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Differential effects of 7-OH-DPAT on the development of behavioral sensitization to apomorphine and cocaine

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Abstract

The primary objective of this study was to determine whether concurrent treatments with a low dose of the dopamine D_3-preferring receptor agonist 7-OH-DPAT would attenuate the development of behavioral sensitization to the indirect dopamine receptor agonist, cocaine, or the direct dopamine receptor agonist, apomorphine. In two experiments, male Wistar rats (250–350 g) were given seven daily injections of 7-OH-DPAT (0.05 mg/kg sc) or vehicle in combination with either cocaine (15 mg/kg ip), apomorphine (1.0 mg/kg sc), or vehicle. After the injections, the rats were tested for activity in photocell arenas for 40 min, and three measures of motor behavior (distance traveled, rearing, and stereotypy) were recorded at 10-min intervals. A total of 24 h after the last preexposure session, all rats were given a challenge injection of either cocaine (10.0 mg/kg ip, Experiment 1) or apomorphine (1.0 mg/kg sc, Experiment 2) and tested for activity. Major findings were as follows: (a) 7-OH-DPAT treatments alone suppressed all measures of locomotor activity and did not affect subsequent behavioral sensitivity to either cocaine or apomorphine; (b) cocaine treatments acutely increased all measures of activity, and repeated treatments produced behavioral sensitization to the horizontal locomotor-activating effects of cocaine; (c) apomorphine treatments alone increased horizontal activity and stereotypy but completely abolished rearing behavior; (d) like cocaine, repeated treatments with apomorphine induced behavioral sensitization; (e) concurrent treatments of 7-OH-DPAT with cocaine acutely attenuated cocaine-induced increases in motor behavior but enhanced the development of behavioral sensitization to cocaine; and (f) concurrent 7-OH-DPAT treatments did not significantly affect either the acute or chronic effects of apomorphine. It is evident from these results that concurrent treatment with 7-OH-DPAT does not block the development of behavioral sensitization to either cocaine or apomorphine. Moreover, the differential acute and chronic effects of 7-OH-DPAT on cocaine- and apomorphine-induced hyperactivity appear to be mediated by dopamine autoreceptor stimulation.

Keywords: Behavioral sensitization; 7-OH-DPAT; Cocaine; Apomorphine; Locomotor activity; Dopamine D_3 receptors; Dopamine D_2 receptors; Autoreceptors

1. Introduction

It has been repeatedly suggested in recent years that the dopamine D_3 receptor may be an important target for the development of pharmacotherapies for the treatment of psychostimulant drug abuse (e.g., Caine and Koob, 1993; Caine et al., 1997; Khroyan et al., 1998; Shafer and Levant, 1998). Consistent with this suggestion, dopamine D_3 receptors appear to be preferentially located in limbic regions important for incentive motivation and reward (see Shafer and Levant, 1998 for review), and the number of binding sites for D_3 receptors are significantly elevated in cocaine-overdosed victims (Staley and Mash, 1996). In addition, the dopamine D_3-preferring receptor agonist, 7-OH-DPAT, modulates cocaine self-administration in rats at doses that are not readily self-administered alone (Caine and Koob, 1993, 1995; Caine et al., 1999; Nader and Mach, 1996; Parsons et al., 1996), substitutes for cocaine, amphetamine, and apomorphine in drug discrimination tasks (Acri et al., 1995; Baker et al., 1998; Sanger et al., 1997), and attenuates the incentive motivational properties of amphetamine, cocaine, and morphine as measured by conditioned place preference (CPP) tests (DeFonseca et al., 1995; Khroyan et al., 1998, 1999). Based upon these and other findings, it has been suggested that 7-OH-DPAT maybe useful in the treatment of drug craving in recovering addicts (e.g., Caine and Koob, 1993; Caine et al., 1997).

