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MDMA (3,4-methylenedioxymethamphetamine) inhibits the firing of dorsal raphe neurons in brain slices via release of serotonin

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The effects of MDMA (3,4-methylenedioxymethamphetamine) on the activity of serotonin (5-HT)-containing dorsal raphe neurons were characterized using extracellular single-unit recording and microdialysis techniques in the in vitro midbrain slice preparation. Addition of (+)-MDMA, (−)-MDMA or p-chloroamphetamine (PCA) to the superfusate (final concentration 3-100 μM) produced a concentration-dependent inhibition of 5-HT cell firing which was reversible and reproducible. Based upon IC_{50} values, (+)-MDMA was 2- to 3-fold more potent than (−)-MDMA. Pretreatment with the selective 5-HT uptake inhibitor fluoxetine, at a concentration which had no effect on baseline firing (20 μM), blocked the inhibitory effect of (+)-MDMA and PCA on dorsal raphe neurons. The selective norepinephrine uptake inhibitor desipramine (20 μM) was ineffective. In a parallel series of experiments, microdialysis probes resting on the brain slice surface provided a means to estimate 5-HT release from the dorsal raphe nucleus. (+)-MDMA (100 μM) caused the release of measureable quantities of 5-HT with a time course which corresponded to the change in dorsal raphe cell firing rate. Taken together, these data suggest that MDMA acts indirectly to inhibit dorsal raphe neurons through release of endogenous 5-HT.

MDMA (3,4-methylenedioxymethamphetamine); Dorsal raphe nucleus; Single unit recording; Microdialysis; Neurotransmitter release

1. Introduction

The synthetic amphetamine derivative MDMA (3,4-methylenedioxymethamphetamine) is a novel psychedelic agent whose neurochemical effects closely resemble those of the serotonergic neurotoxin, p-chloroamphetamine (PCA). Like PCA, administration of MDMA produces an immediate and a long-term decrease in brain serotonin (5-HT) (Schmidt, 1987; Schmidt et al., 1987). The acute depletion appears reversible and may be due to inhibition of transmitter synthesis and stimulation of carrier-mediated 5-HT release. These effects on brain 5-HT are relatively selective in that both MDMA and PCA are more potent in releasing [^{3}H]5-HT than [^{3}H]dopamine. Release of both neurotransmitters by MDMA is stereoselective with (+)-MDMA the more potent releasing agent in vitro and independent of Ca^{2+} in that removal of this ion from the superfusion media is not inhibitory (Schmidt et al., 1987). The delayed effects on brain 5-HT measured 1 week following drug administration have been correlated with a decrease in the number of [^{3}H]5-HT uptake sites with no change in the affinity of the carrier for 5-HT (Schmidt, 1987; Schmidt et al., 1987). The persistent decrease in 5-HT concentration is restricted to the serotonergic system and may represent selective nerve terminal damage (Schmidt et

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al., 1986; Stone et al., 1986; Schmidt et al., 1987). Such a neurotoxic effect is the property of the (+) stereoisomer only. Pretreatment with a 5-HT uptake inhibitor blocks both the acute and long-term depletion of 5-HT (Schmidt et al., 1987; Fuller and Wong, 1987; Battaglia et al., 1988b).

The relationship between these neurochemical effects and the electrophysiological responses which they may elicit has been examined for PCA (Sheard, 1974). In chloral hydrate-anesthetized rats, PCA in doses of 0.25-2.5 mg/kg i.v. reversibly suppressed the spontaneous firing of serotonergic dorsal raphe neurons, presumably as a result of a transient increase in extracellular 5-HT. The purpose of the present study was to compare the effects of (+)-MDMA and (−)-MDMA to PCA on dorsal raphe neurons in the midbrain slice preparation. The ability of fluoxetine to block PCA- and MDMA-induced electrophysiological responses was also assessed. In parallel experiments, the effect of (+)-MDMA on the efflux of endogenous 5-HT from raphe slices was monitored by microdialysis.

