Available online at www.sciencedirect.com
ScienceDirect
Pharmacology, Biochemistry and Behavior 86 (2007) 477–484
PHARMACOLOGY BIOCHEMISTRY AND BEHAVIOR
www.elsevier.com/locate/pharmbiochembeh

N-Methyl-1-(4-methoxyphenyl)-2-aminopropane (PMMA) and *N*-Methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropane (MDMA) produce non-identical discriminative stimuli in rats

Richard A. Glennon *, Richard Young, Małgorzata Dukat, Jean Chang-Fong, Mohamed El-Zahabi

Department of Medicinal, Chemistry School of Pharmacy Medical College of Virginia, Virginia Commonwealth University Richmond, VA 23298-0540, United States

Received 6 October 2006; received in revised form 3 January 2007; accepted 10 January 2007
Available online 18 January 2007

Abstract

N-Methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropane (methylenedioxymethamphetamine, MDMA, *Ecstasy*) and its structurally abbreviated congener *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane (*para*-methoxymethamphetamine, PMMA) are chemically related designer drugs, and PMMA is sometimes sold on the clandestine market as a substitute for MDMA. Prior drug discrimination studies have found that MDMA and PMMA substitute for one another suggesting that they produce similar discriminative stimulus effects in rats. However, there also are some indications that the two agents produce distinct stimulus effects. In this study, further comparisons were made between the stimulus effects of these two agents. Sprague–Dawley rats were trained to discriminate either 1.25 mg/kg of PMMA or 1.5 mg/kg of MDMA from saline vehicle in a two-lever operant paradigm. A structure–activity comparison revealed that MDMA and PMMA behave similarly upon homologation of their terminal amine substituents. In contrast, the PMMA stimulus, unlike an MDMA stimulus, failed to generalize completely to the psychostimulant cocaine, 8-hydroxy-2-(*N*,*N*-di-*n*-propylamino)tetralin (8-OH DPAT), and *R*(−)-1-(3-methoxyphenyl)-2-aminopropane [*R*(−)MMA]. In an additional group of animals, a (+)amphetamine stimulus partially generalized to *R*(−)MMA. Taken together, the results argue and re-emphasize the conclusion that the stimulus effects produced by MDMA and PMMA are similar, but non-identical, and that PMMA is the less "stimulant-like" of the two.
© 2007 Elsevier Inc. All rights reserved.

Keywords: MDMA; PMMA; Cocaine; 8-OH DPAT; *N*-Ethyl PMA (PMEA); *N*-Propyl PMA (PMPA); *R*(−)MMA; *S*(+)MMA

1. Introduction

N-Methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropane (also known as methylenedioxymethamphetamine; MDMA, *Ecstasy*) is a popular recreational drug (Baylen and Rosenberg, 2006; Green et al., 2003). Its structurally simpler congener *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane (*para*-methoxymethamphetamine, PMMA) (see Fig. 1 for structural comparison) is sometimes sold as a substitute for MDMA on the clandestine market (Dal Cason, 2001). Comparative studies in humans have not been reported, but in drug discrimination studies using rats, MDMA and PMMA produce what appear to be similar discriminative stimulus effects. That is, stimulus generalization occurs between MDMA and PMMA regardless of which of the two is used as training drug (Glennon and Higgs, 1992; Glennon et al., 1997). Moreover, for both agents, the *S*(+)-optical isomer is the more potent, the α-methyl group can be homologated to an α-ethyl group with retention of action, and the preferred side chain conformation of both agents appears to be mimicked by an aminotetralin structure (Glennon et al., 1997; Young et al., 1999). Despite their structural and stimulus similarities, the two agents do not seem to produce identical pharmacological effects. One of the salient differences between the two agents is that MDMA seems to possess greater central stimulant character than PMMA. For example, MDMA produces a hyperlocomotor effect in rodents (e.g.; Glennon et al., 1988a) whereas this effect is essentially absent following administration of PMMA (Glennon et al., 1988b; Bustamante et al., 2004). Furthermore, it has been demonstrated in rats that a

* Corresponding author. Tel.: +1 804 828 8487; fax: +1 804 828 7404.
E-mail address: glennon@vcu.edu (R.A. Glennon).

0091-3057/$ - see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.pbb.2007.01.007

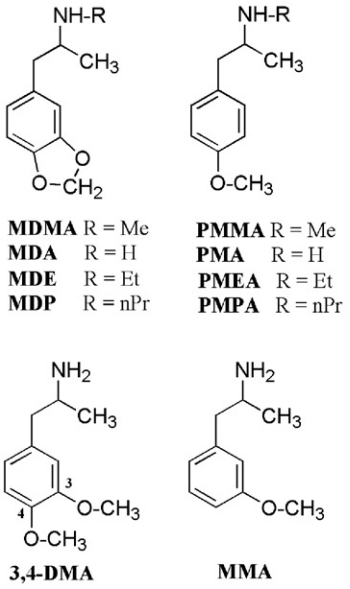

Fig. 1. Chemical structures showing the structural relationships of the agents described in this investigation.

considerable degree of stimulus generalization occurs between MDMA and the psychostimulant (+)amphetamine regardless of which is used as training drug (Glennon, 1989; Nichols and Oberlender, 1989; for further discussion, see Bondareva et al., 2005); this is not the case with PMMA and (+)amphetamine (Glennon and Misenheimer, 1989; Glennon et al., 1997).

