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Pizotyline effectively attenuates the stimulus effects of *N*-Methyl-3,4-methylenedioxyamphetamine (MDMA)

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Abstract

MDMA (*N*-methyl-3,4-methylenedioxyamphetamine) produces a discriminative stimulus (DS) effect in animals, but attempts to completely block this action with selective neurotransmitter antagonists have not been very successful. Biochemically, MDMA can increase synaptic levels of serotonin, dopamine, and norepinephrine that, conceivably, might interact with multiple populations or subpopulations of neurotransmitter receptors. The present study attempted to antagonize the DS effects of MDMA using the nonselective agents clozapine, cyproheptadine, and pizotyline. An extensive and comparative radioligand binding profile was also obtained for the latter two agents. The purported antagonists were administered in combination with the training dose of MDMA to groups of Sprague–Dawley rats trained to discriminate 1.5 mg/kg of MDMA from saline vehicle in a standard two-lever operant paradigm using a VI-15s schedule of reinforcement. Clozapine was without effect at the doses evaluated, and cyproheptadine only partially attenuated MDMA-appropriate responding. In contrast, pizotyline (AD_{50}=2.5 mg/kg), in combination with the MDMA training dose, resulted in a dose related decrease in percent drug-appropriate responding to saline levels. In a separate group of animals trained to discriminate the structurally-related agent *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane (PMMA) from vehicle, pretreatment with pizotyline also resulted in a substantial decrease in drug-appropriate responding. The results with cyproheptadine and pizotyline in the binding assays confirmed that these agents display high affinity for multiple subpopulations of serotonergic, dopaminergic, adrenergic, histaminergic, and cholinergic receptors. The overall results of the present investigation indicate that pizotyline, which is clinically available in some countries, might be of clinical utility in the treatment of MDMA overdose.

Keywords: Designer drugs; Clozapine; Cyproheptadine; Pizotyline; Pizotifen; BC-105; Stimulus antagonism; Radioligand binding; MDMA; PMMA

The empathogen MDMA or *N*-methyl-3,4-methylenedioxyamphetamine ("Ecstasy"), currently a popular recreational drug and a potential psychotherapeutic agent, is thought to produce its behavioral effects primarily via release of serotonin and dopamine (Johnson et al., 1986; McKenna et al., 1991; Rudnick and Wall, 1992). Administered acutely or chronically to animals, MDMA can modulate synaptic levels of serotonin (5-HT), dopamine (DA), and norepinephrine (NE) (Mayerhofer et al., 2001; Rothman et al., 2001; Setola et al., 2003). Studies with human volunteers have demonstrated that the overall psychological effects of MDMA are dependent on carrier-mediated release of 5-HT whereas the stimulant-like euphoric actions are related, at least in part, to stimulation of dopamine receptors (Liechti and Vollenweider, 2000, 2001; Vollenweider et al., 1998). In particular, the mild perceptual effects induced by MDMA might involve stimulation of 5-HT_2 serotonin receptors (Liechti and Vollenweider, 2001; Liechti et al., 2000; Vollenweider et al., 1998). Also, MDMA users display increases in plasma concentrations of NE in the interval following drug use (Stuerenberg et al., 2002).

MDMA serves as a discriminative stimulus in animals and, although its stimulus mechanism has yet to be elucidated fully, there is evidence for the involvement of 5-HT and DA receptors. However, prior attempts to produce complete antagonism of the MDMA stimulus using relatively selective neurotransmitter receptor antagonists (e.g., serotonin 5-HT_{1A} and 5-HT_2 antagonists, dopamine D_1 and D_2 antagonists, β_2-adrenoceptor antagonists) have not met with much success; these antagonists, when administered in combination with MDMA, typically lead

