

*Short Communication*

# Enhancement of Conditioned Place Preference Response to Cocaine in Rats Following Subchronic Administration of 3,4-Methylenedioxymethamphetamine (MDMA)

BRYAN HORAN,<sup>1</sup> ELIOT L. GARDNER,<sup>2</sup> AND CHARLES R. ASHBY, JR.<sup>1\*</sup>

<sup>1</sup>PHS Department, College of Pharmacy and Allied Health Professions, St. John's University, Jamaica, New York

<sup>2</sup>Departments of Psychiatry and Neuroscience, Albert Einstein College of Medicine, Bronx, New York

**KEY WORDS** conditioned place preference; cocaine; MDMA; vulnerability; ecstasy; drug abuse; dependence

**ABSTRACT** In this study, we measured conditioned place preference (CPP) responses to cocaine following subchronic administration of the recreationally abused drug ( $\pm$ )-3,4-methylenedioxymethamphetamine (MDMA, "ecstasy") in male Sprague-Dawley rats. Animals were given either vehicle (1 ml/kg of distilled water, s.c.) or MDMA (20 mg/kg, s.c.) twice a day for 4 consecutive days. Two weeks later, CPP responses to cocaine (5, 10, or 20 mg/kg, i.p.) were measured. The MDMA-treated animals showed a significantly greater CPP response to cocaine than the vehicle-treated animals. Since conditioned place preference is believed to be a measure of appetitive behavior, these results suggest that MDMA abuse could lead to an increased vulnerability to the rewarding actions of cocaine and, hence, to increased vulnerability to cocaine addiction and dependence. **Synapse 35:160–162, 2000.** © 2000 Wiley-Liss, Inc.

The recreationally abused drug 3,4-methylenedioxymethamphetamine (MDMA, "ecstasy") appears to be neurotoxic to serotonergic neurons in the mammalian brain. The administration of MDMA primarily destroys fine serotonergic nerve terminals and significantly decreases serotonin uptake sites and serotonin content (Battaglia et al., 1990). However, despite these well-documented findings little is known about the functional consequences of serotonergic neurotoxicity following the administration of MDMA.

It is well accepted that serotonergic neurons innervate and functionally modulate the mesocorticolimbic dopamine system. This innervation and functional modulation is complex, with serotonin inhibiting dopaminergic function in some cases and facilitating it in others (Kelland and Chiodo, 1996). Considerable evidence indicates that depletion of brain serotonin can alter the behavioral response of animals to compounds that augment dopaminergic activity. For example, the lesioning of serotonergic neurons by the neurotoxin 5,7-dihydroxytryptamine increases breaking points on a progressive ratio schedule rein-

forced by intravenous cocaine (Roberts, 1992) and increases self-administration of D-amphetamine on a fixed ratio schedule (Leccese and Lyness, 1984). In contrast, treatments which augment serotonergic activity appear to attenuate the reinforcing actions of cocaine and amphetamine (Roberts, 1992).

In light of this evidence, the possibility arises that MDMA use could, by acting on serotonergic mechanisms, alter cocaine's rewarding value and abuse potential. This possibility becomes especially troubling in view of the fact that MDMA abuse has been on the rise since the early 1990s, particularly in the 13–18-year-old age group (Mathias, 1997). In addition, MDMA is a commonly used recreational drug at "rave parties" by

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\*Correspondence to: Charles R. Ashby, Jr., Ph.D., PHS Department, College of Pharmacy and Allied Health, St. John's University, 8000 Utopia Parkway, Jamaica, NY 11439. E-mail: CRASHBY@IX.NETCOM.COM.