2. Materials and methods

2.1. Brain slice preparation

Brain slices were obtained from male rats (125-175 g) anesthetized with chloral hydrate (400 mg/kg i.p.) and perfused with ice-cold sucrose-enriched artificial cerebrospinal fluid (ACSF) before decapitation. The sucrose-enriched ACSF was of a standard composition (in mM: KCl 5.0, $CaCl_2$ 2.0, $MgSO_4$ 2.0, $NaHCO_3$ 28, $NaHPO_4$ 1.25, D-glucose 10) except that NaCl, normally present at 126 mM, was replaced by a calculated equiosmolar concentration of sucrose (252 mM). The substitution of sucrose for NaCl in the ACSF for the preparatory and recovery phases of the experiment improved viability (Aghajanian and Rasmussen, in press). Immediately following decapitation, the brain was removed rapidly, placed in a petri dish containing ice-cold sucrose-enriched ACSF and trimmed to expose the posterior midbrain. Coronal slices (500 μm) then were cut through the dorsal raphe using a vibrating-knife microtome (WPI, Vibroslice) and transferred to the stage of a gas-fluid interface brain slice chamber modified from the design of Haas et al. (1979).

The stage of the slice chamber was raised so that humidified 95% O_2 - 5% CO_2 was able to circulate below as well as above the slice. It was constructed by mounting a polypropylene mesh onto a rectangular plastic frame $3.4 \times 2.2 \times 0.4$ cm. A thin layer of paraffin was used to coat the stage (including the mesh) to prevent diversion of fluid away from the top of the stage. A strip of lens paper (Macalaster Bicknell, No. 32215) 2.3 cm wide and 12 cm long was placed over the stage to serve as a wick drawing fluid across the stage to a lower collecting container. Sucrose-enriched ACSF, warmed (33° C) and oxygenated, reached the stage via a small spout. Upon placement of the brain slice on the lens paper, a thin strand of surgical gauze was arranged to draw fluid directly from the spout onto the slice. One hour from the time of decapitation, sucrose-enriched ACSF was replaced by standard ACSF containing NaCl. An additional period of 1 h was allowed prior to recording.

The otherwise silent 5-HT neurons in the brain slice were induced to fire with 2.5 μM phenylephrine in the ACSF (Vandermaelen and Aghajanian, 1983). Solutions of serotonin creatinine sulfate (Regis Chemical Co., Morton Grove, IL), p-chloroamphetamine HCl (PCA; Regis) and the two stereoisomers of (±)-3,4-methylenedioxymethamphetamine HCl, (+)-MDMA and (−)-MDMA (NIDA), in standard ACSF containing phenylephrine were introduced into the slice chamber by means of a stopcock arrangement. The perfusion fluid routinely flowed at 1-1.5 ml/min. The period of drug applications was fixed at 3 min; the period of pretreatment with fluoxetine HCl (Eli Lilly and Company, Indianapolis, IN) or desipramine HCl (Sigma) was minimally 10 min.

2.2. Recording

Extracellular single unit recordings were performed using single barrel glass micropipettes filled with 2% pontamine sky blue in 2 M NaCl. Tip diameters ranged from 1-2 μm; in vitro imped-

ances ranged from 2-4 MΩ when measured at 60 Hz. A Burleigh Inchworm microdrive in conjunction with a stereotaxic apparatus was used for electrode positioning. The electrode signal was passed through a high-impedance amplifier and monitored on an oscilloscope. A variable time and voltage window discriminator facilitated the discrimination of single units. Integrated firing rate was computed in 10 s samples from an electronic counter triggered by individual neuronal spikes. Putative 5-HT neurons were identified on the basis of their wide-duration action potentials (1-2 ms, positive-negative spikes), regular rhythm and slow firing rate (Vandermaelen and Aghajanian, 1983). Cells with these characteristics in the dorsal raphe nucleus have been confirmed to be 5-HT neurons by intracellular double-labeling (Aghajanian and VanderMaelen, 1982).

