

**DIMINISHED CORTICOTROPIN AND ENHANCED PROLACTIN RESPONSES
TO 8-HYDROXY-2(DI-N-PROPYLAMINO)TETRALIN IN
METHYLENEDIOXYMETHAMPHETAMINE PRETREATED RATS**

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Summary

Although ($\pm$) methylenedioxymethamphetamine (MDMA) has been reported to deplete serotonin (5-HT) and destroy 5-HT terminals in the brains of animals, the functional sequelae of such alterations remain to be established. In the present study, a blunted corticotropin and an enhanced prolactin response to the 5-HT_{1A} agonist 8-hydroxy-2(di-n-propylamino)tetralin (8-OH DPAT) was found in rats treated two weeks previously with a single dose of MDMA (2.0 or 20.0 mg/kg, sc). These results suggest that neurochemical changes produced by MDMA are associated with functional alterations as manifested by abnormal 5-HT receptor-coupled neuroendocrine responses.

Key words: 3,4-methylenedioxymethamphetamine (MDMA), corticotropin, adrenocorticotrophic hormone (ACTH), prolactin, serotonin, 8-hydroxy-2(di-n-propylamino)tetralin (8-OH DPAT)

Evidence indicates that methylenedioxymethamphetamine (MDMA) can be neurotoxic to serotonergic (5-HT) systems in the brains of rats and nonhuman primates (Ricaurte, DeLanney, Irwin and Langston, 1988; McKenna and Peroutka, 1990). However, it is unknown if, and to what extent, a functional impairment is associated with the neurochemical changes. Since the central nervous system is the predominant regulator of neuroendocrine function, with pituitary secretion of adrenocorticotrophic hormone (ACTH) and prolactin being strongly influenced by 5-HT pathways in the brain (Tuomisto and Mannisto, 1985), a potential functional measure of 5-HT neurotoxicity would be to assess changes in response of these hormones to administration of 5-HT agonists. In the present study, rats were injected with a single dose of saline or one of two doses of MDMA (2.0 or 20.0 mg/kg, sc), and two weeks later the stimulatory effects of 8-hydroxy-2(di-n-propylamino)tetralin (8-OH DPAT), a 5-HT_{1A} agonist, on ACTH (Gilbert, Brazell, Tricklebank and Stahl, 1988) and prolactin (Simonovic, Gudelsky and Meltzer, 1984) secretion were assessed.

Methods

Adult VAF male Sprague Dawley rats (200-225 g) were purchased from Charles River Laboratories. Animals were maintained under a 12-12 light-dark cycle with lights on at 0400 h and housed four to five per cage with food and water available *ad libitum*. After one week of acclimatization to the vivarium, rats were injected at 0600 h with saline or one of two doses of ($\pm$) MDMA (2.0 or 20.0 mg/kg, sc) and, thereafter, handled daily.

At 0600 h 14 days later, the rats were injected with saline or 8-OH DPAT (0.1 mg/kg, sc), the approximate ED₅₀ and ED₉₀ for stimulation of ACTH (Gilbert, Brazell, Tricklebank and Stahl, 1988) and prolactin (Simonovic, Gudelsky and Meltzer, 1984; unpublished observations) secretion, respectively. Animals were sacrificed by decapitation 20 minutes after saline or 8-OH DPAT administration, and trunk blood was collected into chilled plastic tubes containing aprotinin (1250 KIU), EDTA (7.5 mg) and sodium azide (0.3 mg). Plasma concentrations of ACTH and prolactin were measured by specific radioimmunoassays (Poland, Hanada and Rubin, 1989; Pechnick, George and Poland, 1989). All samples for each hormone were analyzed in triplicate in the same assay. Maximum intra-assay variability was 12%. Data were analyzed by analysis of variance (ANOVA) and t-tests. Significance levels were corrected for multiple t-tests. A $p < 0.05$ was considered statistically significant.

Results

As determined by ANOVA, there was a significant effect of treatment on both plasma ACTH ($p < .001$) (Figure 1) and prolactin concentrations (Figure 2) ($p < .001$). MDMA did not affect either the basal hormone concentrations or the neuroendocrine responses to acute saline administration (all p values $> .4$). Compared to non-injected and saline

injected animals, 8-OH DPAT significantly increased both ACTH ($p < .001$) and prolactin ($p < .001$) concentrations.