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to ≤50% attenuation of the MDMA response (Glennon and Young, 2000; Glennon et al., 1992; Schechter, 1989). Rather than continue examining the effects of "selective" neurotransmitter receptor antagonists to attenuate the stimulus effects of MDMA, the present study employed the opposite approach. That is, several non-selective (i.e., "broad spectrum") receptor antagonists including clozapine, cyproheptadine and pizotyline (pizotifen, BC-105) were chosen for investigation. By virtue of releasing several neurotransmitters, MDMA could indirectly activate multiple neurotransmitter receptor subpopulations, and it was hypothesized that a non-selective antagonist might be more effective than a selective antagonist in attenuating the MDMA stimulus. Clozapine, cyproheptadine and pizotyline are known to bind at multiple receptor populations, including several 5-HT and DA receptor subpopulations (Bymaster et al., 1996; Leysen et al., 1981; Millan et al., 2000; Schmidt et al., 2001) – although an extensive binding profile has not yet been published for cyproheptadine or pizotyline. The present study presents a more complete binding profile for the latter two agents and examines the effects of these three nonselective agents in rats trained to discriminate 1.5 mg/kg of MDMA from saline vehicle. Lastly, a comparative analysis evaluated pizotyline in rats trained to discriminate the stimulus effects of the MDMA-related agent PMMA [*N*-methyl-1-(4-methoxypheny)-2-aminopropane] from saline. Structurally, PMMA can be viewed as an abridged analog of MDMA that lacks the stimulus and stimulant character of amphetamine but that, nonetheless, cross-substitutes with MDMA regardless of which of the two agents is used as training drug (Glennon and Higgs, 1992; Glennon et al., 1997).

1. Materials and methods

1.1. Drug discrimination studies

Fourteen male Sprague–Dawley rats (Charles River Laboratories), weighing 250–300 g at the beginning of the study, were trained to discriminate (15-min presession injection interval) either 1.5 mg/kg of MDMA ($n=7$) or 1.25 mg/kg of PMMA ($n=7$) from saline vehicle (sterile 0.9% saline) under a variable interval 15-s schedule of reward (i.e., sweetened condensed milk) using standard two-lever Coulbourn Instruments operant equipment as previously described (Dukat et al., 2002; Glennon et al., 1997; Glennon and Young, 2000). Animal studies were conducted under an approved Institutional Animal Care and Use Committee protocol.

In brief, animals were food-restricted to maintain body weights of approximately 80% that of their free-feeding weight, but were allowed access to water ad lib in their individual home cages. Daily training sessions were conducted with the training dose of the training drugs or saline. For approximately half the animals, the right lever was designated as the drug-appropriate lever, whereas the situation was reversed for the remainder of the animals. Learning was assessed every fifth day during an initial 2.5-min non-reinforced (extinction) session followed by a 12.5-min training session. Data collected during the extinction session included response rate (i.e., responses per minute) and number of responses on the drug-appropriate lever (expressed as a percent of total responses). Animals were not used in the subsequent stimulus generalization or antagonism studies until they consistently made ≥80% of their responses on the drug-appropriate lever after administration of training drug and ≤20% of their responses on the same drug-appropriate lever after administration of saline (Young and Glennon, 1986). During the testing (i.e., stimulus generalization and antagonism) phase of the study, maintenance of the training-drug/saline discrimination was insured by continuation of the training sessions on a daily basis (except on a generalization or antagonism test day). On one of the two days before a generalization or antagonism test, approximately half the animals would receive the training dose of training drug and the remainder would receive saline; after a 2.5-min extinction session, training was continued for 12.5 min. Animals not meeting the original training criteria during the extinction session were excluded from the subsequent generalization or antagonism test session. During the investigations of stimulus generalization, test sessions were interposed among the training sessions. The animals were allowed 2.5 min to respond under non-reinforcement conditions. An odd number of training sessions (usually 5) separated any two generalization test sessions. Doses of test drugs were administered in a random order, using a 15-min presession injection interval (unless otherwise noted), to the groups of rats. Stimulus generalization was considered to have occurred when the animals, after a given dose of drug, made ≥80% of their responses (group mean) on the training drug-appropriate lever. In the antagonism studies, antagonism was considered to have occurred when the animals made ≤20% (group mean) of their responses on the drug-appropriate lever following a combination of test drug and the training dose of training drug. Animals making fewer than 5 total responses during the 2.5-min extinction session were considered as being behaviorally disrupted. Percent drug-appropriate responding and response rate data refer only to animals making ≥5 responses during the extinction session (Young and Glennon, 1986). If >50% of the animals were disrupted following administration of a given drug dose, data were not plotted. Where applicable, an ED_{50} or AD_{50} dose was calculated by the method of Finney (1952). These doses represent the drug dose where animals would be expected to make 50% of their responses on the drug-appropriate lever.