2.3. Data analysis

The percentage suppression of firing was calculated by comparing the baseline firing rate for a 3 min period prior to drug exposure to the maximal drug response, which occurred within 5 min following drug application. In each cell recorded, a concentration-response relationship was determined for a given compound. The concentration required to produce a 50% suppression of cell firing (IC_{50}) was interpolated following linear regression analysis. Differences were determined by Student's t-test.

2.4. Estimation of 5-HT release

In a parallel series of experiments, microdialysis probes were positioned directly on the brain slice surface, providing a means to estimate 5-HT release from the dorsal raphe nucleus. Probes were constructed using Cuprophan (Enka, West Germany) hollow fibers (300 μm I.D., 330 μm O.D.) housed in a section of 23 gauge (0.64 mm O.D.) hypodermic tubing. The fiber extended approximately 2.0-2.5 mm beyond the tip of the tubing exposing an active surface of 1.5-2.0 mm. Perfusion buffer (in mM: KCl 2.4, NaCl 120, $CaCl_2$ 1.2, $MgCl_2$ 1.2, NaH_2PO_4 0.9, Na_2HPO_4 1.4, ascorbic acid 0.3, pH 7.4) was pumped through a section of vitreous silica (170 μm O.D., Scientific Glass Engineering) which extended to the tip of the hollow fiber. The hypodermic tubing containing the dialysis assembly was bent 90° to allow the probe to rest on the brain slice surface over the dorsal raphe nucleus and perpendicular to the flow of ACSF over the slice. Collection periods were fixed at 10 or 20 min, with flow rate set at 2.1 or 1.1 μl/min, respectively. The average 'percentage recovery' (Ungerstedt, 1984) for these probes at a flow rate of 1.1 μl/min was 11 ± 4; recovery at 2.1 μl/min averaged 9 ± 2.

5-HT content in dialysate aliquots was determined by HPLC with electrochemical detection. Columns, 10 × 2.1 mm I.D., packed with 3 μm C-18 particles (Shandon) were used in conjunction with a thin-layer amperometric detection electrode assembly (Bioanalytical Systems). A lab-designed potentiostat (Bradberry et al., in preparation) was used for the application of a potential to the detector electrode and conversion of the resulting current to voltage for output to a strip chart recorder. A pneumatic fluid pump (Bradberry and Roth, in preparation) was used for pumping mobile phase through the column, providing extremely smooth flow and isolation from electrical noise. Mobile phase consisted of 0.05 M Na_2HPO_4, 350 mg/l sodium octanesulfonate, 0.1 mM disodium EDTA, 300 μl/l triethylamine and 150 ml/l methanol. pH was unadjusted and was 5.6. Flow rate was 0.2 ml/min. The limit of detection attainable was routinely 0.1 nM injected in volume of 20 μl or 2 fmol.

3. Results

3.1. Inhibition of dorsal raphe cell firing by (+)-MDMA, (−)-MDMA and PCA

Addition of (+)-MDMA (final concentration of 3-100 μM) to the superfusate inhibited dorsal raphe cell firing in a concentration-dependent manner (fig. 1A). Generally, the onset of the response occurred after 1-2 min of drug exposure (3 min total); peak effects occurred during the first few min following drug application. The time required for recovery to baseline firing rates ranged