Given the purported similarities, yet possible differences, between the discriminative stimulus actions of MDMA and PMMA, the purpose of the present investigation was to further compare the stimulus actions of these agents in rats. The tactic employed for this investigation was as follows. Of the two agents, MDMA is the better investigated as a discriminative stimulus in rats. Based on available information, several agents were selected for specific comparison.

First, MDMA stimulus generalization occurs to the central stimulant cocaine (Khorana et al., 2004); thus, cocaine was selected for examination in PMMA-trained animals. Second, it has been found that the 5-$HT_{1A/7}$ receptor agent 8-hydroxy-2-(*N*,*N*-di-*n*-propylamino)tetralin (8-OH DPAT) unexpectedly substitutes in MDMA-trained rats (Glennon and Young, 2000); hence, 8-OH DPAT was selected for examination (i.e., both in tests of substitution and modulation) in PMMA-trained animals.

Because homologation of the *N*-methyl group of MDMA to its *N*-ethyl analog (i.e., MDE – also known as MDEA – see Fig. 1) results in an agent that substitutes for an MDMA stimulus (Glennon, 1990), a third approach was to compare the effect of this structural modification using the *N*-ethyl counterpart of PMMA (i.e., PMEA). PMEA was synthesized and examined in PMMA-trained animals.

A final approach examined a ring-opened analog of MDMA (i.e., MMA). Inspection of the chemical structures of MDMA and PMMA (Fig. 1) reveals a close structural relationship, and that both agents can be related back to, or envisioned as having been structurally derived from, 3,4-DMA (Fig. 1). In concert with this concept is that both an MDMA stimulus and a PMMA stimulus generalize to *S*(+)- and *R*(−)-3,4-DMA (Rangisetty et al., 2001). MDA (Fig. 1) represents an "intermediate step" in the structural progression from 3,4-DMA to MDMA, as does PMA (i.e., the 3-*des*-methoxy analog of 3,4-DMA) for PMMA. An MDMA stimulus generalizes to (±)-, *R*(−)- and *S*(+)MDA (Oberlender and Nichols, 1988; Nichols and Oberlender, 1989), as does a PMMA stimulus generalize to *R*(−)PMA (Dukat et al., 2002). Furthermore, the PMMA stimulus also generalizes to *R*(−)- and *S*(+)MDA (Glennon and Young, 2002). Moreover, it seems that the presence of the 4-position oxygen substituent might account for, or at least contribute to, the stimulus properties (similarities?) of these agents because it is a structural feature they all have in common. The corresponding 4-*des*-methoxy analog of 3,4-DMA (i.e., MMA; Fig. 1) has not yet been examined. MMA might be viewed as an alternative intermediate in the structural progression of 3,4-DMA to MDMA-like agents, but not in the progression to PMMA. That is, unlike PMA (which lacks the 3-position substituent of 3,4-DMA), MMA lacks the 4-position substituent. Thus, MMA might substitute in MDMA-trained animals, but not in PMMA-trained animals. To test this hypothesis, the two optical isomers of MMA were synthesized and their stimulus effects examined in MDMA- and PMMA-trained rats.

Overall, then, the stimulus actions of PMMA were compared with those of MDMA using four distinct strategies: evaluations based on the stimulus effects induced by cocaine, 8-OH DPAT, and two types of structural modifications of the PMMA/MDMA molecules.

Finally, (+)amphetamine-trained rats have exhibited a significant degree of (+)amphetamine-like responding following the administration of racemic MMA (Glennon et al., 1985; Huang and Ho, 1974). As such, the enantiomers of MMA also were evaluated in rats trained to discriminate (+)amphetamine from saline vehicle to determine if either MMA optical isomer can produce (+)amphetamine-like discriminative stimulus effects.

2. Materials and methods

2.1. Drug discrimination

Eighteen male Sprague–Dawley rats (Charles River Laboratories), weighing 250–300 g at the beginning of the study, were employed in this investigation. The animals were trained to discriminate (15-min pre-session injection interval) doses of either PMMA (1.25 mg/kg; $n=8$), MDMA (1.5 mg/kg; $n=5$), or (+)amphetamine (1.0 mg/kg; $n=5$) from saline vehicle (sterile 0.9% saline) under a variable interval 15-s schedule of reinforcement (for sweetened milk reward) using standard two-lever Coulbourn Instruments operant equipment. We have previously employed each of these agents (and training doses) as training drugs, and the training procedures have been reported (Glennon and Young, 2000; Glennon et al., 1997; Young et al., 2006). Animal studies were conducted under an approved Institutional Animal Care and Use Committee protocol.

In brief, animals were food-restricted to maintain body weights at approximately 80% of free-feeding weight but were allowed access to water *ad lib* in their individual home cages. Daily training sessions (15 min) 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 number of responses on the drug-appropriate lever (expressed as a percent of total responses) and response rate (i.e., responses per minute). The animals were not used in the subsequent stimulus generalization 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. During the testing (i.e., stimulus generalization) phase of the study, maintenance of the training drug/saline discrimination was ensured by continuation of the training sessions on a daily basis (except on a generalization test day). On one of the two days before a generalization 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 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, and were then returned to their individual home cages. An odd number of training sessions (usually 5) separated any two generalization test sessions. Doses of test drugs were administered to the groups of rats in a random order using a 15-min pre-session injection interval. A determination of complete, partial, or no generalization (or antagonism) was based on previously described criteria (Young and Glennon, 1986). The results of stimulus generalization (or antagonism) tests can produce one of three possible results: 1) complete stimulus generalization, where the animals following a given dose of drug or drug combination make ≥80% of their responses (group mean) on the training drug-appropriate lever, 2) partial generalization, where a challenge drug produces an intermediate (ca. 40–70%) level of responding on the drug-appropriate lever, and 3) no generalization (saline-like responding) where the test drug elicits about 0–20% drug-appropriate responding. Animals making fewer than 5 total responses during the 2.5-min extinction session were considered to be behaviorally disrupted because they failed to meet the testing criteria. 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 for that dose was not plotted. Where complete stimulus generalizations occurred, potency comparisons were made between the training drug and the test agent via calculation of the 50% effective dose (i.e., ED_{50} dose as calculated by the method of Finney, 1952). The ED_{50} dose represents the drug dose where animals would be expected to make 50% of their responses on the drug-appropriate lever. Response rate data were evaluated by the Dunnett's *t*-test ($p<0.05$) for comparison of a control group (i.e., saline or dose of training drug response rate) *versus* experimental dose groups of a test compound.