1.2. Binding profile

Cyproheptadine HCl and pizotyline maleate and were examined in a number of different radioligand binding assays by the NIMH Psychoactive Drug Screening Program; PMMA was examined at 5-HT_{2A} and 5-HT_{2C} serotonin receptors. The agents were initially screened several times at a concentration of 10,000 nM; if an agent produced >50% inhibition, a Ki value was determined in quadruplicate. Where an agent produced <50% inhibition of binding, a Ki of >10,000 nM is reported. Assay details and standard binding protocols can be found at:http://pdsp.cwru.edu/pdspw/binding.php.

1.3. Drugs

Racemic *N*-methyl-3,4-methylenedioxyamphetamine HCl [*N*-methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropne HCl; MDMA] was obtained as a gift from NIDA, whereas racemic *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane HCl was synthesized in our laboratories. Clozapine free base was purchased from Sigma/Aldrich, and cyproheptadine HCl (Merck, Sharp and Dohme Research Labs) and pizotyline maleate (Pizotifen, BC-105) (Sandoz Pharmaceuticals) were obtained as gifts.

MDMA and PMMA were administered (i.p.) 15 min prior to testing, and clozapine, cyproheptadine and pizotyline were examined 45 min prior to testing. One drop of Tween 80 was used to aid the suspension of clozapine. Following drug administration, animals were returned to their individual home cages prior to testing. Doses refer to the weight of base (clozapine) or the salts. Solutions in sterile 0.9% saline were freshly prepared daily, and injected in a constant volume of 1 ml/kg.

2. Results

Groups of rats trained to discriminate either 1.5 mg/kg of MDMA (Figs. 1 and 2) or 1.25 mg/kg of PMMA (Fig. 3) from saline vehicle were used in the study. The administration of training-drug doses lower than the training dose, to the respective group, resulted in decreased percent drug-appropriate responding (Figs. 2 and 3). Response rates were not markedly different following the administration of the different doses of training drug or saline (Figs. 2 and 3). Dose–response curves for MDMA and PMMA are consistent with prior results for these agents (Dukat et al., 2002; Glennon and Young, 2000; Glennon et al., 1997).