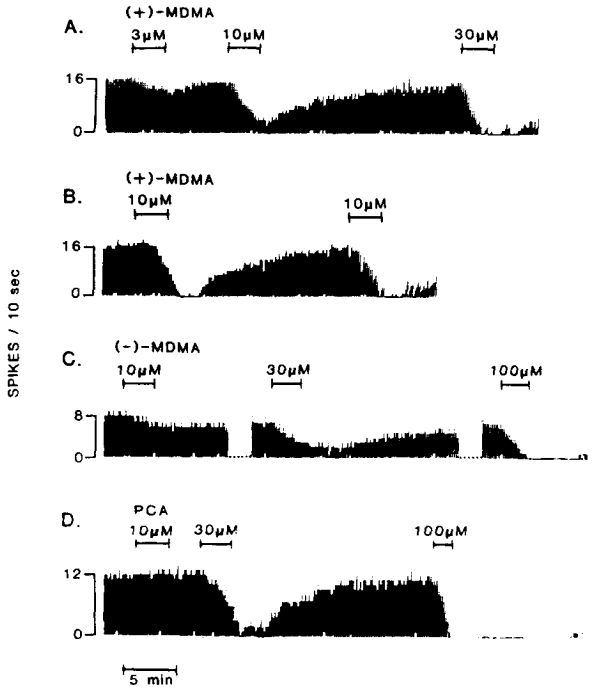

Fig. 1. Effects of (+)-MDMA, (−)-MDMA and PCA on the firing rate of four dorsal raphe neurons recorded in midbrain slices (A-D). Compounds were added to the superfusate to produce the concentrations indicated; the duration of drug exposure, indicated by the length of the bar, was fixed at 3 min. (A) (+)-MDMA reversibly inhibited dorsal raphe cell firing rate in a concentration-dependent manner. (B) Repeated applications of (+)-MDMA to the same cell produced similar inhibitory responses. (C) Like (+)-MDMA, (−)-MDMA inhibited unit activity as a function of drug concentration. Dotted lines indicate 10-20 min of elapsed time between tracings. (D) PCA also inhibited dorsal raphe cell firing within the same concentration range. Note that the 100 μM application lasted only 1.5 min.

10-50 min for the 3, 10 and 30 μM applications and was directly related to the drug concentration. For the 100 μM application, recovery was not complete in three of four cells examined for 2 h following drug exposure. Repeated applications of a fixed concentration of (+)-MDMA to the same cell produced similar responses (fig. 1B). Like (+)-MDMA, (−)-MDMA and PCA (10-100 μM) inhibited dorsal raphe neuronal discharges as a function of drug concentration (fig. 1C, D). Concentration-response curves for (+)-MDMA, (−)-MDMA and PCA constructed from these data appeared to be parallel (fig. 2), i.e. the hypothesis

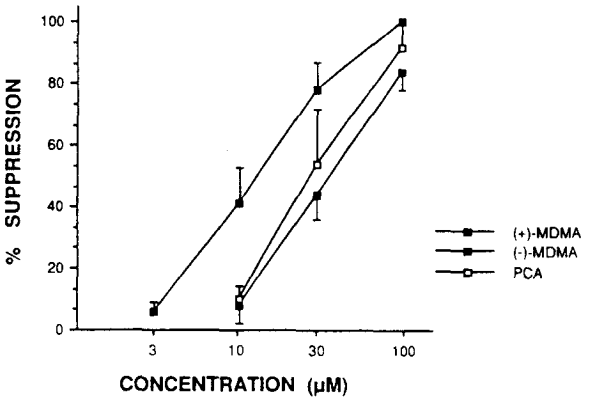

Fig. 2. Concentration-response curves for the inhibition of dorsal raphe cell firing by superfusions of (+)-MDMA, (−)-MDMA and PCA in midbrain slices (N = 5-8 cells).

of parallelism could not be rejected, $P > 0.1$ (Tallarida and Murray, 1982). Based upon IC_{50} values, (+)-MDMA was 2- to 3-fold more potent than (−)-MDMA in suppressing unit activity. There was a trend for (+)-MDMA to be more potent than PCA, but IC_{50} values for (−)-MDMA and PCA were indistinguishable. Potency differences among the three drugs could not be attributed to differences in the baseline firing rates of the cells sampled (table 1).