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Like dopamine D_2 receptors, D_3 receptors appear to be located on both pre- and postsynaptic membranes (e.g., Aretha et al., 1995; Booth et al., 1994; Gilbert et al., 1995; Svensson et al., 1994b). Moreover, considerable evidence suggests that the D_3 presynaptic receptor functions as an autoreceptor (see Shafer and Levant, 1998 for review). For example, acute treatment with 7-OH-DPAT produces hypoactivity in rats (e.g., Mattingly et al., 1996; Svensson et al., 1994b), as well as a significant decrease in dopamine synthesis, release, and dopamine cell firing rates (e.g., Acri et al., 1995; Gilbert et al., 1995; Kreiss et al., 1995; Lejeune and Millan, 1995). Despite this evidence, however, it has been more recently reported that the dopamine D_3-preferring agonist, PD 128907, produces similar effects in models of autoreceptor function in wild-type and D_3 "knock-out" mice (Koeltzow et al., 1998). Based upon this result, it has been argued that D_3 receptors do not function as autoreceptors (Koeltzow et al., 1998). Consistent with this conclusion, decreases in locomotor activity have been reported following low doses of 7-OH-DPAT that do not affect dopamine synthesis or release (Mattingly et al., 1996; Svensson et al., 1994b). Based upon this latter finding, it has been suggested that dopamine D_3 postsynaptic receptors, unlike postsynaptic D_2 receptors, may be inhibitory with respect to locomotor behavior (Svensson et al., 1994a,b).

Many of the behavioral effects of psychostimulant drugs, including cocaine and amphetamine, increase with repeated drug exposure (Kalivas and Stewart, 1993; Robinson and Becker, 1986; Stewart and Badiani, 1993). This phenomenon, termed behavioral sensitization, has now been extensively documented for both the locomotor-activating and rewarding effects of psychostimulant drugs (e.g., Pierre and Vezina, 1997; Robinson and Berridge, 1993; Shippenberg and Heidbreder, 1995), and has been proposed as an animal model for the development and persistence of drug craving (Kalivas et al., 1998; Robinson and Berridge, 1993). Previous research indicates that nearly all dopamine agonists, including 7-OH-DPAT, produce behavioral sensitization with repeated treatments (e.g., Mattingly et al., 1996). However, although high doses of 7-OH-DPAT result in the development of behavioral sensitization with repeated treatment, repeated treatments with low doses of 7-OH-DPAT, presumably selective for D_3 receptors (Levant et al., 1996), inhibit locomotor activity and do not induce behavioral sensitization (Mattingly et al., 1996).

The purpose of the current study, therefore, was to determine whether concurrent treatments with a low dose 7-OH-DPAT would attenuate the development of behavioral sensitization. Consequently, in Experiment 1, rats were treated daily with a low dose of 7-OH-DPAT combined with either cocaine or vehicle and tested for locomotor activity. After this pretreatment, all rats were tested for activity following a challenge dose of cocaine. Experiment 2 was the same as Experiment 1, except that the direct dopamine agonist, apomorphine, was used in place of cocaine. Although the development of behavioral sensitization to both cocaine and apomorphine is presumed to result from repeated dopamine receptor stimulation (e.g., Henry et al., 1989; Henry and White, 1995; Mattingly et al., 1991), the two drugs differ in mechanism of action. Cocaine promotes an increase in extracellular dopamine by blocking the dopamine transporter, whereas, apomorphine is a relatively nonselective direct dopamine receptor agonist. Thus, concurrent 7-OH-DPAT treatments may differentially affect the development of sensitization to these two drugs (cf. Khroyan et al., 1999).

2. Materials and methods

2.1. Subjects

In each experiment, 32 male Wistar albino rats (Harlan Sprague–Dawley; Indianapolis, IN) weighing between 250 and 350 g served as subjects. The rats were housed in the colony room for at least 1 week prior to behavioral testing. All rats were housed individually in hanging wire-mesh cages in a colony room with a 12-h light–dark cycle and food and water available continuously. All behavioral testing was conducted during the light phase of the cycle.

