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Lorazepam Attenuates the Behavioral Effects of Dizocilpine

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FAHEY, J. M., G. A. PRITCHARD, J. S. PRATT, R. I. SHADER AND D. J. GREENBLATT. *Lorazepam attenuates the behavioral effects of dizocilpine.* PHARMACOL BIOCHEM BEHAV **62**(1) 103–110, 1999.—To characterize the potential interaction between the excitatory and inhibitory neurotransmitter systems, the effects of dizocilpine, CPP, and lorazepam on open-field behavior and pentylenetetrazol-induced seizures were evaluated in mice. Dizocilpine (0.01–0.1 mg/kg), CPP (1–10 mg/kg), or vehicle was administered intraperitoneally 15 min prior to lorazepam (0.2–2 mg/kg) or vehicle. Behavioral monitoring began 25 min after the lorazepam injection. Upon completion of testing, unrestrained mice were infused intravenously with pentylenetetrazole until the onset of a full tonic–clonic seizure. The highest dose of dizocilpine by itself significantly increased the average distance traveled, the number of rears, and the number of stereotypies during the test period. Lorazepam alone dose dependently decreased activity on all behavioral parameters. Lorazepam also completely antagonized the hyperactivity produced by dizocilpine when the two compounds were coadministered. This antagonism is most likely due to an interaction in the regulation of dopaminergic tone which underlies motor activity. Lorazepam exerted a dose-dependent anticonvulsant effect. Dizocilpine alone had no effect on seizure induction and did not potentiate the anticonvulsive effect of lorazepam when coadministered with lorazepam. CPP reduced the number of rears and the number of stereotypies during the test period. CPP did not alter the pentylenetetrazol-induced seizure threshold and did not influence the anticonvulsant effect of lorazepam. © 1998 Elsevier Science Inc.

Behavior Benzodiazepine CPP Dizocilpine GABA Lorazepam NMDA antagonist Open-field activity Pentylenetetrazol-induced seizures

RECENT evidence suggests a regulatory role of excitatory amino acid (EAA) receptors in the function of $GABA_A$/benzodiazepine responses in vivo and in vitro. The EAA receptors, like $GABA_A$ receptors, are widely distributed throughout the central nervous system (33), and are the principal neurotransmitters involved in fast excitation, while the $GABA_A$ system is the principal inhibitory system within the brain. It is, therefore, likely that these two neurotransmitter systems have compensatory and regulatory effects on one another. The EAA receptor family is composed of both metabotropic (G-protein coupled) and ligand-gated ionotropic receptors. The latter group can be further subdivided into receptors with selective affinity for *N*-methyl-d-aspartate (NMDA), α-amino-3-hydroxy-5-methylisoxazole-4-propionate (AMPA), and kainate. In addition, the NMDA receptor family contains a number of ligand binding sites that regulate receptor function.

A number of investigators have recently implicated alterations in ligand-gated EAA receptor efficacy and expression as contributing factors in adaptation to chronic antidepressant treatment, opiate tolerance, and withdrawal and benzodiazepine tolerance and withdrawal [for review, see (29)]. Skolnick and colleagues found that, in mice, a wide variety of antidepressant therapies, including tricyclic antidepressants, monoamine oxidase inhibitors, serotonin selective reuptake inhibitors as well as electroconvulsive shock reproducibly altered glycine binding and efficacy at the NMDA receptor complex (29). Additionally, chronic administration of NMDA receptor antagonists directed at both competitive and noncompetitive binding sites were found to possess antidepressant-like activity (28). Extensive evidence has also accrued which indicate the efficacy of both competitive and noncompetitive NMDA receptor antagonists in ameliorating symptoms associated

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with opiate tolerance and withdrawal (2,15). Two recent in vivo benzodiazepine studies have implicated a link between the compensatory effects of ionotropic EAA receptor activity and withdrawal from chronic benzodiazepine administration (12,31). These investigators observed a diminution of tolerance to and withdrawal from chronic diazepam administration in the presence of a variety of ionotropic EAA receptor antagonists. In addition, our laboratory has recently demonstrated that 3-(2-carboxypiperazin-4-yl)propyl-1-phosphonic acid (CPP) can attenuate tolerance to and withdrawal from chronic lorazepam treatment (17). CPP was equally effective whether administered during or after chronic benzodiazepine treatment. Two studies utilizing in vitro hippocampal slice preparations have demonstrated that glutamate or NMDA reduces the efficacy of $GABA_A$ receptor-mediated responses (26,30). The regulation appears to be calcium dependent, and involves calcium-mediated phosphorylation and dephosphorylation events, further suggesting a role of ionotropic EAA receptors in the regulation of or adaptation to benzodiazepine administration in vivo.

The present study examines the potential interaction between the glutamatergic and GABAergic neurotransmitter systems in a well-characterized behavioral paradigm in male CD-1 mice. Other investigators have demonstrated that both competitive and noncompetitive NMDA antagonists produce hyperlocomotion in mice (3,18,32). Based on the evidence for a functional interaction between the major excitatory and inhibitory neurotransmitter systems outlined above, it is expected that coadministration of a benzodiazepine will antagonize the hyperlocomotion typically observed with NMDA antagonists. We administered the NMDA open channel blocker, dizocilpine, a competitive NMDA antagonist, CPP, and a positive allosteric modulator of the $GABA_A$ receptor complex, lorazepam, alone or in combination prior to evaluating open-field activity and pentylenetetrazole-induced seizure threshold. We have previously used this model to describe both acute and chronic responses to a number of different benzodiazepine site ligands including "typical" benzodiazepine agonists (13), the beta-carboline inverse agonist FG 7142 (25), and the benzodiazepine antagonist flumazenil (23).