3.2. Blockade of MDMA- and PCA-induced inhibitory effects on dorsal raphe cell firing by fluoxetine

The selective 5-HT uptake inhibitor fluoxetine, when superfused at a concentration which had no effect on baseline firing rate (20 μM, 10 min),

TABLE 1

Potency differences among (+)-MDMA, (−)-MDMA and PCA in inhibiting the firing of dorsal raphe neurons recorded in midbrain slices. Values are means ± S.E.M. IC_{50} is the drug concentration required to produce a 50% suppression in unit activity.

	N	Firing rate (Hz)	IC_{50}
(+)-MDMA	8	1.6 ± 0.2	16 ± 4
(−)-MDMA	5	1.4 ± 0.2	40 ± 8 [a]
PCA	5	1.3 ± 0.3	35 ± 11 [b]

[a] $P < 0.01$, relative to (+)-MDMA; [b] $0.05 < P < 0.1$, relative to (+)-MDMA.

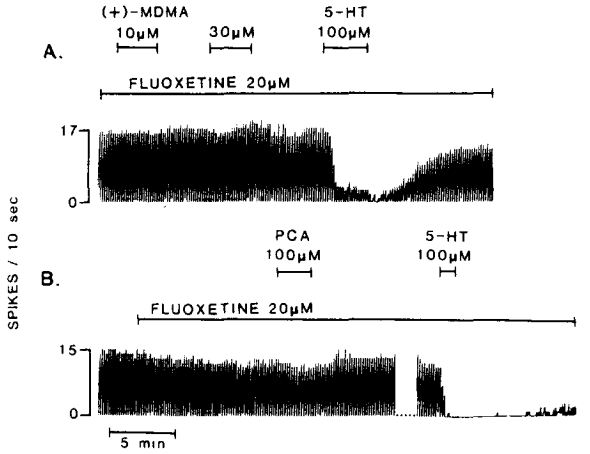

Fig. 3. Effect of fluoxetine pretreatment on (+)-MDMA- and PCA-induced inhibition of firing of two dorsal raphe neurons recorded in midbrain slices (A, B). Compared to untreated slices (fig. 1A, B, D), fluoxetine (20 μM) had no effect on baseline unit activity but blocked the inhibitory effects of (+)-MDMA (A) and PCA (B). The magnitude of the response to 5-HT (100 μM) generally was unaffected; however, the time to recovery was prolonged.

blocked the inhibitory effects of (+)-MDMA (30 μM) and PCA (100 μM) (figs. 3, 4). In these cells, the magnitude of the response to 5-HT (100 μM) was unchanged by fluoxetine; however, recovery to baseline firing generally was prolonged. Pretreatment with the selective norepinephrine uptake inhibitor desipramine (20 μM for a minimum of 10 min) had no effect on the responses of

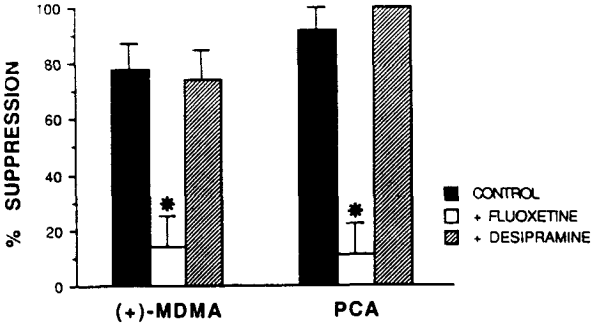

Fig. 4. Effect of fluoxetine and desipramine pretreatment on (+)-MDMA- and PCA-induced inhibition of dorsal raphe cell firing recorded in midbrain slices. Compared to untreated slices (CONTROL), 10 min of pretreatment with fluoxetine (20 μM) significantly blocked the inhibition of unit activity by (+)-MDMA (30 μM) and PCA (100 μM) whereas desipramine pretreatment (20 μM) had no effect (N = 4-8 cells, * $P < 0.01$).

dorsal raphe neurons to (+)-MDMA or PCA (fig. 4).

