 to 474±35 and 43±4 to 402±22 min, respectively. The intravenous administration of 1,4-butanediol also elicited prolonged increases in mean arterial pressure and heart rate (55±6 to 109±8 and 31±8 to 132±7 min, respectively). The magnitudes of the increase in mean arterial pressure elicited by the intraperitoneal administration of 1,4-butanediol were not significantly different from the increase in mean arterial pressure elicited in animals receiving 1,4-butanediol intravenously. The duration of the mean arterial pressure response was significantly shorter (P<0.001) in the intravenous group except at the smallest dose tested (P=0.248). Similarly, the intraperitoneal administration of 1,4-butanediol elicited large and prolonged increases in heart rate that were not significantly different from those elicited by the intravenous administration of 1,4-butanediol; however, the duration of the tachycardic response was significantly shorter with intravenous administration (P<0.001). The intravenous admin-

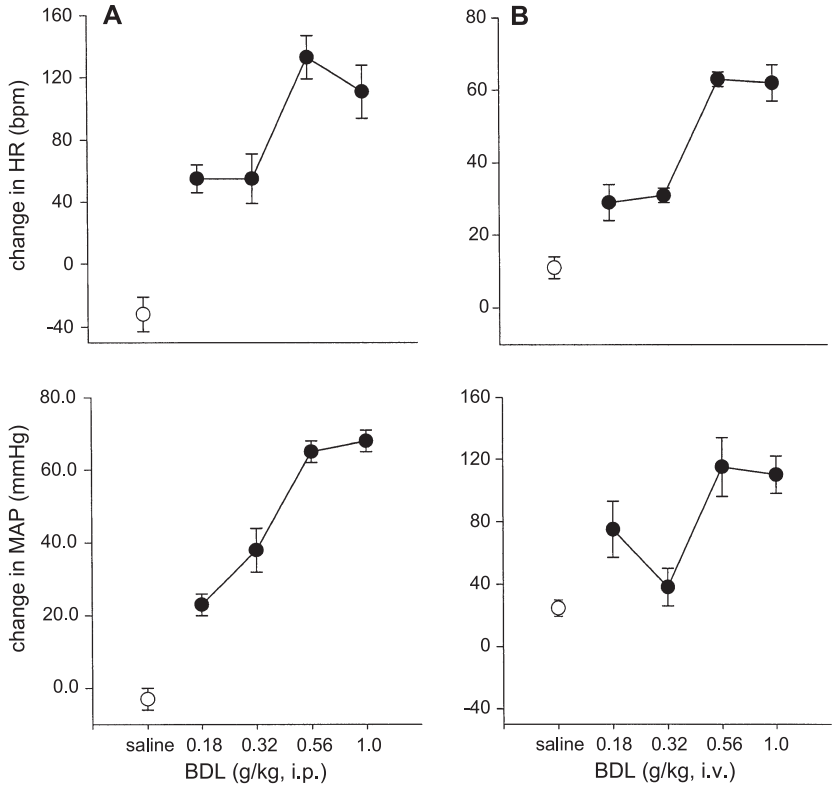

Fig. 7. Summary of the average peak mean arterial pressure (MAP) and heart rate (HR) responses elicited by intraperitoneal (panel A, n=6) or intravenous (panel B, n=7) administration of 1,4-butanediol (0.18–1.0 g/kg) and saline in conscious, telemetered rats. Values are mean±S.E.M.

istration of saline significantly increased both mean arterial pressure and heart rate; however, the duration (2±0 min) of the saline responses for both measures was significantly shorter ($P<0.001$ and $P<0.015$, respectively) than those elicited by 0.18 g/kg of 1,4-butanediol. On the other hand, the intraperitoneal administration of saline significantly lowered heart rate ($P<0.008$) without altering mean arterial pressure. Independent of route of administration, doses of 1,4-butanediol larger than 0.32 g/kg appeared to produce sedation that coincided with the sustained hypertension.

The administration of ethanol alone significantly increased heart rate (37±12 bpm) from baseline without significantly altering mean arterial pressure (4±3 mm Hg). Conversely, 1,4-butanediol alone significantly increased both mean arterial pressure and heart rate (63±2 mm Hg and 59±6 bpm, respectively). The administration of ethanol 10 min prior to the administration of 1,4-butanediol significantly attenuated both the pressor and tachycardic responses (4±3 mm Hg and 16±12 bpm, respectively) elicited by 1,4-butanediol.

4. Discussion

The current studies correlate the ability of 1,4-butanediol to decrease operant responding with its cardiovascular effects. In fact, these studies are the first to characterize the sympathomimetic cardiovascular effects elicited by 1,4-butanediol. The observations that 1,4-butanediol can produce central nervous system-depressant effects while eliciting sympathoexcitatory cardiovascular responses are not typical of most sedative-hypnotics. However, this characteristic is not unique to 1,4-butanediol as γ-hydroxybutyrate has been shown to elicit similar cardiovascular and behavioral responses (Hicks et al., 2004; Nicholson and Balster, 2001). The similarities between 1,4-butanediol and γ-hydroxybutyrate are not surprising considering that 1,4-butanediol is degraded to γ-hydroxybutyrate in vivo by alcohol dehydrogenase. Data from the present studies characterize this pharmacokinetic interaction between 1,4-butanediol and ethanol within both a behavioral paradigm and the cardiovascular system.