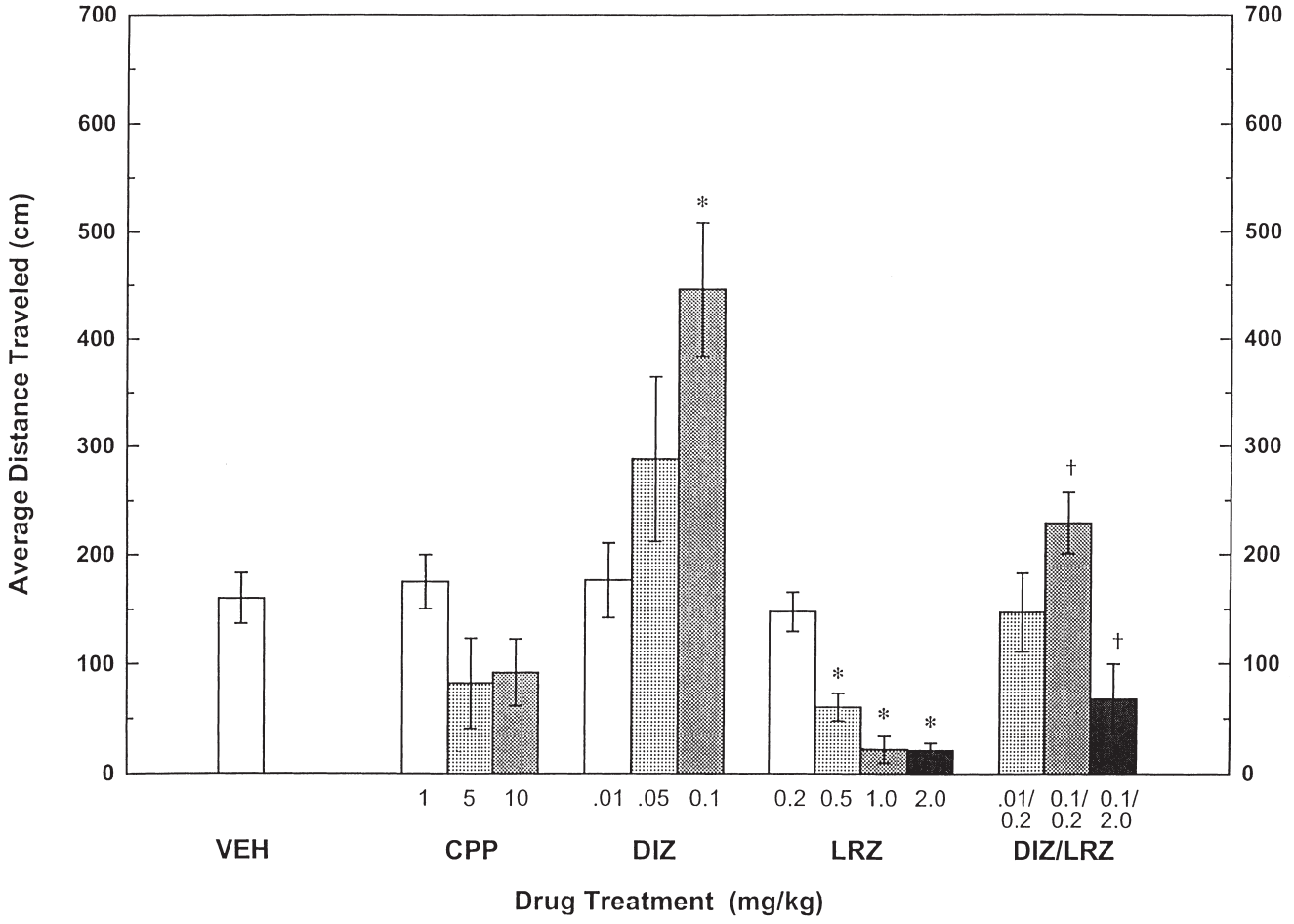

FIG. 1. Effect of CPP, dizocilpine (DIZ), and/or lorazepam (LRZ) on the average distance traveled. Data from distance traveled was recorded at 5-min intervals for a total of 20 min. Dizocilpine (0.01, 0.05, or 0.1 mg/kg), CPP (1, 5, or 10 mg/kg) or vehicle (VEH) was administered intraperitoneally 15 min prior to lorazepam (0.2, 0.5, 1, or 2 mg/kg) or vehicle ($n \geq 6$). Behavioral testing began 25 min after the lorazepam injection. Bars represent mean response ± SEM. Significant differences ($p < 0.05$) from vehicle control or from 0.1 mg/kg dizocilpine are indicated by * and †, respectively, as determined by ANOVA and Dunnetts Multiple Comparison or Student–Newman–Keuls post hoc tests.

METHOD

Materials

Male Crl: CD-1(ICR)BR mice, 6–8 weeks of age, were purchased from Charles River Laboratories (Wilmington, MA), maintained on a 12 L:12 D cycle and given food and water ad lib. Lorazepam (M.W. 321.2) was generously donated by Wyeth Ayerst (Radnor, PA). (R)-CPP (M.W. 252.21) and dizocilpine [(+)-MK-801 maleate—M.W. 337.37] were purchased from Tocris Cookson, Inc. (Ballwin, MO). All other reagents were obtained from standard commercial sources.

Drug Administration

CPP, lorazepam, and dizocilpine were dissolved in polyethylene glycol (PEG) 400:saline (1:1) and administered in a constant volume (100 μl) intraperitoneally. Vehicle-treated mice received PEG 400:saline (1:1) alone.

Open-Field Activity

Activity for all groups, including distance traveled, rears and stereotypy, was assessed in 5-min intervals for 20 min in an Omnitech Digiscan apparatus (Columbus, OH). The use of this automated activity monitor has been described in detail in previous publications (4,13,17,19,23,25). Animals were randomly assigned to one of several treatment groups ($n \geq 6$ per group). Dizocilpine (0.01, 0.05, or 0.1 mg/kg), CPP (1, 5, or 10 mg/kg) or vehicle was administered intraperitoneally 15 min prior to lorazepam (0.2, 0.5, 1, or 2 mg/kg) or vehicle. Behavioral testing began 25 min after the lorazepam injection. Between each run, the interior of the activity chamber was cleaned with 70% ethanol and dried. All testing occurred between 0900 and 1200 h.