2.2. Drugs

N-Methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropne hydrochloride (MDMA) was obtained as a gift from NIDA, and *N*-methyl-1-(4-methoxyphenyl)-2-aminopropane hydrochloride (PMMA) was synthesized in our laboratory as previously described (Glennon et al., 1988b). 8-Hydroxy-2-(*N*,*N*-di-*n*-propylamino)tetralin hydrobromide (8-OH DPAT) was purchased from Sigma-Aldrich (St. Louis, MO). (+)Amphetamine sulfate (Sigma-Aldrich) was available in our laboratories from previous studies. *N*-Ethyl- and *N*-*n*-propyl-1-(4-methoxyphenyl)-2-aminopropane hydrochloride (PMEA and PMPA, respectively), and the optical isomers of 1-(3-methoxyphenyl)-2-aminopropane hydrogen oxalate (MMA) were synthesized as described below. All drugs were administered *via* the intraperitoneal (i.p.) route 15 min prior to testing. 8-OH DPAT, when administered in combination with PMMA, was administered at

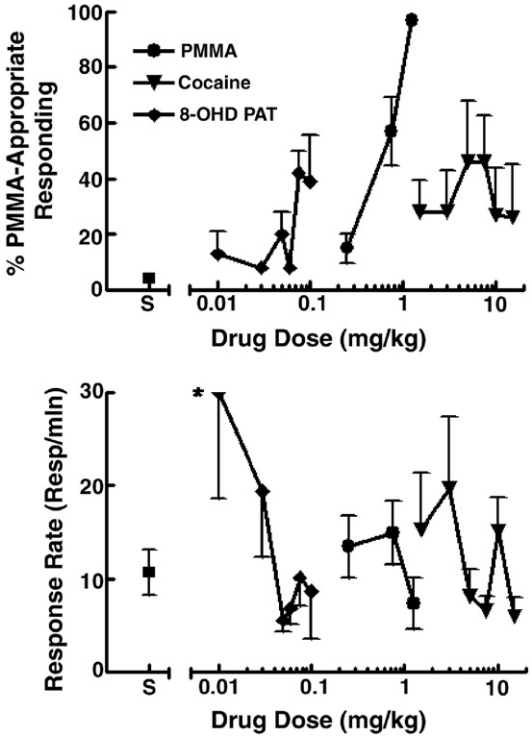

Fig. 2. Results (group mean±SEM) of stimulus generalization studies with PMMA, cocaine, and 8-OH DPAT in rats ($n=8$) trained to discriminate 1.25 mg/kg of PMMA from saline vehicle (upper panel), and animals' response rates (lower panel). *S*=responses following administration of saline. *Response rate following administration of 8-OH DPAT was statistically ($p<0.05$) different from the saline control rate. Administration of cocaine and 8-OH DPAT doses higher than those shown resulted in disruption of the animals' lever-pressing behavior.

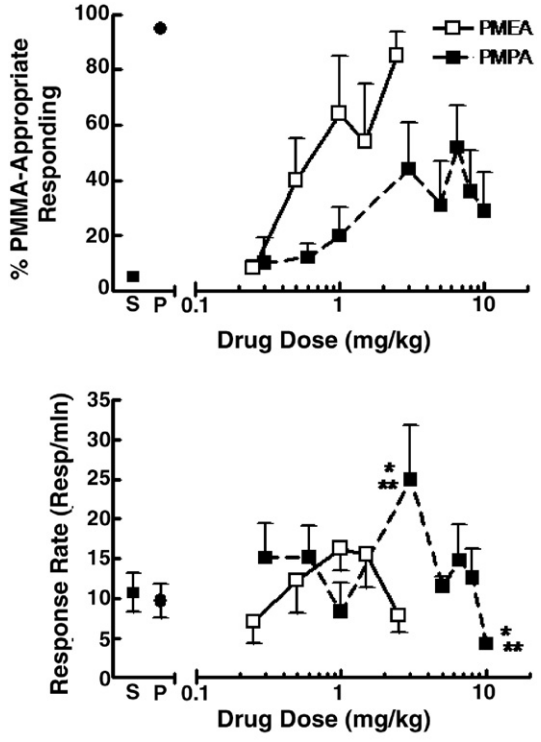

Fig. 3. Results (group mean±SEM) of stimulus generalization studies with *N*-Et PMA (PMEA) and *N*-Pr PMA (PMPA) in rats ($n=5$) trained to discriminate 1.25 mg/kg of PMMA from saline vehicle (upper panel). The animals' response rates are shown in the lower panel. Response rate following administration of PMPA was statistically ($p<0.05$) different from the response rate following administration of the training dose of the training drug (*) or saline (**). PMMA=responses following 1.25 mg/kg of PMMA, and *S*=responses following administration of saline vehicle.

the same time but by separate injection. Doses refer to the weight of the salts. Solutions in sterile 0.9% saline were freshly prepared each day, and administered in a constant volume of 1.0 ml/kg.