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16–24-year-olds. Thus, the issue arises that recent increases in MDMA abuse may lead to increased vulnerability to cocaine abuse. To examine this possibility, we measured the conditioned place preference (CPP) response to cocaine following administration of a subchronic regimen of MDMA in male Sprague-Dawley rats. In the CPP paradigm, animals are tested in a drug-free state to determine whether they prefer an environment in which they previously received cocaine injections as compared to an environment in which they previously received vehicle injections. If the animal, in a drug-free state, consistently chooses the environment previously associated with cocaine, the inference is drawn that the appetitive value of cocaine was encoded in the brain and is accessible in the drug-free state (Gardner, 1997).

Male Sprague-Dawley rats (175–200 g at the beginning of the experiment; Taconic Farms, Germantown, NY) were given either MDMA (20 mg/kg, s.c.) or vehicle (1 ml/kg of distilled water, s.c.) twice daily for 4 consecutive days. Two weeks after the last injection, animals were habituated to the experimental room and handled for 5 days prior to the CPP experiments. A three-chambered CPP apparatus, as described previously (Horan et al., 1997b), was used, with modifications. Prior to the start of the CPP experiments, the animals were allowed to freely explore the apparatus for 15 min. Animals ( $n = 8$  per group) were randomly assigned to receive either vehicle (1 ml/kg of distilled water, s.c.) or 5, 10, or 20 mg/kg, i.p., of (-)-cocaine in either of the conditioning chambers in a counterbalanced design. Each animal received four pairings each (one injection per day) with saline or cocaine. The animals were confined to one of two conditioning chambers for 30 min during each pairing session. On the test day, the animals were allowed access to the entire apparatus for 15 min. The amount of time spent in each chamber was recorded using an automated recording device. The data were expressed as the raw time spent in each chamber. The data were analyzed using a one-way ANOVA followed by Tukey's post-hoc test.

The data are presented in Table I. The administration of vehicle (0 mg/kg cocaine) did not produce a place preference in either the MDMA or vehicle-pretreated animals. Above the 0 mg/kg control dose, cocaine produced a dose-dependent increase in place preference in both the MDMA- ( $F(3,28) = 6.84$ ,  $P < 0.002$ ) and vehicle-pretreated ( $F(3,28) = 12.4$ ,  $P < 0.0001$ ) rats. However, statistical analysis indicated that the CPP response to cocaine was significantly greater in the MDMA-pretreated animals compared to the vehicle-pretreated animals ( $F(1,62) = 5.74$ ,  $P < 0.019$ ). Post-hoc statistics revealed that the cocaine CPP response in the MDMA-pretreated animals was significantly greater

TABLE I. Effect of subchronic administration of vehicle or MDMA on the conditioned place preference response to cocaine

| Pretreatment | Dose of cocaine (mg/kg) | Time spent in chambers      |               |
|--------------|-------------------------|-----------------------------|---------------|
|              |                         | Paired                      | Non-paired    |
| Vehicle      | 0                       | 7.2 $\pm$ 0.7               | 7.8 $\pm$ 0.6 |
|              | 5                       | 7.8 $\pm$ 0.5 <sup>††</sup> | 7.2 $\pm$ 0.5 |
|              | 10                      | 9.3 $\pm$ 0.4*              | 5.7 $\pm$ 0.4 |
|              | 20                      | 11.5 $\pm$ 0.7**            | 3.5 $\pm$ 0.7 |
| MDMA         | 0                       | 7.1 $\pm$ 0.6               | 7.9 $\pm$ 0.8 |
|              | 5                       | 9.7 $\pm$ 0.6***            | 5.3 $\pm$ 0.5 |
|              | 10                      | 11.2 $\pm$ 0.6 <sup>†</sup> | 3.8 $\pm$ 0.4 |
|              | 20                      | 11.6 $\pm$ 1.2 <sup>†</sup> | 3.5 $\pm$ 0.7 |

Animals received subcutaneous injections of either vehicle (distilled water, 1 ml/kg) or MDMA twice daily for four consecutive days. Two weeks later, the pairings with vehicle or cocaine were started. Dose 0 represents the animals that were paired with vehicle. A total of eight animals were examined at each dose of cocaine for both vehicle- and MDMA-pretreated animals.