Pentylenetetrazole-Induced Seizures

As previously described (27), unrestrained mice were infused intravenously via tail vein with a solution of pentylene-

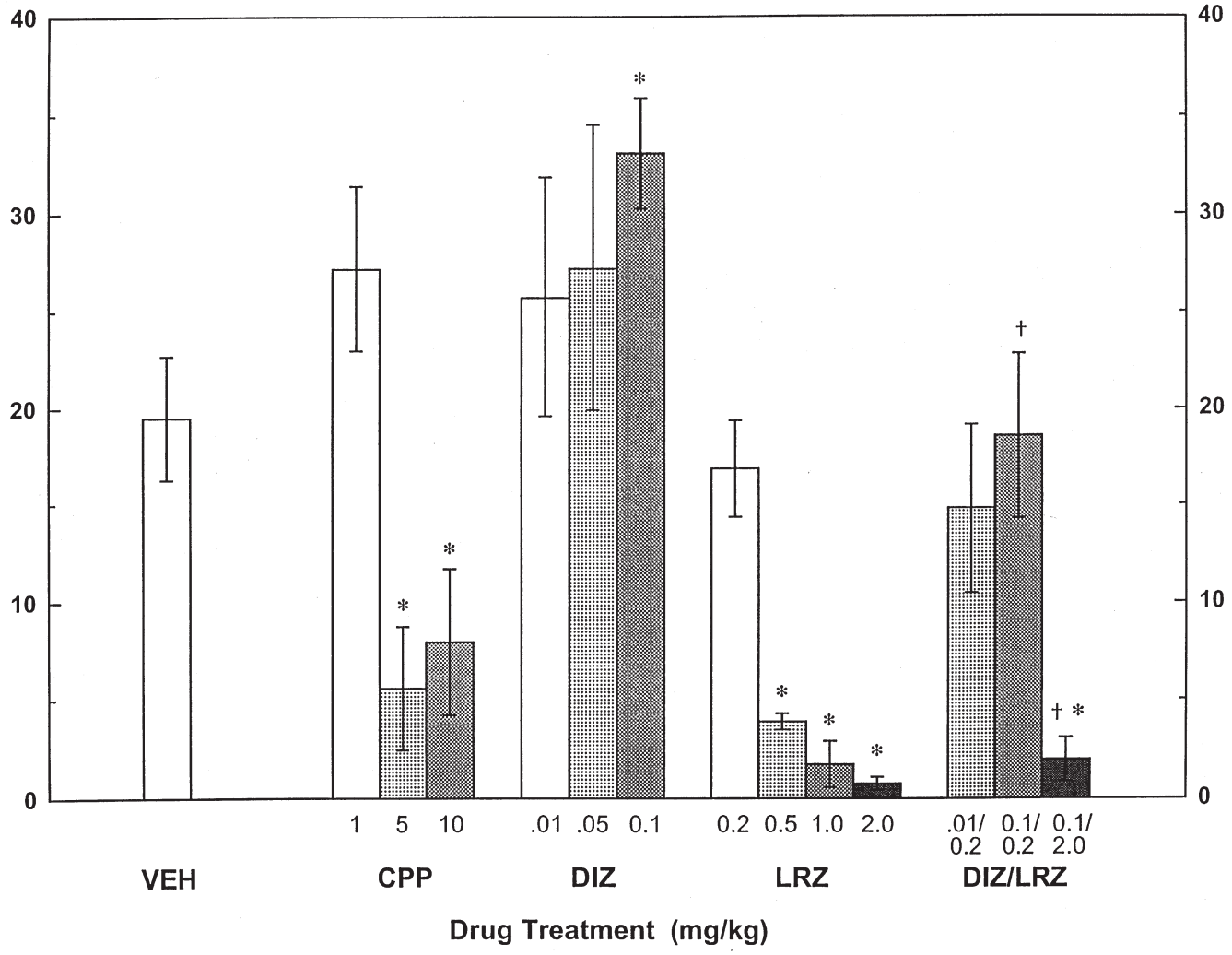

FIG. 2. Effect of CPP, dizocilpine (DIZ), and/or lorazepam (LRZ) on the average number of rears. Data from number of rears was recorded at 5-min intervals for a total of 20 min. Dizocilpine (0.01, 0.05, or 0.1 mg/kg), CPP (1, 5, or 10 mg/kg) or vehicle (VEH) was administered intraperitoneally 15 min prior to lorazepam (0.2, 0.5, 1, or 2 mg/kg) or vehicle ($n \geq 6$). Behavioral testing began 25 min after the lorazepam injection. Bars represent means response ± SEM. Significant differences ($p < 0.05$) from vehicle control or from 0.1 mg/kg dizocilpine are indicated by * and †, respectively, as determined by ANOVA and Dunnetts Multiple Comparison or Student–Newman–Keuls post hoc tests.