PMEA was prepared following the general procedure of Bach et al. (1999) and its homogeneous hydrochloride salt (mp 156–157 °C) was consistent with what has been previously reported in the literature (Woodruff et al., 1940). PMPA hydrochloride (mp 150–154 °C) was prepared in an analogous manner; its assigned structure is consistent with spectral data and the product analyzed within 0.4% of theory for C, H, and N.

Racemic MMA was synthesized by the condensation of 3-methoxybenzaldehyde with nitroethane followed by reduction of the resultant nitrostyrene using lithium aluminum hydride by the general procedure of Shulgin (1968). Resolution of the racemate was achieved using the isomers of tartaric acid to afford the individual MMA optical isomers. The isomers were converted to their free bases and isolated as their hydrogen oxalate salts. The salts of both isomers were homogeneous to thin-layer chromatography and analyzed within 0.4% of theory for C, H, and N. Characteristics: *R*(−)MMA, mp=148–150 °C following recrystallization from an absolute ethanol/anhydrous ether mixture, $[\alpha]_D=-7.3°$ ($c=2\%$, MeOH, 37 °C); *S*(+)MMA, mp=149–151 °C following recrystallization from an absolute ethanol/anhydrous ether mixture, $[\alpha]_D=+7.3°$ ($c=2\%$, MeOH, 37 °C). Samples of the salts were converted to their free bases and their optical rotations were found to be $[\alpha]_D=-34.0°$ ($c=2\%$, CH_2Cl_2, 37 °C) and $[\alpha]_D=+35.1°$ ($c=2\%$, CH_2Cl_2, 37 °C) for *R*(−)- and *S*(+)MMA, respectively. The MMA isomers were used as their water-soluble hydrogen oxalate salts in the animal studies reported herein.

3. Results

3.1. PMMA-trained animals

In the group of eight rats trained to discriminate PMMA from vehicle, administration of lower PMMA doses resulted in reduced percent PMMA-appropriate responding ($ED_{50}=0.5$ mg/kg; 95% CL=0.3–1.0 mg/kg) (Fig. 2). The animals' response rates were fairly consistent following administration of the various doses of PMMA (Fig. 2).

Doses of cocaine were examined in the PMMA-trained animals (Fig. 2). Cocaine produced a maximum of 46% PMMA-appropriate responding (at 5.0 and 7.5 mg/kg). Higher cocaine doses (i.e., 10 and 15 mg/kg) failed to elicit greater PMMA-appropriate responding (Fig. 2), or disrupted the animals' lever-pressing behavior (data not shown). Fig. 2

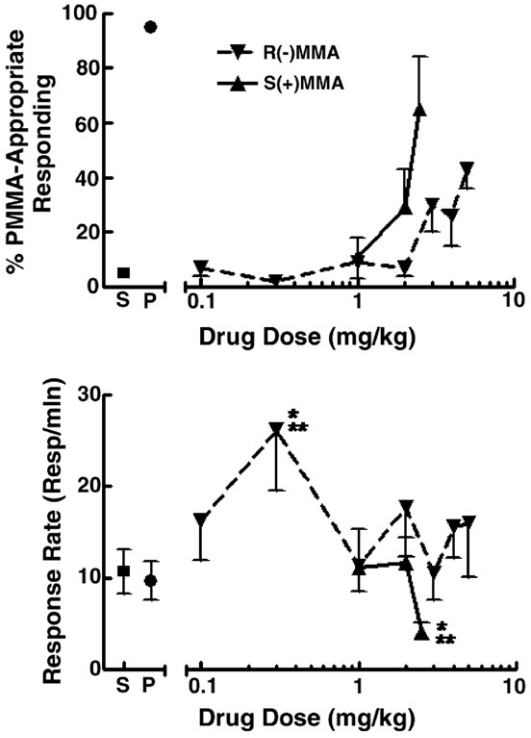

Fig. 4. Results (group mean±SEM) of stimulus generalization studies with the optical isomers of MMA in rats ($n=5$) trained to discriminate 1.25 mg/kg of PMMA from saline vehicle (upper panel). The animals' response rates are shown in the lower panel. Response rate following administration of *R*(−)MMA or *S*(+)MMA was statistically ($p<0.05$) different from response rates following administration of the training dose of the training drug (*) or saline (**). PMMA=responses following 1.25 mg/kg of PMMA, and *S*=responses following administration of saline vehicle.