3.3. MDMA-induced release of 5-HT

(+)-MDMA caused the release of detectable quantities of 5-HT under the same conditions in which the compound caused the inhibition of dorsal raphe cell firing. Peak levels of 5-HT following

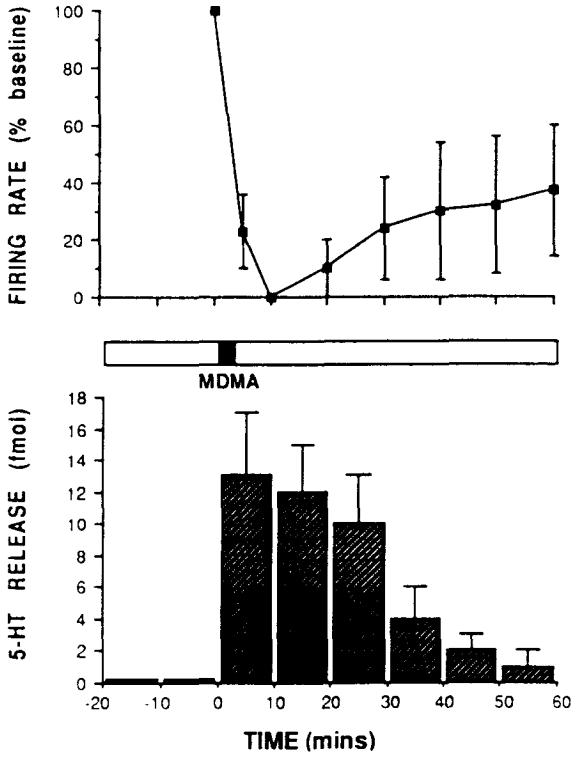

Fig. 5. (+)-MDMA-induced release of 5-HT from the dorsal raphe nucleus as measured by microdialysis probes resting on the brain slice surface. Lower panel: in the two collection periods prior to the application of (+)-MDMA, 5-HT release was below the limit of detection (2 fmol). In the collection period in which (+)-MDMA was applied (100 μM for 3 min as indicated by the bar), 5-HT release peaked to 13±4 fmol; thereafter, 5-HT release declined gradually but remained detectable 50 min following exposure to (+)-MDMA (N = 4 slices). Upper panel: in a parallel series of experiments, dorsal raphe cell firing, plotted along the same time base as 5-HT release in the lower panel, declined rapidly following the (+)-MDMA application (N = 4 cells). Firing rate gradually recovered thereafter; percentage recovery 100 min following (+)-MDMA exposure averaged 66±16 (not shown). This rapid decline followed then by slow recovery correlated with the changes in 5-HT release (lower panel).

a continuous 20 min exposure to (+)-MDMA (100 μM) averaged 19 ± 9 fmol (N = 3, 20 min collection period), uncorrected for differences in probe efficiency. Detectable levels of 5-HT persisted for three consecutive collection periods (60 min total). When MDMA superfusion (100 μM) was fixed at 3 min, as in the electrophysiological experiments, peak 5-HT levels occurred during the first or second 10 min collection period and averaged 13 ± 4 fmol (N = 4). Thereafter, 5-HT release gradually declined but remained detectable 50 min following MDMA. This prolonged time course correlated with the suppression of dorsal raphe cell firing (fig. 5): during the period of peak 5-HT release, dorsal raphe cell firing rate dropped to zero. As 5-HT release gradually declined to undetectable levels, unit activity gradually recovered to approximately 40% of baseline.