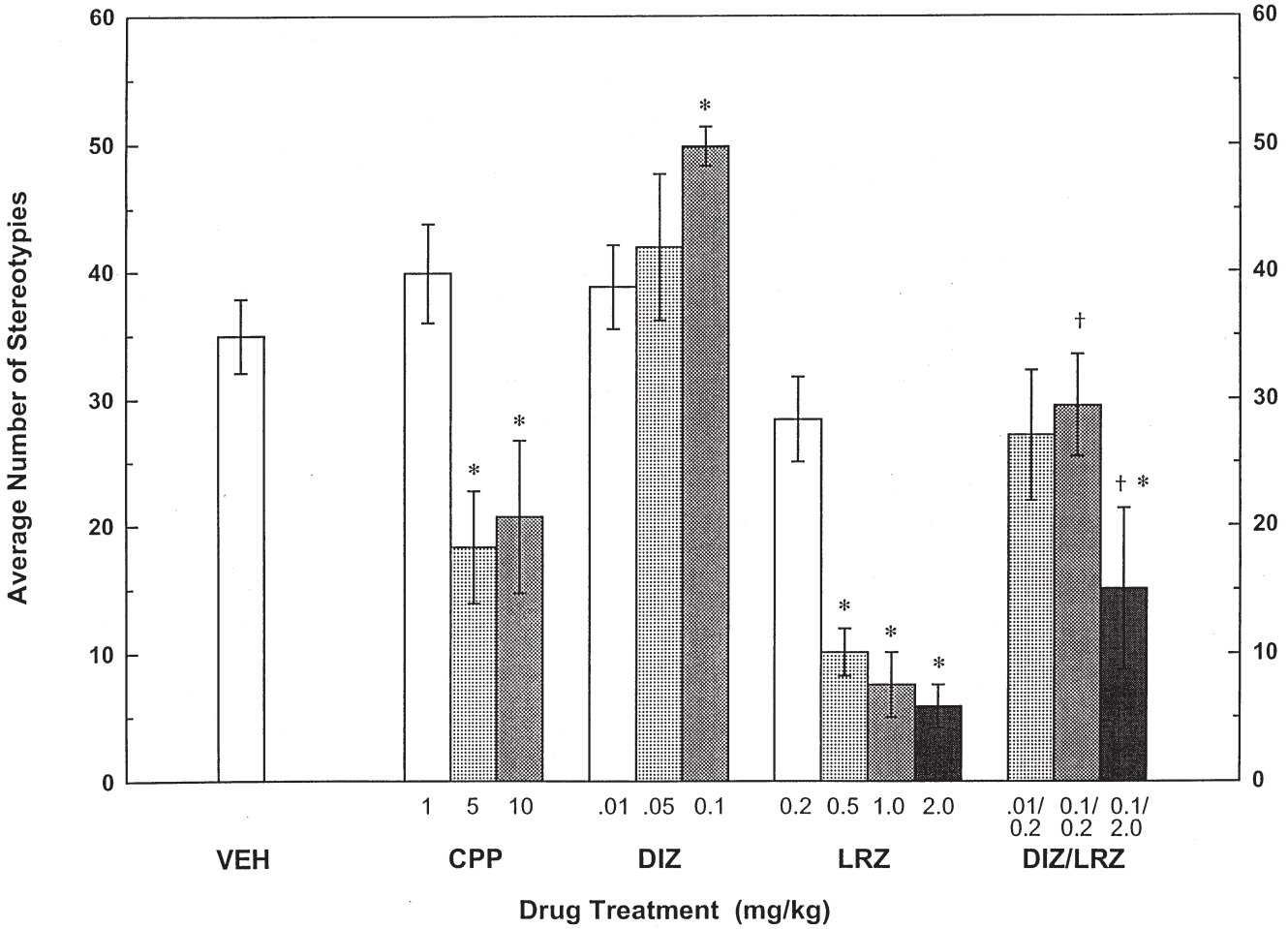

FIG. 3. Effect of CPP, dizocilpine (DIZ), and/or lorazepam (LRZ) on the average number of stereotypies. Data from number of steroetypies was recorded at 5-min intervals for a total of 20 min. Dizocilpine (0.01, 0.05, or 0.1 mg/kg), CPP (1, 5, or 10 mg/kg) or vehicle (VEH) was administered intraperitoneally 15 min before lorazepam (0.2, 0.5, 1, or 2 mg/kg) or vehicle ($n \geq 6$). Behavioral testing began 25 minutes after the lorazepam injection. Bars represent mean response ± SEM. Significant differences ($p < 0.05$) from vehicle control or from 0.1 mg/kg dizocilpine are indicated by * and † respectively, as determined by ANOVA and Dunnetts Multiple Comparison or Student-Newman-Keuls posthoc tests.

tetrazole (7.5 mg/ml) at a rate of 0.60 ml/min, beginning 15 min following the conclusion of open-field activity measurement. Infusion was terminated at the onset of a full tonic–clonic seizure, as determined by two trained observers. Seizure threshold was quantitated as the total dose of pentylenetetrazole (in mg/kg) at the time of seizure onset.

Data Analysis

Comparisons between groups were performed using analysis of variance and Dunnetts Multiple Comparison tests for 1) vehicle vs. CPP; 2) vehicle vs. dizocilpine, and 3) vehicle vs. lorazepam. Student–Neuman–Keuls comparisons were made among the vehicle, dizocilpine, lorazepam, and dizocilpine/lorazepam groups at single indicated concentrations.

RESULTS

Dizocilpine alone significantly increased ($p \leq 0.05$) all three behavioral parameters at a dose of 0.1 mg/kg (Figs. 1–3). The time course of drug effects on horizontal activity is presented in Fig. 4, and demonstrates increased activity at all time points. The two lower doses of dizocilpine (0.01 and 0.05 mg/kg) increased average total activity to a lesser extent, and differences from vehicle were not significant. CPP at 5 and 10 mg/kg reduced the average total distance traveled (Fig. 1), although the difference was not significant. CPP did not significantly decrease ($p \leq 0.05$), and the average total number of rears (Fig. 2) and the average total number of stereotypies (Fig. 3) at 5 and 10 mg/kg. As expected, the three highest doses (0.5, 1.0, and 2.0 mg/kg) of lorazepam dose dependently ($p \leq 0.05$) decreased the average total distance traveled in male Crl: CD-1(ICR)BR mice (Fig. 1). This decrease in open-field activity occurred at all time points (Fig. 4). Lorazepam also significantly decreased ($p \leq 0.05$) the average total number of rears (Fig. 2) and stereotypies (Fig. 3) in a dose-dependent manner at concentrations ranging from 0.5–2.0 mg/kg. Only the lowest dose of lorazepam (0.2 mg/kg) failed to have a significant effect on any of the parameters.