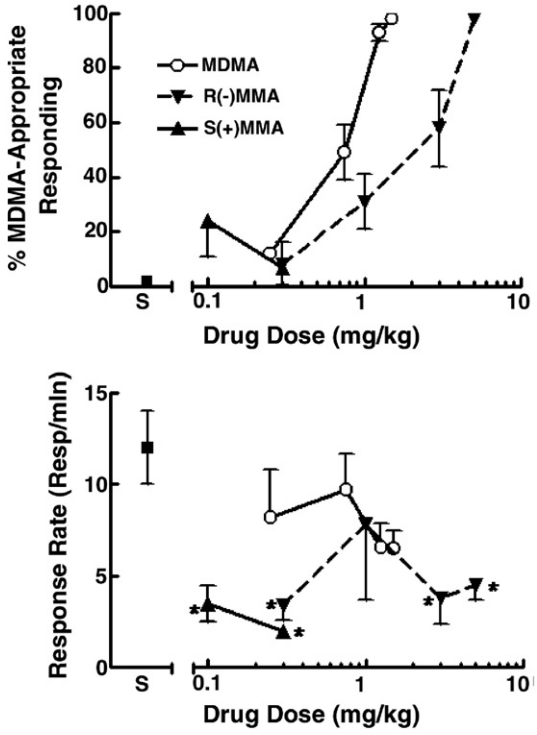

Fig. 5. Results (group mean±SEM) of stimulus generalization studies with the optical isomers of MMA in rats ($n=5$) trained to discriminate 1.5 mg/kg of MDMA from saline vehicle (upper panel). The animals' response rates are shown in the lower panel. *Response rate was statistically ($p<0.05$) different from that of the saline control response rate. *S*=responses following administration of saline vehicle.

shows the animals' response rates. Except at the lowest dose evaluated, one animal failed to respond following administration of each of the higher cocaine doses shown in Fig. 2.

The administration of 8-OH DPAT doses to the PMMA-trained animals ($n=5$–6) resulted in partial (i.e., in a maximum of 43%) PMMA-appropriate responding at a dose of 0.075 mg/kg. Higher doses (e.g. 0.1 mg/kg) either failed to produce greater drug-appropriate responding (Fig. 2), or (at 0.15 and 0.2 mg/kg; data not shown) resulted in disruption of lever-pressing behavior. Following administration of 0.01 mg/kg of 8-OH DPAT the animal's response rate was statistically ($p<0.05$) different from, and nearly three times, the saline control rate (Fig. 2); however, response rates were reduced to control rates following administration of higher 8-OH DPAT doses. At each dose (except for the 0.06 mg/kg dose), one animal failed to make >5 responses during the entire 2.5-min extinction session.

Administration of the calculated ED_{50} dose of PMMA resulted in the animals making 51(±10)% of their responses (response rate=12.9±3.2 responses/min) on the PMMA-appropriate lever (data not shown). Administered in combination with the ED_{50} dose of PMMA, 8-OH DPAT doses of 0.06 to 0.1 mg/kg failed to enhanced (or antagonize) the PMMA stimulus effect. Doses of 8-OH DPAT (followed by percent PMMA-appropriate responding and response rate) were 0.06 mg/kg (53±16%; 12.3±2.9 resp/min), 0.075 mg/kg (40±13%; 10.2±2.8 resp/min) and 0.1 (58±11%; 8.1±4.5 resp/min) (data not shown). All six animals responded following administration of 0.06 mg/kg of 8-OH DPAT, but only 4/5 and 5/6 animals responded following administration of 0.075 and 0.1 mg/kg, respectively. Administration of 0.15 mg/kg of 8-OH DPAT in combination with the ED_{50} dose of PMMA disrupted the lever-pressing behavior of all six animals.

Administration of the *N*-ethyl homolog of PMMA (PMEA; Fig. 3) to the PMMA-trained animals ($n=5$) resulted in dose-related substitution ($ED_{50}=0.9$ mg/kg; 95% CL=0.4–1.8 mg/kg). Administration of the *N*-propyl homolog of PMMA (i.e., PMPA), however, elicited a maximum of 52% PMMA-appropriate responding (at 6.5 mg/kg), with no further increase in percent PMMA-appropriate responding following administration of higher doses (Fig. 3). The animals' response rates were fairly consistent across drug treatments following administration of 2.0 mg/kg of PMPA, however, there was a significant ($p<0.05$) spike in response rate; higher drug doses resulted in response rates that were not markedly different from control rates except that 10 mg/kg of PMPA produced a significant ($p<0.05$) decrease in the animals' responding.

The optical isomers of MMA were administered to the PMMA-trained animals, and neither isomer produced ≥80% PMMA-appropriate responding (Fig. 4). Nine doses of *R*(−)MMA were examined. The administration of *R*(−)MMA at 5.0 mg/kg produced a maximum of 43% PMMA-appropriate responding. *R*(−)MMA doses of 5.5 and 6.0 mg/kg produced disruption of behavior (data not shown). Following

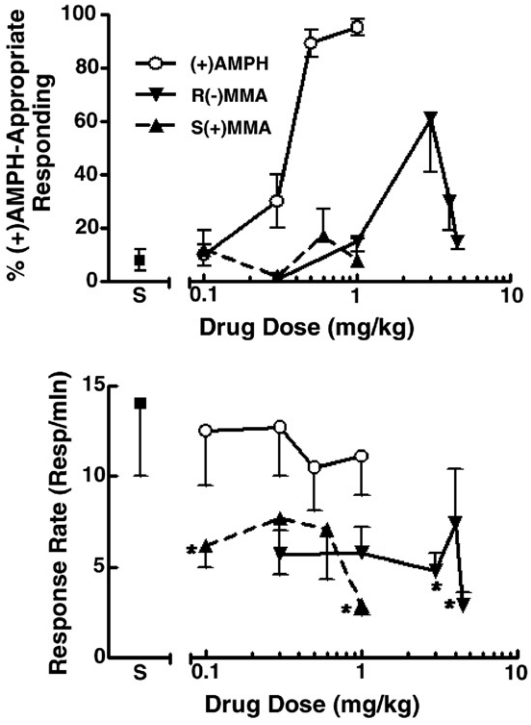

Fig. 6. Results (group mean±SEM) of stimulus generalization studies with the optical isomers of MMA in rats ($n=5$) trained to discriminate 1.0 mg/kg of (+)amphetamine from saline vehicle (upper panel). The animals' response rates are shown in the lower panel. *Response rate was significantly ($p<0.05$) different from the saline control response rate. *S*=responses following administration of saline vehicle.