1,4-Butanediol and ethanol dose-dependently decreased responding when administered alone. The rate-decreasing effects of ethanol (Bird et al., 1985; Lamb et al., 2003) and 1,4-butanediol (Carter et al., 2004b; Eckermann et al., 2004) have been well characterized and are consistent among studies. For example, despite differences in rat strain and operant schedules, ED_{50} values for ethanol varied by <1.3-fold [current study; (Bird et al., 1985; Lamb et al., 2003)]. When the rate-decreasing effects of 1,4-butanediol were determined in Sprague–Dawley rats under different schedule conditions (Carter et al., 2004b), a 2-fold smaller dose of 1,4-butanediol was required to eliminate responding as compared to the dose that eliminated responding in the current study.

The interactions between ethanol and 1,4-butanediol demonstrated in the current study are consistent with both drugs being substrates for alcohol dehydrogenase. The ability of ethanol to attenuate the rate-decreasing effects of 1,4-butanediol indicates that these effects most likely reflect the conversion of 1,4-butanediol to γ-hydroxybutyrate. This conclusion is also supported by the changes in the time course when ethanol was administered with 1,4-butanediol. The maximum rate-decreasing effects of 1,4-butanediol occurred 30–40 min after administration [current study, (Carter et al., 2004b)]. γ-Hydroxybutyrate has a more rapid onset of action with the maximum rate-decreasing effects evident 20 min after administration (Carter et al., 2004b). Therefore, the time required to convert 1,4-butanediol to γ-hydroxybutyrate could account for the longer onset of action of 1,4-butanediol. These differences suggest that 1,4-butanediol is active only after it has been converted to an active metabolite. Doses of ethanol (0.25 and 0.5 g/kg) that were ineffective when administered alone completely attenuated the rate-decreasing effects of 1,4-butanediol with response rates >90% of control following administration of both ethanol and 1,4-butanediol as compared to response rates that were <10% of control when 1,4-butanediol was administered alone. As ethanol was metabolized, allowing 1,4-butanediol to be converted to γ-hydroxybutyrate, the time course of the combination more closely resembled that of 1,4-butanediol alone; this effect is dependent on the dose of ethanol. The rate-decreasing effects of larger doses of ethanol alone were not altered when administered in combination with 1,4-butanediol (0.32 g/kg). Although the rate-decreasing effects of ethanol dissipated as it was metabolized, ethanol continued to prevent the degradation of 1,4-butanediol. As such, the rate-decreasing effects of 1,4-butanediol were only evident in the absence of ethanol, indicating the need for conversion of 1,4-butanediol to γ-hydroxybutyrate by alcohol dehydrogenase.

The present study also showed that the intravenous and intraperitoneal administration of 1,4-butanediol consistently increases mean arterial pressure and heart rate in conscious rats. To our knowledge, the cardiovascular effects of 1,4-butanediol have not been characterized in other species. The magnitudes of the increase in mean arterial pressure and tachycardia elicited by individual doses of 1,4-butanediol are independent of the route of administration. The dose–response relationships for 1,4-butanediol and mean arterial pressure and heart rate are remarkably similar to those elicited by γ-hydroxybutyrate (Hicks et al., 2004; Persson and Henning, 1980b). Interestingly, the intraperitoneal and intravenous administration of 1,4-butanediol appeared to produce sedation that corresponded to the prolonged hypertension. The ability of ethanol pretreatment to block the cardiovascular responses elicited by 1,4-butanediol indicates that increases in mean arterial pressure and heart rate occur after the degradation of 1,4-butanediol to γ-hydroxybutyrate by alcohol dehydrogenase. This observation is consistent with the conclusion that the rate-

decreasing effects of 1,4-butanediol also require the conversion of 1,4-butanediol to γ-hydroxybutyrate.

The pharmacodynamics of 1,4-butanediol within the behavioral paradigm and the cardiovascular system are most likely due to γ-hydroxybutyrate via activation of $GABA_B$ and γ-hydroxybutyrate receptors. In fact, increases in mean arterial pressure and sympathetic nerve activity elicited by the acute administration of γ-hydroxybutyrate are mediated by the activation of central $GABA_B$ receptors (Hicks et al., 2004). Increases in heart rate elicited by γ-hydroxybutyrate appear to be mediated by the activation of specific γ-hydroxybutyrate receptors (Hicks et al., 2004). These receptor systems are also thought to be involved in the discriminative stimulus effects of γ-hydroxybutyrate (Carter et al., 2003). Taken together, these data suggest that the rate-decreasing effects and the cardiovascular responses elicited by 1,4-butandiol are mediated by $GABA_B$ and γ-hydroxybutyrate receptors following the conversion of 1,4-butanediol to γ-hydroxybutyrate. Despite differences in the patterns of misuse of 1,4-butanediol and γ-hydroxybutyrate, the current findings suggest that the behavioral and cardiovascular toxicities of 1,4-butanediol could be directly related to the toxicity associated with γ-hydroxybutyrate.

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