Coadministration of the highest doses of lorazepam (2 mg/kg) and dizocilpine (0.1 mg/kg) significantly decreased ($p \leq 0.05$) the average total number of rears and stereotypies com-

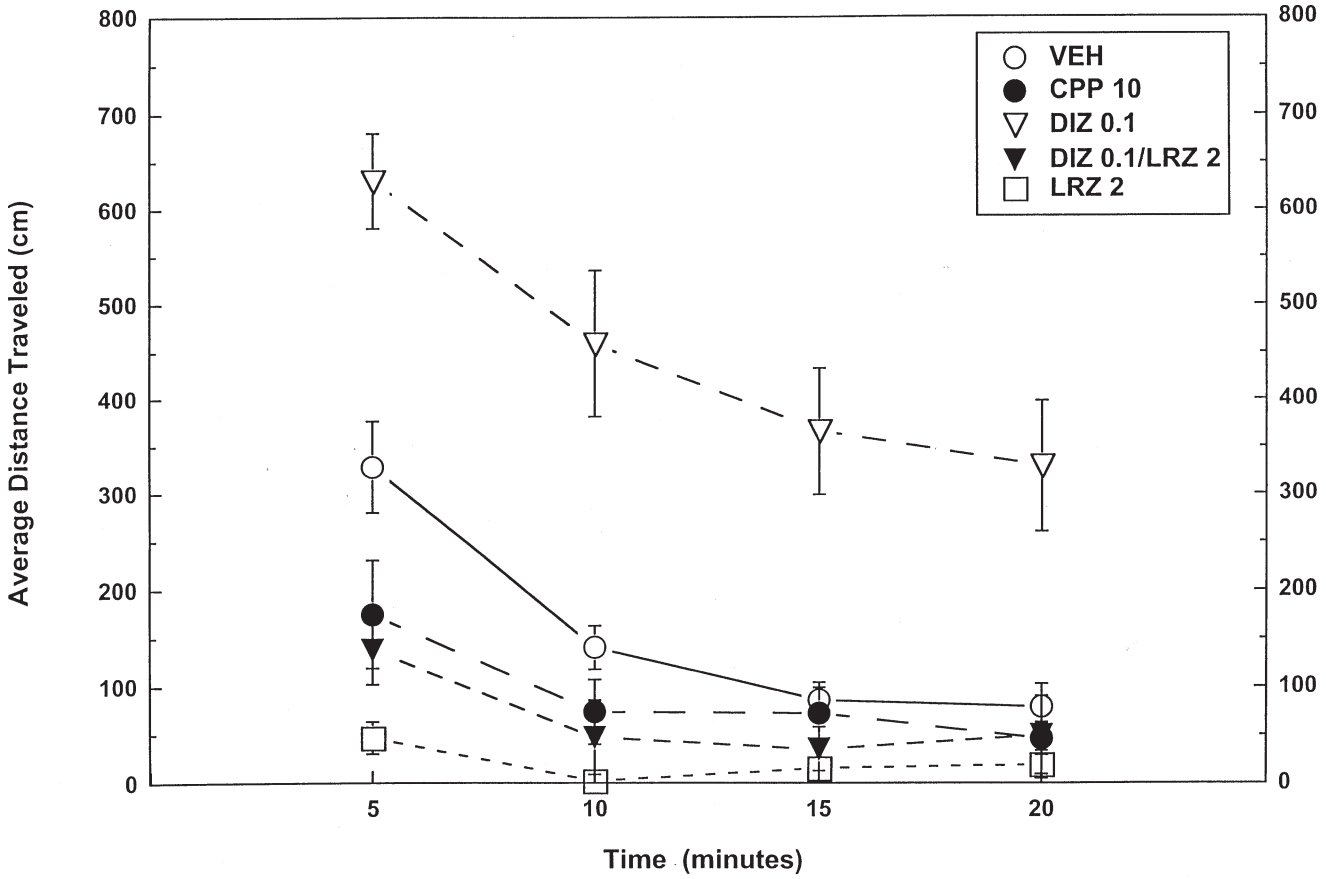

FIG. 4. Time course of CPP, dizocilpine (DIA), and/or lorazepam (LRZ) effects on the average distance traveled. Data from distance traveled was recorded at 5-min intervals for a total of 20 min. Dizocilpine (0.1 mg/kg), CPP (10 mg/kg), or vehicle (VEH) was administered intraperitoneally 15 min before lorazepam (2 mg/kg) or vehicle ($n \geq 6$). Behavioral testing began 25 min after the lorazepam injection. Symbols respresent mean response ± SEM.

pared to both vehicle-treated and dizocilpine-treated animals. The average distance traveled was also reduced compared to the dizocilpine-treated animals. Lorazepam at 0.2 or 2 mg/kg, when coadministered with 0.1 mg/kg dizocilpine, completely attenuated ($p \leq 0.05$) the increase in all three behavioral parameters produced by dizocilpine alone (Figs. 1–3). Values for animals coadministered lorazepam (0.2 or 2 mg/kg) and dizocilpine (0.1 mg/kg) were nearly identical to those for animals receiving the same dose or lorazepam alone. The combination of the lowest doses of lorazepam (0.01 mg/kg) and dizocilpine (0.2 mg/kg) had no significant effect on open-field activity.

Neither dizocilpine nor CPP had any effect on pentylenetetrazol-induced seizure threshold (Fig. 5). Lorazepam (0.2–2.0 mg/kg) exhibited a dose-dependent increase in the amount of pentylenetetrazol that was needed to induce a seizure, which paralleled alterations in open-field activity (Fig. 5). Coadministration of either the highest or lowest doses of dizocilpine and lorazepam significantly protected the animals from seizures compared to vehicle- or dizocilpine-treated groups, but was indistinguishable from those receiving lorazepam alone (Fig. 5).

DISCUSSION

Open-field behavior was evaluated as a pharmacodynamic parameter due to its noninvasive nature, simplicity of measurement, and the availability of data regarding acute effects of benzodiazepines (19). Horizontal activity, rears, and stereotypy have been previously shown to be the most sensitive and reliable measures following benzodiazepine administration. The observed effects of lorazepam on open-field activity are consistent with previous studies (4,9,13). Lorazepam uniformly and clearly decreased open-field activity on several different behavioral parameters. The behavioral results following administration of NMDA receptor antagonists are also consistent with previously published reports. Several investigators have observed hyperlocomotion following dizocilpine administration, which has been attributed to increased dopamine synthesis and metabolism (3,11,18,20). The lack of a uniformly significant effect of CPP on open-field activity is also consistent with Svensson and colleagues, who found, using a similarly automated system, that analogous doses (3–20 mg/kg) of D-CPPene decreased locomotor activity during the first 30 min of observation, had no effect at 30–60 min, and increased activity when the observation period was extended to 160 min (32). The contrasting effects of CPP and dizocilpine on open-field behavior may reflect a different mechanism of action, as several investigators have concluded that competitive NMDA antagonists, such as CPP, have little or no effect on dopamine metabolism (3,32). In contrast to effects observed with dizocilpine, other investigators have reported sed-