4. Discussion

There are several lines of evidence to suggest that the inhibition of dorsal raphe cell firing by (+)-MDMA, (−)-MDMA and PCA observed in the present study is indirect, mediated by the release of endogenous 5-HT. (1) Serotonergic dorsal raphe neurons can be inhibited via a 5-HT autoreceptor located in the somatodendritic region of the cell. Previous studies have shown that 5-HT released from recurrent axon collaterals and/or dendrite bundles as a result of cell firing binds to these receptors, thereby acting to maintain a slow and regular spontaneous firing rate (Aghajanian et al., 1987). Pharmacologic manipulations which increase synaptic availability of 5-HT (e.g. monoamine oxidase inhibitors, uptake inhibitors) consequently decrease dorsal raphe discharge rate (Aghajanian and Wang, 1978). (2) In the present study, the inhibition of dorsal raphe cell firing by MDMA was stereoselective with the (+) stereoisomer displaying significantly greater potency. Similarly, (+)-MDMA is more potent than (−)-MDMA at inducing 5-HT release in vitro (Schmidt et al., 1987). (3) The suppression of dorsal raphe neuronal discharges by (+)-MDMA and PCA was blocked by a selective 5-HT uptake inhibitor (fluoxetine) as is the acute and long term depletion of 5-HT (Schmidt et al., 1987; Fuller and Wong, 1987; Battaglia et al., 1988b). (4) Most compelling, however, was the evidence that the time course of MDMA-induced 5-HT release correlates closely with the time course of MDMA-induced inhibition of dorsal raphe unit activity. In experiments to be reported elsewhere, MDMA-induced 5-HT release in the brain slice, like MDMA-induced inhibition of dorsal raphe neuronal firing, was reduced significantly by fluoxetine pretreatment (Bradberry et al., in preparation). We propose therefore that MDMA elicits 5-HT release independently of cell firing, and that the released 5-HT binds to the inhibitory somatodendritic autoreceptor.

An alternative mechanism of action for (+)-MDMA, (−)-MDMA and PCA in inhibiting dorsal raphe unit activity might be the direct activation of the somatodendritic autoreceptor. However, it has been shown that this receptor is of the 5-HT_1 subtype (specifically, 5-HT_{1A}; Sprouse and Aghajanian, 1987) and (+)-MDMA and (−)-MDMA bind to 5-HT_1 receptors with very low affinity (Lyon et al., 1986; Battaglia et al., 1988a). Furthermore, the (+) stereoisomer of MDMA binds to 5-HT_1 sites with lower affinity than the (−) stereoisomer (Lyon et al., 1986), in contrast to the greater potency of (+)-MDMA in suppressing neuronal discharges (present study). A second line of evidence against a direct action for these compounds comes from the finding that fluoxetine, at a concentration which had no effect on baseline firing rate, blocked the inhibitory effects of (+)-MDMA and PCA. And finally, in preliminary studies with L-tryptophan, pretreatment with this 5-HT precursor (100 μM) increased the potency of (+)-MDMA and PCA to inhibit dorsal raphe cell firing (data not shown).

The neurochemical effects of MDMA most closely resemble those of PCA in that both compounds produce an acute and a long-term depletion of 5-HT after single or multiple doses and both compounds were more potent releasers of [^{3}H]5-HT than [^{3}H]dopamine (Schmidt et al., 1986; 1987; Stone et al., 1986). Dose-response curves for these compounds appear parallel, a finding consistent with the notion that MDMA and PCA induce acute 5-HT release by a similar

mechanism (Schmidt et al., 1987). Likewise, in the present study, dose-response curves for MDMA and PCA appear parallel, suggesting that in electrophysiological studies too these compounds are similar with regard to their mechanism of action.

There is some question as to the mechanism by which uptake inhibitors interfere with the release of monoamines by MDMA (Schmidt et al., 1987). Blockade of carrier-mediated transport of MDMA into the nerve terminal is an attractive hypothesis, yet [^{3}H]MDMA is not actively accumulated by rat striatal synaptosomes. Alternatively, MDMA may enter neurons by passive means where it causes the displacement and release of monoamines. Selective 5-HT uptake inhibitors such as fluoxetine and citalopram may not have any effect on the entrance of the releasing agent into the nerve terminal and instead prevent the carrier-mediated transport of monoamines out of the nerve terminal. As demonstrated in the present study, uptake inhibitors selective for norepinephrine should have no effect on entrance of the releasing agent or 5-HT release.