administration of 0.3 mg/kg of *R*(−)MMA there was a significant ($p<0.05$) increase in response rate in comparison to saline or PMMA control rates. The administration of *S*(+)MMA at 2.5 mg/kg induced a maximum of 65% PMMA-appropriate responding and at this dose 4/8 animals met criteria; animals' response rate at this *S*(+)MMA dose was significantly ($p<0.05$) less than the saline or PMMA response rate. Administered *S*(+)MMA doses of 2.75 and 3.0 mg/kg, a majority of the animals failed to respond (disruption data not shown).

3.2. MDMA-trained animals

MDMA stimulus generalization occurred to *R*(−)MMA (ED_{50}=1.4 mg/kg; 95% CL=0.6–3.1 mg/kg) whereas *S*(+)MMA produced saline-appropriate responding at 0.1 and 0.3 mg/kg (Fig. 5). Administration of higher *S*(+)MMA doses produced behavioral disruption (data not shown); that is, at 0.6 or 1.0 mg/kg of *S*(+)MMA only 2/5 animals met criteria. Following administration of 0.1 and 0.3 mg/kg of *S*(+)MMA, and 0.3, 3.0, and 5.0 mg/kg of R(−)MMA, animals' response rate was significantly ($p<0.05$) less than the saline control response rate.

3.3. (+)Amphetamine-trained animals

Fig. 6 shows that *R*(−)MMA produced a maximum of 61% (+)amphetamine-appropriate responding (at a dose of 3.0 mg/kg) with 4/5 animals responding; administration of higher *R*(−)MMA doses failed to engender greater (+)amphetamine-appropriate responding, whereas following administration of 5.0 mg/kg of *R*(−)MMA the animals were behaviorally impaired (data not shown). Following administration of 3.0 and 4.5 mg/kg of *R*(−)MMA the animals' response rates were significantly ($p<0.05$) less than the saline control response rate. Administration of *S*(+)MMA produced saline-appropriate responding at all doses examined (Fig. 6). The animals' response rates appeared depressed relative to saline control at all *S*(+)MMA doses examined (Fig. 6), and only 3/5 animals met the criteria at the two highest doses.

4. Discussion

In prior drug discrimination studies in rats, MDMA and PMMA substituted for one another implying that the two agents are capable of inducing similar stimulus effects. However (see Introduction), there are indications that their stimulus effects are not identical. To further examine this concept, several issues were selected for closer inspection: a) because an MDMA stimulus generalizes to cocaine, will cocaine produce PMMA-like stimulus effects, b) because MDMA stimulus generalization occurs to 8-OH DPAT, will the PMMA-stimulus generalize to 8-OH DPAT, and if not, might 8-OH DPAT modulate the stimulus effects of PMMA, c) will *N*-homologation have the same effect on PMMA as it has on MDMA, and d) will optical isomers of MMA (a positional isomer of PMA) substitute in PMMA- and/or MDMA-trained animals?

It has been previously shown that PMMA lacks central stimulant-like actions associated with MDMA. For example, a (+)amphetamine training stimulus generalizes to MDMA in rats (Glennon et al., 1982), pigeons (Evans and Johanson, 1986), and monkeys (Kamien et al., 1986) (see also Bondareva et al., 2005 for additional discussion); similarly, an MDMA stimulus generalizes (Oberlender and Nichols, 1988) or partially generalizes (Glennon and Misenheimer, 1989) to (+)amphetamine. In contrast, an amphetamine stimulus failed to generalize to PMMA (Glennon et al., 1988b) and PMMA-stimulus generalization did not occur to (+)amphetamine (i.e., the maximum PMMA-appropriate responding occasioned following administration of (+)amphetamine was 11%) (Glennon et al., 1997). Because an MDMA stimulus generalizes to cocaine (Khorana et al., 2004), cocaine was examined in PMMA-trained animals. As shown in Fig. 2, cocaine engendered a maximum of 46% PMMA-appropriate responding. This result is consistent with the prior suggestion that MDMA possesses greater central stimulant character than PMMA (Glennon et al., 1997) and, taken together with the above mentioned findings with (+)amphetamine, represent a major difference between the stimulus effects produced by MDMA and PMMA in rats.

Although the mechanism of action of MDMA as a discriminative stimulus is complex, it seems to involve, at least in part, 5-$HT_{1A/7}$ serotonin receptors; that is, an MDMA stimulus has been shown to generalize to 8-OH DPAT (Glennon and Young, 2000). Unlike what was found with MDMA-trained animals, the PMMA stimulus did not generalize to the 5-$HT_{1A/7}$ agent 8-OH DPAT (Fig. 2). Furthermore, although 8-OH DPAT does not substitute for a (+)amphetamine stimulus, pretreatment of (+)amphetamine-trained rats with 8-OH DPAT enhanced its stimulus actions (Young et al., 2006). In the present investigation, 8-OH DPAT failed to enhance (or attenuate) the actions of PMMA in PMMA-trained animals.