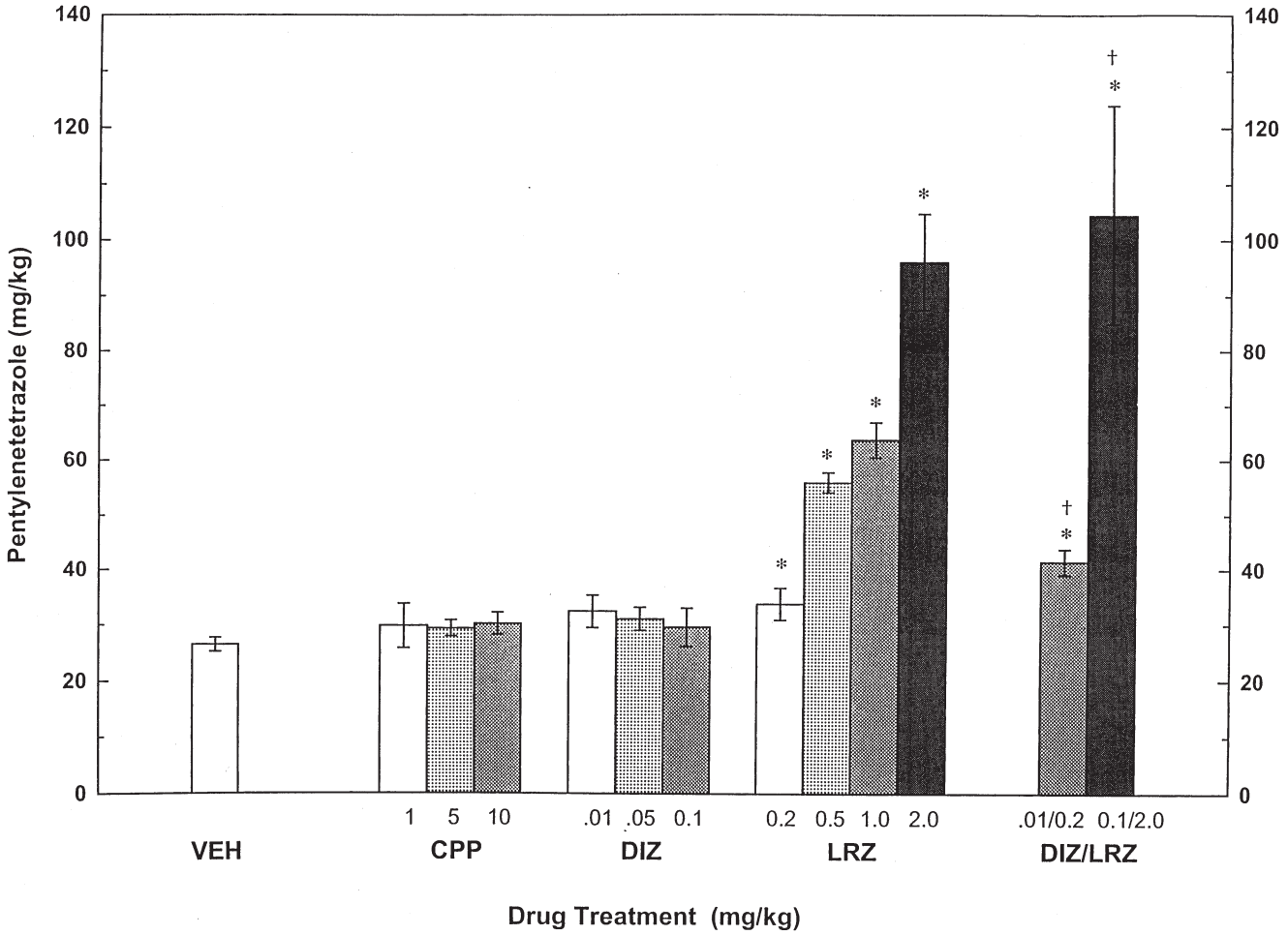

FIG. 5. Effect of CPP, dizocilpine (DIZ), and/or lorazepam (LRZ) on seizure protection. Dizocilpine (0.01, 0.05, or 0.1 mg/kg), CPP (1, 5, or 10 mg/kg) or vehicle (VEH) was administered intraperitoneally 15 min prior to lorazepam (0.2, 0.5, 1, or 2 mg/kg) or vehicle ($n \geq 3$). Fifteen minutes following the conclusion of open-field activity measurement, pentylenetetrazol (7.5 mg/ml) was infused via the tail vein at a rate of 0.6 ml/min, and was terminated at the induction of a clonic–tonic seizure. Bars represent mean response ± SEM. Significant differences ($p < 0.05$) from vehicle control or from 0.1 mg/kg dizocilpine are indicated by * and †, respectively, as determined by ANOVA and Dunnett's Multiple Comparison or Student–Newman–Keuls post hoc tests.

ative effects of both competitive and noncompetitive NMDA antagonists in both rats and mice using time on an inclined plane (TIP) and the elevated plus maze (8,16).

Coadministration of lorazepam and dizocilpine completely attenuated the hyperactivity on all three parameters seen with dizocilpine alone and returned activity levels to or below control values. This is in direct contrast to Behrens and Gattaz, who showed that diazepam caused a dose-dependent enhancement of the stereotypies induced by dizocilpine (1). It has been reported that the increase in spontaneous locomotion observed following dizocilpine administration can be attributed to stimulation of dopamine synthesis and release (18,32). Other investigators have found that benzodiazepines significantly reduce dopamine synthesis and release through action at the benzodiazepine binding site on the GABA receptor complex (14,24). This would result in a functional antagonism of the hyperlocomotion that is due to a dizocilpine-induced increase in dopaminergic tone. However, animals that received both compounds were similar to those that received lorazepam alone. Several investigators have demonstrated that glutamatergic system may modulate GABAergic responses via an increase in cytosolic calcium concentration (26,30). This mechanism of action is supported by the present results because the open channel blocker dizocilpine inhibits this NMDA-mediated calcium flux and would prevent modulation of the GABA receptor. Since the hyperlocomotion observed following dizocilpine administration may be due to a non-NMDA mechanism, the attenuation demonstrated with coadministration of dizocilpine and lorazepam does not conclusively indicate a functional interaction of the glutamatergic and GABAergic neurotransmitter systems. However, this argument is strengthened by the present data, which demonstrate that a dose of lorazepam, which by itself does not produce sedation, also reduces the hyperlocomotion seen with dizocilpine alone. In addition, previous work in our laboratory has shown that administration of CPP partly attenuated behavioral tolerance to chronic lorazepam, additional evidence of the functional interaction of these two systems (17).