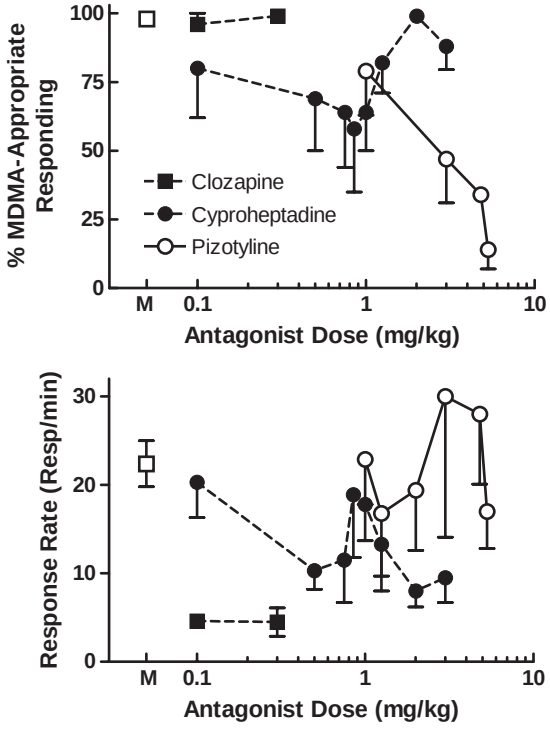

Fig. 1. Results of antagonism tests in rats ($n=5$ to 7/dose) trained to discriminate MDMA (1.5 mg/kg) from saline vehicle (upper panel). Mean (±SEM) percent drug-appropriate responding following administration of MDMA in combination with doses of clozapine, cyproheptadine, and pizotyline; M=effect of MDMA (1.25 mg/kg) in the absence of the antagonist. The animals' response rates are shown in the lower panel. Animals making <5 total responses during the 2.5-min extinction session were considered as behaviorally disrupted; percent drug-appropriate responding and response rate data refer only to animals making ≥5 responses during the extinction session.

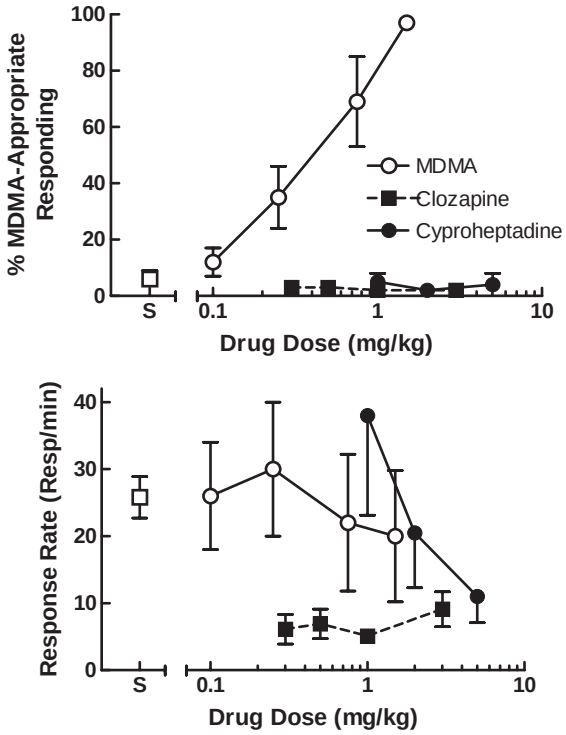

Fig. 2. Results of generalization tests in rats ($n=5$ to 7/dose) trained to discriminate MDMA (1.5 mg/kg) from saline vehicle (upper panel). Mean (± SEM) percent drug-appropriate responding following administration of various doses of MDMA clozapine, and cyproheptadine; S=effect of saline (1 ml/kg). The animals' response rates are shown in the lower panel. Animals making <5 total responses during the 2.5-min extinction session were considered as behaviorally disrupted; percent drug-appropriate responding and response rate data refer only to animals making ≥5 responses during the extinction session.

Clozapine was examined in five animals trained to discriminate MDMA from saline vehicle (Fig. 1). When administered at 0.1 mg/kg in combination with the training dose of MDMA, the animals made 96% of their responses on the MDMA-appropriate lever and, following a dose of 0.3 mg/kg, 3/5 animals responded and made 100% of their responses on the MDMA-appropriate lever. Administered 0.5 mg/kg of clozapine in combination with the training dose of MDMA, none of the animals responded (data not shown). Administered alone (Fig. 2), clozapine (doses of 0.3, 0.5, 1, and 3 mg/kg) failed to produce >3% drug-appropriate responding; although all animals responded at each dose, their response rates were substantially depressed (Fig. 2).

A total of eight doses of cyproheptadine (0.1 to 3 mg/kg) were examined in combination with MDMA in groups of five to seven animals per dose (Fig. 1). Percent MDMA-appropriate responding decreased to 58% (at 0.85 mg/kg of cyprohepta-

dine); administration of higher cyproheptadine doses in combination with the training dose of MDMA produced an increase in MDMA-appropriate responding. At 2 mg/kg, four of five animals made ≥5 total responses during the 2.5-min extinction session, whereas at 3 mg/kg, only four of seven animals made ≥5 total responses. Administered alone in tests of stimulus generalization, cyproheptadine failed to engender >5% MDMA-appropriate responding at doses of up to 5 mg/kg (Fig. 2). Administration of 7.5 mg/kg of cyproheptadine disrupted the animals' lever-pressing behavior and none of five animals made any responses (data not shown).