In previous reports, fluoxetine has been shown to suppress dorsal raphe cell firing at a bath concentration of 2-10 μM (Trulson and Frederickson, 1987; Rigdon and Wang, 1987). In the present experiments, however, the uptake inhibitor had no effect on baseline firing rate when superfused at 20 μM. Apparently, in our brain slice preparation, perhaps because of a more efficient perfusion system, there is not sufficient 5-HT in the extracellular fluid to accumulate in the presence of fluoxetine and affect discharge rate. When 5-HT was present in sufficient quantities, i.e. when the brain slice was exposed to brief high concentrations of 5-HT, fluoxetine at 20 μM delayed the recovery of the firing rate to baseline.

That MDMA-induced release of 5-HT returned to baseline before the suppression of dorsal raphe cell firing (fig. 5) suggests that release may continue at levels below the limit of detection which nevertheless can suppress neuronal discharge rate. This persistence of 5-HT release (detectable and undetectable) following MDMA application may be due to multiple actions of the compound. Beyond an increase in transmitter release, both stereoisomers of MDMA also directly inhibit uptake mechanisms and monoamine oxidase activity (Schmidt et al., 1986; 1987). Regardless of the mechanism(s) involved, it is likely that the prolonged time course of 5-HT release is the result of an action of the compound occurring within the serotonergic neuron because the rapid exchange afforded by an interface slice chamber (Haas et al., 1979) should effectively remove extracellular MDMA shortly after the superfusion has stopped (see Reid et al., 1988).

Immunocytochemical studies have shown that chronic treatment of rats with MDMA produces a profound loss of 5-HT axons throughout the forebrain. Fine 5-HT axon terminals are extremely vulnerable whereas raphe cell bodies are spared (O'Hearn et al., 1988). However, chronic treatment of monkeys has revealed that while MDMA produced no obvious cell loss, the drug induced striking cytopathological changes in the nerve cells of the dorsal raphe nucleus (Ricaurte et al., 1988). It is unlikely that the effects of MDMA observed on dorsal raphe cell firing in the present study were the result of neurotoxicity. The reversible nature of the MDMA-induced inhibition and the reproducibility of the drug effect argue against any toxicity. Nevertheless, MDMA-induced release of 5-HT as evidenced by decreases in dorsal raphe discharge rate and dialysis measurements of 5-HT efflux may be a necessary step in the pathologic process. It has been proposed that MDMA and related compounds destroy serotonergic neurons by releasing large amounts of 5-HT and inducing the endogenous formation of the serotonergic neurotoxin, 5,6-dihydroxytryptamine (Commins et al., 1987; see also Schmidt, 1987). The requirement for changes in spontaneous firing rate in initiating, sustaining or conversely slowing cell damage in the dorsal raphe nucleus or in postsynaptic areas is unknown.

In drug discrimination studies in rats, the interoceptive stimulus to the hallucinogenic phenylisopropylamine DOM (1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane) does not generalize to MDMA or to its N-monoethyl (MDE) or N-hydroxyl analogs (N-OH MDA) (Glennon et al., 1988). However, MDMA does result in stimulus generalization when (+)-amphetamine is used as the training drug. MDE and N-OH MDA do not

share this property of MDMA, yet all three compounds apparently share the same pharmacological profile in humans (Braun et al., 1980). Based upon these findings, it has been suggested that these agents produce a pharmacological effect that is neither amphetamine-like or DOM-like and may represent a new class of psychoactive compounds (Glennon et al., 1988). Presumably, neurotransmitter release from nerve terminals in serotonergic projection areas accounts for the behavioral effects of these unique agents. An analysis of MDMA action on postsynaptic neurons therefore is warranted.

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