The structure–activity relationships for agents to produce MDMA-like stimulus effects have been explored to some extent, and it is known that homologation of the terminal amine substituent from a methyl group to an ethyl group (i.e., MDE) results in an agent with stimulus properties similar to those of MDMA (Glennon, 1989; Nichols and Oberlender, 1989). As with MDMA, the PMMA stimulus generalized to its simple *N*-ethyl homolog (i.e., PMEA; ED_{50}=0.9 mg/kg) (Fig. 3). Evidently, MDMA and PMMA behave similarly with regard to homologation of the terminal amine substituent from a methyl group to an ethyl group. The *N*-propyl homolog of MDMA (i.e., MDP) has not been examined as a discriminative stimulus, but seems to lack MDMA-like actions in humans (Shulgin and Shulgin, 1991). It was of interest, then, to determine whether or not further homologation of PMMA to its propyl homolog, PMPA, would retain PMMA-like stimulus character. If so, this might represent a potential difference in structure–activity relationships between MDMA and PMMA. As shown in Fig. 3, however, administration of PMPA resulted only in partial substitution (i.e., PMPA produced a maximum of 52% PMMA-appropriate responding) in PMMA-trained animals. Thus, it was found that although homologation of the *N*-methyl group of PMMA to an *N*-ethyl group results in retention of its

stimulus actions, the *N*-propyl homolog, PMPA, does not substitute in PMMA-trained animals. With regard to one-carbon homologation, both MDMA and PMMA behaved in a similar manner.

Because 3,4-DMA seems to be the structural parent both of MDMA and PMMA (i.e., conversion of the 3,4-dimethoxy to a 3,4-methylenedioxy group ultimately leads to MDMA whereas elimination of the 3-methoxy group of 3,4-DMA ultimately leads to PMMA), and because both optical isomers of 3,4-DMA substitute in MDMA- and PMMA-trained animals, it was important to determine the role of the 4-oxygen substituent common to each of these agents. Consequently, the 4-methoxy group of 3,4-DMA was eliminated to afford MMA. The PMMA stimulus only partially generalized either to *R*(−)- or *S*(+)MMA (Fig. 4). In contrast, the MDMA stimulus generalized completely to *R*(−)MMA (ED_{50}=1.4 mg/kg) but not to *S*(+)MMA (Fig. 5). It would seem that the presence of the 3-oxygen function of MDMA is what makes MDMA behave somewhat differently than PMMA as a discriminative stimulus.

Perhaps the most significant quality differentiating the actions of MDMA and PMMA is that the former are more "stimulant-like". This raises the question of whether *R*(−)MMA substituted in the MDMA-trained, but not in PMMA-trained, animals because of its amphetamine-like stimulus character. As a consequence, both MMA isomers were evaluated in (+) amphetamine-trained rats (Fig. 6). Interestingly neither MMA isomer substituted completely for the (+)amphetamine stimulus, but *R*(−)MMA resulted in partial generalization (i.e., 61% (+) amphetamine-appropriate responding). Although *R*(−)-MMA cannot be considered a simple amphetamine-like agent, it seems to possess some amphetamine-like stimulus character. But, it is unlikely that the partial amphetamine-like character of *R*(−) MMA, by itself, can account for the difference seen upon its administration to the PMMA- and MDMA-trained animals.

Overall, the results of the present investigation support the contention that similarities and differences exist between the stimulus properties of MDMA and PMMA. Adding to the list of similarities is that homologation of the terminal amine substituent of MDMA and PMMA from a methyl to an ethyl group result in agents that substitute in MDMA- and PMMA-trained animals, respectively. In contrast, whereas MDMA seems to exert amphetamine-like and cocaine-like actions in animals (Glennon, 1989; Nichols and Oberlender, 1989), a PMMA stimulus, as shown here, did not substitute fully for cocaine. Also, whereas *R*(−)MMA substituted in MDMA-trained animals the MMA optical isomers failed to substitute in the PMMA-trained animals. In summary, then, the results of this investigation show that although MDMA and PMMA might share some stimulus similarities, they produce stimulus effects in rats that are clearly distinguishable from one another. Moreover, a 3-position oxygen substituent (common to MDMA, MDA, and 3,4-DMA, but absent in PMMA), might structurally account for differences observed between the stimulus actions of MDMA and PMMA, whereas any similarities in their effects might be more related to the presence of an oxygen substituent at the aromatic ring 4-position. An interesting study to further investigate the stimulus effects of MDMA and PMMA would be to train animals to discriminate these agents from one another; such studies are currently under consideration in our laboratories.

Acknowledgment

This work was supported, in part, by DA 01642 (RAG).

References

Bach MV, Coutts RT, Baker GB. Involvement of CYP2D6 in the *in vivo* metabolism of amphetamine, two *N*-alkylamphetamines, and their 4-methoxylated derivatives. Xenobiotica 1999;29:719–32.

Baylen CA, Rosenberg H. A review of the acute subjective effects of MDMA/ecstasy. Addiction 2006;101:933–47.

Bondareva T, Wesolowska A, Dukat M, Lee M, Young R, Glennon RA. *S*(+)- and *R*(−)*N*-methyl-1-(3,4-methylenedioxyphenyl)-2-aminopropane (MDMA) as discriminative stimuli: effect of cocaine. Pharmacol Biochem Behav 2005;82:531–8.

Bustamante D, Diaz-Veliz G, Paeile C, Zapata-Torres G, Cassels BK. Analgesic and behavioral effects of amphetamine enantiomers, *p*-methoxyamphetamine and *n*-alkyl-*p*-methoxyamphetamine derivatives. Pharmacol Biochem Behav 2004;79:199–212.