Pizotyline (AD_{50} dose=2.5 mg/kg; 95% CL=1.3–4.8 mg/kg) was the only one of the three agents that elicited <20% MDMA-appropriate responding when administered in combination with the training dose of MDMA; 5.25 mg/kg of pizotyline together with the training dose of MDMA produced 14% MDMA-appropriate responding (Fig. 1). Administered alone 45 min prior to testing, pizotyline (5.25 mg/kg) produced 5 (±2)% MDMA-appropriate responding; the animals' response rate was 18.9 (±2.9) resp/min at this dose (data not shown).

The administration of 5.0 mg/kg of pizotyline in combination with the training dose of PMMA decreased the animals' PMMA-appropriate responding to 32% (Fig. 3); at doses of 6 and 7 mg/kg, 6/7 and 5/7 animals responded but there was no

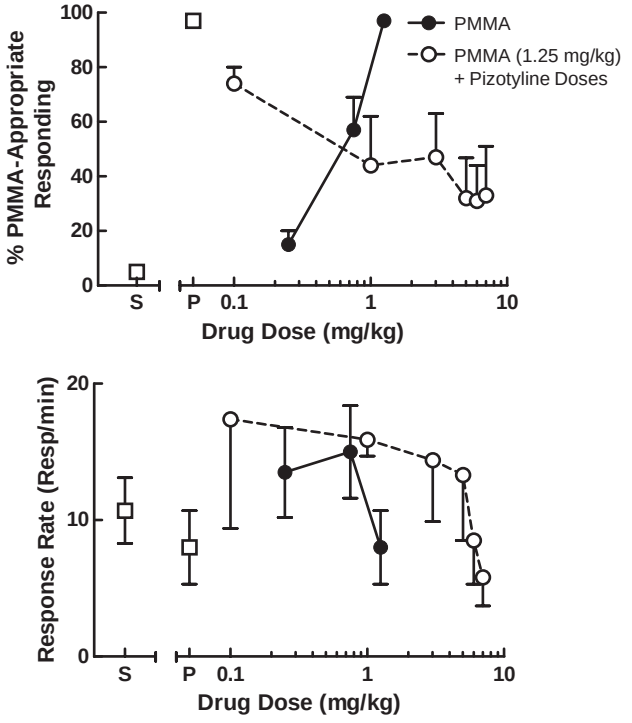

Fig. 3. Results of generalization and antagonism tests in rats (n=6 to 7/dose) trained to discriminate PMMA (1.25 mg/kg) from saline vehicle (upper panel). Mean (±SEM) percent drug-appropriate responding following administration of PMMA doses, or PMMA (1.25 mg/kg) in combination with various doses of pizotyline. S=effect of saline (1 ml/kg) and P=effect of PMMA (1.25 mg/kg) in the absence of antagonists. The animals' response rates are shown in the lower panel. Animals making <5 total responses during the 2.5-min extinction session were considered as behaviorally disrupted; percent drug-appropriate responding and response rate data refer only to animals making ≥5 responses during the extinction session.