Lorazepam alone exerted an anticonvulsant effect in this study that is consistent with previous work (4). In contrast,

neither dizocilpine nor CPP had any effect on pentylenetetrazol-induced seizures. Other investigators have demonstrated that both competitive and noncompetitive NMDA antagonists have anticonvulsant properties in a wide range of seizure models. These antagonists protect against electroconvulsive shock (7), sound-induced seizures in genetically susceptible mice and rats (5,6), kindled seizures (10,22), and chemically induced seizures (7,21). The lack of effect of CPP and dizocilpine in the present study could be due to the nature of the protocol used in our seizure model. Other models of pentylenetetrazol-induced seizures use a kindling protocol with repeated administration of the convulsant. Activation of the NMDA receptor complex, combined with inactivation of the GABA receptor complex, may be critical to the development of kindling (9). In that model, NMDA antagonists block the excitatory component in the model and inhibit the seizure. In an acute model, such as the one used in the present experiments, there may not be enough time for NMDA receptors to be activated above baseline levels. Therefore, inhibition of NMDA receptor function by either competitive or noncompetitive NMDA antagonists would be expected to have little or no effect. In addition, NMDA receptor channels need to be open for dizocilpine to bind and exert an effect. Alternately, if glutamatergic modulation of the GABA receptor complex is due to NMDA-mediated calcium flux, inhibition of this flux by CPP and dizocilpine prevents regulation of the GABA receptor and results in no effect on pentylenetetrazol-induced seizures. Coadministration of dizocilpine and lorazepam produced an anticonvulsant effect in this seizure model. However, administration of both compounds did not result in a larger response than administration of lorazepam alone. The inability of dizocilpine to alter the anticonvulsant effect of lorazepam is additional evidence that the attenuation in locomotor activity described above may not be due to a direct effect at the $GABA_A$/benzodiazepine site. It again implies that the functional interaction of dizocilpine and lorazepam may be via either the dopaminergic neurotransmitter system or the blockade of NMDA-mediated calcium influx.

The present study demonstrates the antagonism of dizocilpine-induced hyperactivity by coadministration of dizocilpine and lorazepam. This attenuation in all three behavioral parameters is likely due to an interaction between the GABAergic and glutamatergic neurotransmitter systems in regulating the dopaminergic tone which underlies motor activity. Alteration of NMDA receptor activity may have promise in limiting the negative symptoms of tolerance and dependence associated with chronic benzodiazepine administration. Similarly, the decreased function of the NMDA receptor complex with aging may underlie the acute sensitivity of aged animals to the hypnotic effects of benzodiazepines, which does not appear to be attributable to alterations in benzodiazepine binding, $GABA_A$ receptor function, or altered clearance of benzodiazepines.

ACKNOWLEDGEMENTS

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REFERENCES

1. Behrens, S.; Gattaz, W. F.: MK-801 induced stereotypies in rats are decreased by haloperidol and increased by diazepam. J. Neural Transm. 90:219–224; 1992.
2. Bhargava, H. N.: Diversity of agents that modify opioid tolerance, physical dependence, abstinence syndrome and self-administration behaviors. Pharmacol. Ref. 46:293–324; 1994.
3. Bubser, M.; Keseberg, U.; Notz, P. K.; Schmidt, W. J.: Differential behavioural and neurochemical effects of competitive and non-competitive NMDA receptor antagonists in rats. Eur. J. Pharmacol. 229:75–82; 1992.
4. Byrnes, J. J.; Miller, L. G.; Perkins, K.; Greenblatt, D. J.; Shader, R. I.: Chronic benzodiazepine administration XI. Concurrent administration of PK11195 attenuates lorazepam discontinuation effects. Neuropsychopharmacology 8:267–273; 1993.
5. Chapman, A. G.; Meldrum, B. S.: Non-competitive *N*-methyl-d-aspartate antagonists protect against sound-induced seizures in DBA/2 mice. Eur. J. Pharmacol. 166:201–211; 1989.
6. Chapman, A. G.; Meldrum, B. S.; Nanji, N.; Watkins, J. C.: Anticonvulsant action and biochemical effects in DBA/2 mice of CPP [3-((±)-2-carboxypiperazin-4-yl)-propyl-1-phosphonate], a novel *N*-methyl-d-aspartate antagonist. Eur. J. Pharmacol. 139:91–96; 1987.
7. Clineschmidt, B. V.; Martin, G. E.; Bunting, P. R.: Anticonvulsant activity of (+)-5-methyl-10,11-dihydro-5H-dibenzo(a,d)cyclohepten-5,10-amine (MK-801), a substance with potent anticonvulsant, central sympathomimetic and apparent anxiolytic properties. Drug Dev. Res. 2:123–134; 1982.
8. Corbett, R.; Dunn, R. W.: Effects of 5,7 dichlorokynurenic acid on conflict, social interaction and plus maze behaviors. Neuropharmacology 32:461–466; 1993.
9. Corda, M. G.; Orlandi, M.; Lecca, D.; Giorgi, O.: Decrease in GABAergic function induced by pentylenetetrazol kindling in rats: Antagonism by MK-801. J. Pharmacol. Exp. Ther. 262:792–800; 1992.
10. Croucher, M. J.; Bradford, H. F.; Sunter, D. C.; Watkins, J. C.: Inhibition of the development of electrical kindling of the prepiriform cortex by daily focal injections of excitatory amino acid antagonists. Eur. J. Pharmacol. 152:29–38; 1988.
11. Dai, H.; Gebhardt, K.; Carey, R. J.: Time course effects of MK-801: The relationship between brain neurochemistry and behavior. Brain Res. Bull. 36:175–180; 1995.
12. File, S. E.; Fernandes, C.: Dizocilpine prevents the development of tolerance to the sedative effects of diazepam in rats. Pharmacol. Biochem. Behav. 47:823–826; 1994.
13. Galpern, W. R.; Miller, L. G.; Greenblatt, D. J.; Shader, R. I.: Differential effects of chronic lorazepam and alprazolam on benzodiazepine binding and $GABA_A$-receptor function. Br. J. Pharmacol. 101:839–842; 1990.
14. Hegarty, A. A.; Vogel, W. H.: The effect of acute and chronic diazepam treatment on stress-induced changes in cortical dopamine in the rat. Pharmacol. Biochem. Behav. 52:771–778; 1995.
15. Herman, B. H.; Vocci, F.; Bridge, P.: The effects of NMDA receptor antagonists and nitric oxide synthase inhibitors on opioid tolerance and withdrawal. Neuropsychopharmacology 13: 269–293; 1995.
16. Kehne, J. H.; McCloskey, T. C.; Baron, B. M.; Chi, E. M.; Harrison, B. L.; Whitten, J. P.; Palfreyman, M. G.: NMDA receptor complex antagonists have potential anxiolytic effects as measured with separation-induced ultrasonic vocalizations. Eur. J. Pharmacol. 193:283–292; 1991.
17. Koff, J. M.; Pritchard, G. A.; Greenblatt, D. J.; Miller, L. G.: The NMDA receptor competitive antagonist CPP modulates benzodiazepine tolerance and discontinuation. Pharmacology 55:217–227; 1997.
18. Liljequist, S.; Ossowska, K.; Grabowska-Anden, M.; Anden, N. E.: Effect of the NMDA receptor antagonist, MK-801, on locomotor activity and on the metabolism of dopamine in various brain areas of mice. Eur. J. Pharmacol. 195:55–61; 1991.
19. Lopez, F.; Miller L. G.; Greenblatt, D. J.; Paul, S. M.; Shader, R. I.: Low-dose alprazolam augments motor activity in mice. Pharmacol. Biochem. Behav. 30:511–513; 1988.
20. Löscher, W.; Honack, D.: The behavioural effects of MK-801 in