\*Significantly greater than 0 mg/kg cocaine and significantly less than MDMA pretreated rats,  $P < 0.05$ , ANOVA + Tukey's post hoc test.

\*\*Significantly greater than 0, 5 or 10 mg/kg cocaine,  $P < 0.01$ , ANOVA + Tukey's post hoc test.

\*\*\*Significantly greater than 0 mg/kg cocaine,  $P < 0.05$ , ANOVA + Tukey's post hoc test.

<sup>†</sup>Significantly greater than 0 mg/kg cocaine,  $P < 0.01$ , ANOVA + Tukey's post hoc test.

<sup>††</sup>Significantly less than MDMA pretreated rats,  $P < 0.05$ , ANOVA + Tukey's post hoc test.

at the 5 and 10 mg/kg doses of cocaine compared to vehicle-pretreated animals.

These results are the first to show that the CPP response to cocaine is significantly increased after administration of MDMA (20 mg/kg s.c., twice a day for 4 consecutive days). Our findings are consistent with studies indicating that depletion of brain serotonin alters the rewarding effects of cocaine. It is unlikely that MDMA per se had a direct effect on cocaine's action because on the test day, no MDMA was present in the rat brain since it had been administered 2 weeks earlier, which is significantly longer than its half-life. In addition, MDMA was administered to the rats in their home cages and thus was not paired with either one of the CPP chambers. Our findings are unlikely related to an MDMA-induced changes in basal DA levels or number of DA neurons, as the MDMA regimen used in this study does not significantly alter brain DA levels or DA uptake transporters in rats (Battaglia et al., 1990).

The explanation for our present findings are not clear. Given that depletion of brain serotonin increases the self-administration of psychostimulants, it is possible that MDMA administration may augment the rewarding/appetitive action of cocaine via its depletion of brain serotonin, subsequently altering the responsiveness of brain dopaminergic systems. Indeed, we have previously shown (using in vivo brain microdialysis) that the increase in extracellular dopamine levels in the nucleus accumbens in male rats produced by a single injection of 20 mg/kg, i.p. of cocaine was significantly greater in animals pretreated with the same MDMA regimen used in this study compared to vehicle-pretreated rats (Horan et al., 1997a). Furthermore, we have shown that the MDMA regimen used in this study

will produce a 70% depletion of 5-HT levels in male Sprague-Dawley rats of the same age and from the same supplier as this study (Granoff and Ashby, 1998). In contrast, midbrain DA levels are not altered. Alternatively, it has been reported that rats show a sensitization to a challenge injection of MDMA following the administration of 20 mg/kg of MDMA twice a day for 4 consecutive days (Spanos and Yamamoto, 1989). Therefore, the presently observed increased CPP response to cocaine following MDMA treatment could be related to sensitization. However, depletion of serotonin by other pharmacologic agents such as 5,7-dihydroxytryptamine, which does not produce sensitization, augments cocaine's rewarding actions (Lecesse and Lyness, 1984; Roberts, 1992). Finally, it seems possible that MDMA-induced depletion of 5-HT may have diminished aversive effects elicited by cocaine, thereby increasing the time the animals spent in the cocaine-paired chamber. Treatments that augment or decrease brain 5-HT levels decrease (McGregor et al., 1993; Richardson and Roberts, 1991) and increase (Loh and Roberts, 1990), respectively, cocaine self-administration on a progressive ratio schedule in rats. Moreover, cocaine is an inhibitor of 5-HT uptake in the brain.

In conclusion, our results indicate that the CPP response to cocaine is augmented following the repeated administration of MDMA. Extrapolating these findings to humans, one might postulate that prior exposure to MDMA might increase vulnerability to cocaine use. We intend to further study this possibility by assessing the effect of MDMA pretreatment on self-administration of cocaine in laboratory animals.

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