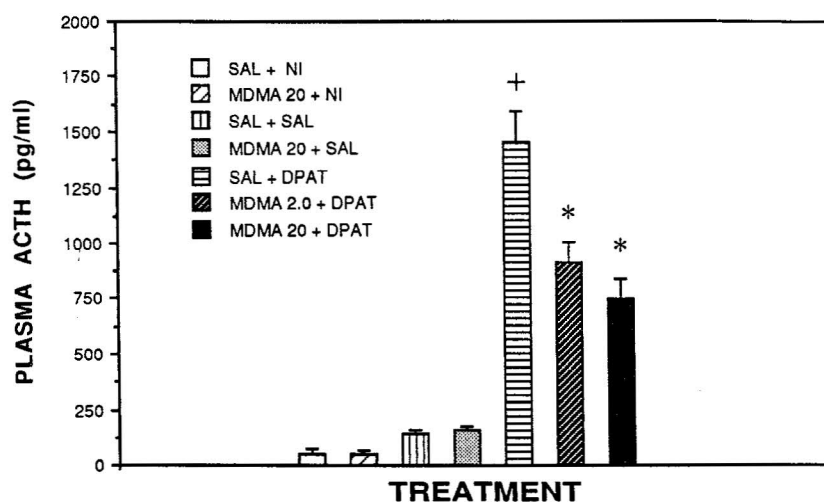


Figure 1. Mean ($\pm$ SEM) ACTH responses to 8-OH DPAT (0.1 mg/kg, sc) in male rats treated 14 days previously with saline (SAL) or one of two doses of MDMA (2.0 or 20.0 mg/kg, sc). MDMA (20.0 mg/kg) pretreatment did not affect plasma ACTH concentrations in non-injected (NI) or SAL injected animals. Compared to the NI and SAL animals, 8-OH DPAT significantly increased plasma ACTH concentrations (+ = $p < .001$). The ACTH response to 8-OH DPAT was significantly blunted in animals treated with either dose of MDMA (* = $p < .01$). There was no significant difference in the ACTH responses to 8-OH DPAT between the two doses of MDMA. (n = 10-12/group)

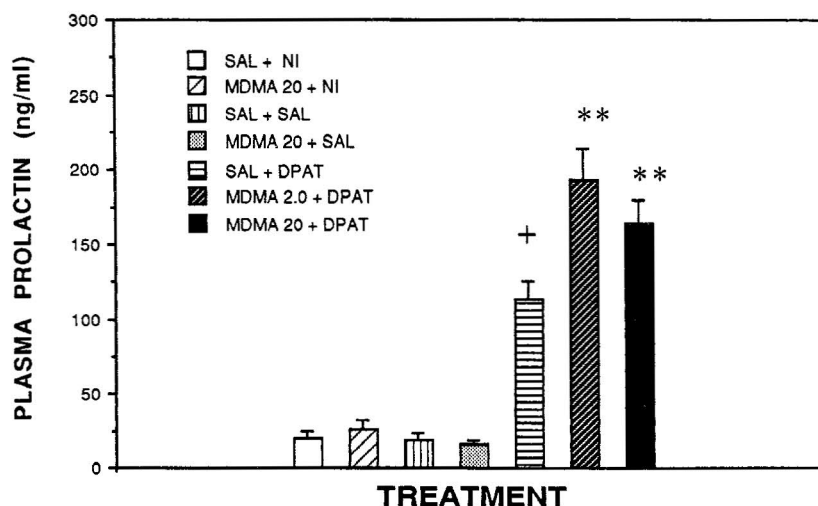


Figure 2. Mean ($\pm$ SEM) prolactin (PRL) responses to 8-OH DPAT (0.1 mg/kg, sc) in male rats treated 14 days previously with saline (SAL) or one of two doses of MDMA (2.0 or 20.0 mg/kg, sc). MDMA (20.0 mg/kg) pretreatment did not affect plasma PRL concentrations in non-injected (NI) or SAL injected animals. Compared to the NI and SAL animals, 8-OH DPAT significantly increased plasma PRL concentrations (+ = $p < .001$). The PRL response to 8-OH DPAT was significantly enhanced in animals treated with either dose of MDMA (** = $p < .02$). There was no significant difference in the PRL responses to 8-OH DPAT between the two doses of MDMA. (n = 10-12/group)

Animals pretreated with either 2.0 or 20 mg/kg MDMA two weeks previously demonstrated a significantly blunted ACTH ($p < .01$) and a significantly augmented prolactin ($p < .02$) response to 8-OH DPAT compared to animals pretreated with saline and challenged with 8-OH DPAT. There were no dose-dependent effects of MDMA, as both doses of MDMA affected the ACTH and prolactin responses to 8-OH DPAT in a comparable fashion (all p values $> .3$).

Discussion

It has been reported that administration of a single dose of MDMA (20 mg/kg) to rats, a dose identical to one of the doses used in the present study, reduces brain 5-HT concentrations for at least two months (Commins, Vosmer, Virus, Woolverton, Schuster and Seiden, 1987). Similarly, a depletion of 5-HT in the thalamus and hypothalamus was found in monkeys when assessed two weeks after a single dose of MDMA (5.0 mg/kg, PO) (Ricaurte, DeLanney, Irwin and Langston, 1988). However, comparable long-term neurochemical alterations have not been reported in rodents or primates following the administration of a single dose of 2.0 mg/kg MDMA, a dosage comparable to that ingested by humans (Peroutka, 1987; Price, Ricaurte, Krystal and Heninger, 1989).

In the present study, both doses of MDMA affected the ACTH and prolactin responses to a single dose of 8-OH DPAT, as measured at a single timepoint, suggesting that the functional integrity of 5-HT_{1A} receptor-coupled neuroendocrine responses has been altered. Since the exact contribution of pre- and post-synaptic 5-HT_{1A} receptors to the regulation of ACTH and prolactin secretion is unclear, the mechanisms underlying the differential effect of MDMA on the 8-OH DPAT induced secretion of these hormones remains to be determined. Nevertheless, if the altered neuroendocrine responses to 8-OH DPAT are a reflection of MDMA induced neurotoxicity, then the results would suggest that, under conditions of provocation, there might be significant health consequences for humans who ingest MDMA even once (Peroutka, 1987).

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