rats: Involvement of dopaminergic, serotonergic and noradrenergic systems. Eur. J. Pharmacol. 215:199–208; 1992.
21. Löscher, W.; Nolting, B.; Honack, D.: Evaluation of CPP, a selective NMDA antagonist, in various rodent models of epilepsy. Comparison with other NMDA antagonists, and with diazepam and epilepsy. Comparison with other NMDA antagonists, and with diazepam and phenobarbitol. Eur. J. Pharmacol. 152:9–17; 1988.
22. McNamara, J. O.; Russel, R. D.; Rigsbee, L.; Bonhaus, D. W.: Anticonvulsant and antiepileptogenic actions of MK-801 in the kindling and electroshock models. Neuropharmacology 27:563–568; 1988.
23. Miller, L. G.; Greenblatt, D. J.; Roy, R. B.; Gaver, A.; Lopez, F.; Shader, R. I.: Chronic benzodiazepine administration. III. Upregulation of γ-aminobutyric $acid_A$ receptor binding and function associated with chronic benzodiazepine antagonist administration. J. Pharmacol. Exp. Therap. 248:1096–1101; 1989.
24. Murai, T.; Koshikawa, N.; Kanayama, T.; Takada, K.; Tomiyama, K.; Kobayashi, M.: Opposite effects of midazolam and beta-carboline-3-carboxylate ethyl ester on the release of dopamine from rat nucleus accumbens measured by in vivo microdialysis. Eur. J. Pharmacol. 261:65–71; 1994.
25. Pritchard, G. A.; Galpern, W. R.; Lumpkin, M.; Miller, L. G.: Chronic benzodiazepine administration VIII. Receptor upregulation produced by chronic exposure to the inverse agonist FG-7142. J. Pharmacol. Exp. Ther. 258:280–285; 1991.
26. Ragozzino, D.; Eusebi, F.: Inhibition of GABA and glycine responses by glutamate in rat hippocampal neurons. Brain Res. 628:115–120; 1993.
27. Schatzki, A.; Lopez, F.; Greenblatt, D. J.; Shader, R. I.; Miller, L. G.: Lorazepam discontinuation promotes "inverse agonist" effects of benzodiazepines. Br. J. Pharmacol. 98:451–454; 1989.
28. Shader, R. I.; Fogelman, S. M.; Greenblatt, D. J.: Newer antidepressants: Further reflections. J. Clin. Psychopharmacol. 17:75–77; 1997.
29. Skolnick, P.; Layer, R. T.; Popik, P.; Nowak, G.; Paul, I. A.; Trullas, R.: Adaptation of *N*-methyl-d-aspartate (NMDA) receptors following antidepressant treatment: Implications for the pharmacotherapy of depression. Pharmacopsychiatry 29:23–26; 1996.
30. Stelzer, A.; Shi, H.: Impairment of $GABA_A$ receptor function by *N*-methyl-d-aspartate-mediated calcium influx in isolated CA1 pyramidal cells. Neuroscience 62:813–828; 1994.
31. Steppuhn, K. G.; Turski, L.: Diazepam dependence prevented by glutamate antagonists. Proc. Natl. Acad. Sci. USA 90:6889–6893; 1993.
32. Svensson, A.; Pileblad, E.; Carlsson, M.: A comparison between the non-competitive NMDA antagonist dizocilpine (MK-801) and the competitive NMDA antagonist D-CPPene with regard to dopamine turnover and locomotor-stimulatory properties in mice. J. Neural Transm. 85:117–129; 1991.
33. Wisden, W.; Seeburg, P. H.: Mammalian ionotropic glutamate receptors. Curr. Opin. Neurobiol. 3:291–298; 1993.