Table 1
Binding profile for cyproheptadine and pizotyline[a]

Receptor	Ki, nM (±SEM)			
	Cyproheptadine	HCl	Pizotyline	Maleate
5-HT_{1A} Serotonin	50	(10)	270	(45)
5-HT_{1B} Serotonin	1600	(560)	1415	(310)
5-HT_{1D} Serotonin	670	(200)	770	(300)
5-HT_{1E} Serotonin	1500	(340)	820	(135)
5-HT_{2A} Serotonin	1.9	(0.2)	2.0	(0.3)
2-HT_{2B} Serotonin	2.6	(0.3)	2.3	(0.2)
5-HT_{2C} Serotonin	18	(2)	8.4	(2.1)
5-HT_3 Serotonin	235	(45)	95	(30)
5-HT_{5A} Serotonin	57	(8)	110	(16)
5-HT_6 Serotonin	96	(40)	74	(40)
5-HT_7 Serotonin	30	(6)	17	(5)
D_1 Dopamine	10	(6)	3.5	(2.0)
D_2 Dopamine	74	(11)	87	(22)
D_4 Dopamine	120	(18)	64	(13)
D_5 Dopamine	60	(23)	50	(12)
SERT	>10,000		>10,000	
NET	2550	(300)	710	(180)
DAT	4100	(1150)	>10,000	
H_1 Histamine	2.3	(0.8)	1.9	(0.2)
H_2 Histamine	4.8	(1.0)	1.4	(0.6)
α_{1A} -Adrenoceptor	45	(2)	65	(4)
α_{1B}-Adrenoceptor	>10,000		>10,000	
α_{2A}-Adrenoceptor	330	(40)	660	(140)
α_{2B}-Adrenoceptor	220	(8)	225	(15)
α_{2C}-Adrenoceptor	160	(20)	390	(20)
β_1-Adrenoceptor	>10,000		>10,000	
β_2-Adrenoceptor	>10,000		>10,000	
m_1 Cholinergic	19	(1)	67	(1)
m_2 Cholinergic	18	(2)	34	(3)
m_3 Cholinergic	12	(1)	29	(1)
m_4 Cholinergic	19	(1)	130	(26)
m_5 Cholinergic	4.5	(0.3)	6.8	(0.4)
I_1 Imidazoline	204	(12)	121	(10)
σ_1 Sigma	>10,000		>10,000	
σ_2 Sigma	750	(110)	6450	(820)

[a] Radioligand binding data were obtained from the NIMH Psychoactive Drug Screening Program. In addition to results shown above, neither agent displayed affinity (i.e., Ki>10,000 nM) for $GABA_A$ receptors, benzodiazepine receptors, μ-, κ-, and δ-opioid receptors, α2β2, α2β4, α3β2, α3β4, α4β2, and α4β4 nicotinic cholinergic receptors, or CB_1 and CB_2 canabinoid receptors.

further decrease in percent PMMA-appropriate responding. Administered alone to six animals, 7 mg/kg of pizotyline elicited 9 (±3)% PMMA-appropriate responding; the animals' response rate was similar to their response rate following 1.25 mg/kg of PMMA or saline (data not shown).

A detailed binding profile was obtained for cyproheptadine and pizotyline (Table 1). In general, both agents displayed high affinity for multiple populations of serotonin, dopamine, histamine, adrenergic, and muscarinic receptors. Results for the two agents were nearly identical with neither showing more than a few-fold difference in affinity from the other. PMMA was found to lack affinity (Ki>10,000 nM) for 5-HT_{2A} and 5-HT_{2C} serotonin receptors.

3. Discussion

Three relatively non-selective neurotransmitter receptor antagonists—clozapine, cyproheptadine, and pizotyline—were

administered to animals trained to discriminate MDMA (1.5 mg/kg) from saline vehicle. These agents were given alone or in combination with the training dose of MDMA to determine if they would generalize to, or antagonize, the MDMA-induced stimulus, respectively. Only pizotyline (AD_{50}=2.5 mg/kg) effectively reduced MDMA-appropriate responding to saline levels (Fig. 1). Cyproheptadine partially (to 58%) attenuated the MDMA response whereas clozapine was without antagonist action (Fig. 1). It is unlikely that the failure of cyproheptadine or clozapine to fully attenuate MDMA-appropriate responding is related to any MDMA-like action because neither agent produced >5% MDMA-appropriate responding when examined in tests of stimulus generalization.