Dal Cason T. A re-examination of the mono-methoxy positional ring isomers of amphetamine, methamphetamine and phenyl-2-propanone. Forensic Sci Int 2001;119:168–94.

Dukat M, Young R, Glennon RA. Effect of PMA optical isomers and 4-MTA in PMMA-trained rats. Pharmacol Biochem Behav 2002;72:299–305.

Evans SM, Johanson CE. Discriminative stimulus properties of (±)methylenedioxymethamphetamine and (±)methylenedioxyamphetamine in pigeons. Drug Alcohol Depend 1986;18:159–64.

Finney D. Probit Analysis. London: Cambridge University Press; 1952.

Glennon RA. Stimulus properties of hallucinogenic phenylalkylamines and related designer drugs: formulation of structure–activity relationships. NIDA Res Monogr 1989;94:43–67.

Glennon RA. Phenylalkylamine stimulants, hallucinogens, and designer drugs. NIDA Res Monogr 1990;105:154–60.

Glennon RA, Higgs R. Investigation of MDMA-related agents in rats trained to discriminate MDMA from saline. Pharmacol Biochem Behav 1992;43:759–63.

Glennon RA, Misenheimer B. Stimulus effects of *N*-mono ethyl-(3,4-methylenedioxyphenyl)-2-aminopropane (MDE) and *N*-hydroxy-1-(3,4-methylenedioxyphenyl)-2-aminopropane (*N*-OH MDA) in rats trained to discriminate MDMA from saline. Pharmacol Biochem Behav 1989;33:909–12.

Glennon RA, Young R. MDMA stimulus generalization to the 5-HT_{1A} serotonin agonist 8-hydroxy-2-(di-*n*-propylamino)tetralin. Pharmacol Biochem Behav 2000;66:483–8.

Glennon RA, Young R. Effects of 1-(3,4-methylenedioxyphenyl)-2-aminopropane and its optical isomers in PMMA-trained rats. Pharmacol Biochem Behav 2002;72:307–11.

Glennon RA, Young R, Hauck AE. Structure–activity studies on methoxy-substituted phenylisopropylamines using drug discrimination methodology. Pharmacol Biochem Behav 1985;22:723–9.

Glennon RA, Yousif M, Patrick G. Stimulus properties of 1-(3,4-methylenedioxyphenyl)-2-aminopropane (MDA) analogs. Pharmacol Biochem Behav 1988a;29:443–9.

Glennon RA, Young R, Rosecrans JA, Anderson GM. Discriminative stimulus properties of MDA analogs. Biol Psychiatry 1982;17:804–7.

Glennon RA, Ismaiel AM, Martin B, Poff D, Sutton M. A preliminary behavioral investigation of PMMA, the 4-methoxy analog of methamphetamine. Pharmacol Biochem Behav 1988b;31:9–13.

Glennon RA, Young R, Dukat M, Cheng Y. Initial characterization of PMMA as a discriminative stimulus. Pharmacol Biochem Behav 1997;57:151–8.

Green AR, Mechan AO, Elliott JM, O'Shea E, Colado MI. The pharmacology and clinical pharmacology of 3,4-methylenedioxymethamphetamine (MDMA, "Ecstasy"). Pharmacol Rev 2003;55:463–508.

Huang JT, Ho BT. Discriminative stimulus properties of D-amphetamine and related compounds in rats. Pharmacol Biochem Behav 1974;2:669–73.

Kamien JB, Johanson CE, Schuster CR, Woolverton WL. The effects of (±) methylenedioxymethamphetamine and (±)methylenedioxyamphetamine in monkeys trained to discriminate (+)amphetamine from saline. Drug Alcohol Depend 1986;18:139–47.

Khorana N, Pullagurla MR, Young R, Glennon RA. Comparison of the discriminative stimulus effects of 3,4-methylenedioxymethamphetamine (MDMA) and cocaine: asymmetric generalization. Drug Alcohol Depend 2004;74:281–7.

Nichols DE, Oberlender R. Structure–activity relationships of MDMA-like substances. NIDA Res Monogr 1989;94:1–29.

Oberlender R, Nichols DE. Drug discrimination studies with MDMA and amphetamine. Pyschopharmacology (Berl) 1988;95:71–6.

Rangisetty JB, Bondarev ML, Chang-Fong J, Young R, Glennon RA. PMMA-stimulus generalization to the optical isomers of MBDB and 3,4-DMA. Pharmacol Biochem Behav 2001;69:261–7.

Shulgin AT. The ethyl homologs of 2,4,5-trimethoxyphenylisopropylamine. J Med Chem 1968;11:186–7.

Shulgin A, Shulgin A. Pihkal. Berkeley: Transform Press; 1991. p. 753–5.

Woodruff EH, Lambooy JP, Burt WE. Physiologically active amines. III. Secondary and tertiary β-phenylpropylamines and β-phenylisopropylamines. J Am Chem Soc 1940;62:922–4.

Young R, Glennon RA. Amphetamine and related phenyalkylamines as discriminative stimuli: a review of the literature. Med Res Rev 1986;6:99–130.

Young R, Dukat M, Malmusi L, Glennon RA. Stimulus properties of PMMA: effect of optical isomers and conformational restriction. Pharmacol Biochem Behav 1999;64:449–53.

Young R, Bondareva T, Wesolowska A, Glennon RA. Modulation of a (+) amphetamine discriminative stimulus in rats by 8-hydroxy-2-(*N*,*N*-di-*n*-propylamino)tetralin (8-OH DPAT). Pharmacol Biochem Behav 2006;83:612–7.