MDMA is an arylalkylamine with recognized abuse liability. Chemically related arylalkylamines can be classified as producing one, or more, of three distinct prototypical stimulus effects in animals: i) a stimulant-like effect, as evidenced by substitution studies employing rats trained to discriminate the stimulant arylalkylamine (+)amphetamine from vehicle, ii) a hallucinogen-like effect, as evidenced by substitution studies employing the arylalkylamine hallucinogen 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (DOM) from vehicle, and iii) a third effect that is typified by that produced by the arylalkylamine designer drug *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane (PMMA) (Glennon, 1999). The latter effect, though not yet fully characterized, bears similarities to MDMA but, unlike MDMA, seems to lack a stimulant component of action. That is, MDMA substitutes both for a (+)amphetamine stimulus and a PMMA stimulus, and both (+)amphetamine and PMMA substitute for an MDMA stimulus (Glennon et al., 1997). However, neither (+)amphetamine nor PMMA substituted for one another regardless of which of the two was used as training drug (Glennon et al., 1997). Hence, MDMA might be best characterized as an agent that produces stimulus effects with some commonalities to those produced by amphetamine and PMMA.

Neither an amphetamine (Winter, 1978) nor a (+)amphetamine stimulus (Minnema and Rosecrans, 1982, 1984; Young and Glennon, 1986) is antagonized by pizotyline. In contrast, the stimulus effects of hallucinogens are effectively antagonized by pizotyline as demonstrated in studies employing any one of several arylalkylamine hallucinogens as training drugs including, for example, mescaline (Winter, 1978), 5-methyoxy-*N*,*N*-dimethyltryptamine (Glennon et al., 1979; Young et al., 1983), (+)lysergic acid diethylamide (LSD) (Colpaert et al., 1982; Holohean et al., 1982; Minnema and Rosecrans, 1984; Rosecrans and Glennon, 1979), and DOM (Young et al., 1980). Because pizotyline completely antagonized the stimulus effects of hallucinogens, but not those of amphetamine, it was of interest to determine the effect of pizotyline on the third type of prototypical arylalkylamine stimulus, namely, the PMMA stimulus. Fig. 3 shows that administration of pizotyline in combination with PMMA, to PMMA-trained animals, resulted in decreased PMMA-appropriate responding. Although drug-appropriate responding did not achieve saline-like levels (i.e., ≤20% PMMA-appropriate responding), substantial (i.e., to about 30% PMMA-appropriate responding) attenuation of the PMMA response was seen. Given the receptor populations to which pizotyline (Table 1) and MDMA (Khorana et al., 2004) bind, there is no ready explanation for the observed results. One possibility that can be proffered involves association with 5-HT_2 serotonin receptors. Pizotyline is an effective antagonist of agents that produce their stimulus effects through a 5-HT_2 mechanism (e.g., DOM). Moreover, it has been demonstrated that 5-HT_2 antagonists partially attenuate the stimulus effects of MDMA (Glennon et al., 1992; Schechter, 1989), that certain actions of MDMA in humans involve activation of 5-HT_2 receptors (Liechti and Vollenweider, 2001; Liechti et al., 2000), and that MDMA binds, although with modest affinity, at 5-HT_2 receptors (Lyon et al., 1986). The inability of pizotyline to fully antagonize the PMMA stimulus might be related to a lack of affinity of PMMA (Ki>10,000 nM; see Results) for this receptor population.

The ability of pizotyline, but not cyproheptadine, to attenuate the MDMA-induced stimulus was initially thought to represent a potential clue to what neurotransmitter mechanism(s) might be most effectual for this action. Perhaps binding differences between these two agents might identify receptor populations to which pizotyline binds but that cyproheptadine does not. Binding profiles were obtained for both agents (Table 1) and it would seem that they share a nearly identical affinity profile. Given the isosteric structural relationship of these two agents (Fig. 4), this might have been expected. One explanation for the observed difference in antagonist ability might relate to different functional activity (i.e., efficacy) of pizotyline and cyproheptadine at a specific receptor population(s), and another possibility is that a receptor population not examined here might play a role in the action of pizotyline.

Although the present investigation does little to identify mechanisms underlying the MDMA stimulus, an indirect finding is demonstration of mechanistic differences in the stimulus actions of arylalkylamines such as amphetamine, MDMA and PMMA. Even though there is some similarity between the stimulus actions of MDMA and amphetamine, as well as between MDMA and PMMA, the stimulus effects of amphetamine (Arnt, 1996; Nielsen and Jepsen, 1985) but not MDMA (Fig. 1) are antagonized by clozapine, whereas

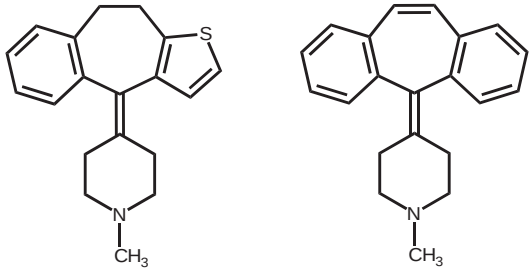

Fig. 4. Chemical structures of pizotyline and cyproheptadine showing their close structural similarity.

those of MDMA (Fig. 1) but not amphetamine (vide supra) are antagonized by pizotyline. Furthermore, whereas pizotyline seems to completely antagonize the MDMA stimulus (Fig. 1), it only partially antagonized the PMMA stimulus (Fig. 3).

An alternative explanation for the present findings with the MDMA/pizotyline combination is that pizotyline (or the combination) produces stimulus effects (i.e., cueing properties) different than those occasioned by MDMA alone; this could cause the animals receiving the combination to respond on the non-MDMA-appropriate lever. This is a possible explanation for any drug discrimination study involving apparent antagonism of a stimulus cue. At a dose slightly higher than the highest dose used in this study, pizotyline (at 6 mg/kg) serves as a discriminative stimulus and produces effects that seem to be antihistaminergic in nature (Minnema et al., 1984). In addition, Yamamoto et al. (1991) have reported that pizotyline substitutes for a 5-hydroxytrptophan (5-HTP) stimulus in pigeons when administered via the intramuscular route suggesting possible serotonergic effects. But this latter effect might be related to species and/or route of administration because pizotyline (2 to 8 mg/kg, i.p.) has been shown to antagonize the effects of 5-HTP in a dose-dependent and surmountable fashion following administration to rats (Cunningham et al., 1985; Friedman et al., 1983), and that given by itself pizotyline (3 mg/kg, i.p.) produces saline-appropriate responding (Friedman et al., 1983). Furthermore, when LSD — an agent that substitutes for a 5-HTP stimulus (Cunningham et al., 1985) — was used as training drug in rats, pizotyline behaved as an antagonist (at doses of about 1 mg/kg and greater) but failed to substitute for the LSD stimulus at doses of up to 40 mg/kg (Colpaert et al., 1982). Nevertheless, a possible masking effect by pizotyline (or the combination) on MDMA-appropriate responding cannot be discounted on the basis of the present results.

The most notable finding of this investigation is that pizotyline can reduce percent drug-appropriate responding to saline levels when administered in combination with MDMA to animals trained to discriminate 1.5 mg/kg of MDMA from saline vehicle. Pizotyline was initially developed in the 1960s as a therapeutic agent for the treatment of migraine (Sicuteri et al., 1967) and depression associated with schizophrenia (Krumholz et al., 1968). Early studies showed that pizotyline behaved, at least in part, as a serotonin antagonist and, following the discovery of 5-HT_1 and 5-HT_2 receptors, pizotyline was shown to bind selectively at the latter — as well as at histamine receptors (Leysen et al., 1981). Table 1 shows that pizotyline is even more non-selective than originally appreciated and binds at a multiplicity of neurotransmitter receptors with nanomolar affinity. Nevertheless, it is possible that pizotyline, which is clinically available in some countries for the treatment of migraine and depression, might prove to be of clinical utility in the treatment of MDMA overdoses.

Acknowledgments

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