2.2. Apparatus

Activity measures were taken in four acrylic open-field chambers (Med-Associates Model OFA-163) located individually in sound-attenuated experimental cubicles. These chambers were equipped with a 16×16 array of photocell beams and detectors 2.5 cm above the floor and singe array of 16 photocell beams and detectors 10 cm above the floor. In each chamber, a clear acrylic cylinder (41 cm in diameter) was placed inside the outer square chamber. The photocell banks were connected to a microcomputer in an adjoining room. Using Med-Associates software, three measures of activity were recorded at 10-min intervals: distance traveled (cm), number of rears, and stereotypy (small movements).

2.3. Drugs

Apomorphine hydrobromide (Sigma), cocaine hydrochloride (NIDA), and (±)-7-hydroxy-dipropylaminotetralin hydrobromide (7-OH-DPAT; Research Biochemicals) were dissolved in distilled H_2O and injected in a volume of 1.0 ml/kg. Doses of each drug were calculated based upon the salt form of the drug. Vehicle injections were given using the same route and volume as the corresponding drug injection. Doses of apomorphine (1.0 mg/kg) and cocaine (15 mg/kg) known to reliably induce behavioral sensitization were chosen based upon previous research in our laboratory (Mattingly et al., 1988a,b, 1991, 1994). The dose of 7-OH-DPAT (0.05 mg/kg) was chosen, because it acutely decreases activity, does not induce behavioral sensitization with repeated treatment, and has been suggested to be

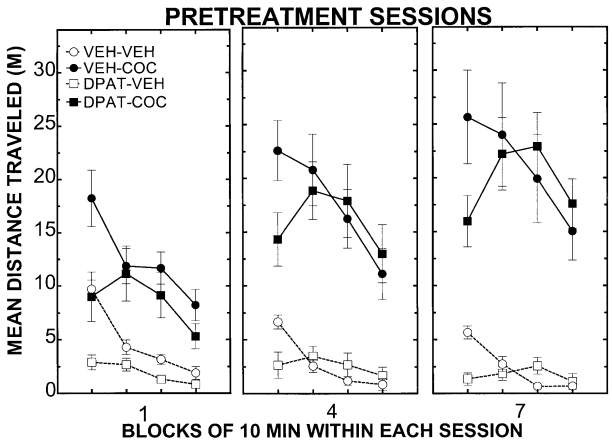

Fig. 1. Mean ± S.E.M. distance traveled across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 15.0 mg/kg cocaine (COC).

selective for dopamine D_3 receptors (Gainetdinov et al., 1996; Levant et al., 1996; Mattingly et al., 1996).

2.4. Design and procedure

At the beginning of Experiment 1, 32 rats were randomly assigned in equal numbers to one of four treatment groups comprising the two (7-OH-DPAT dose: 0 or 0.05 mg/kg) × two (cocaine dose: 0 or 15.0 mg/kg) factorial design. The design of Experiment 2 was the same as Experiment 1, except that apomorphine (0 or 1.0 mg/kg) was used in place of cocaine. In Experiment 1, prior to each of the first seven sessions (training phase), the rats were first injected subcutaneously (sc) with either vehicle or 7-OH-DPAT and then immediately injected intraperitoneally (ip) with either vehicle or cocaine. A total of 5 min following the second injection, each rat was placed into the activity chamber. On Session 8 (sensitization test), all rats received a vehicle injection followed immediately by an injection of cocaine (10.0 mg/kg). The same procedure was followed in Experiment 2, except that 7-OH-DPAT and apomorphine were administered subcutaneously in a cocktail during the training phase, and a single injection of apomorphine (1.0 mg/kg) was administered on the sensitization test. In all phases of the experiments, the rats were returned to their homecage after the injections prior to activity testing. All activity test sessions were 40 min in duration and separated by 24-h drug-free intervals. This experimental protocol was reviewed and approved by the Institutional Animal Care and Use Committee at Morehead State University.

2.5. Data analysis

Significant differences among the groups in mean activity counts during the training phase (Sessions 1–7) were determined with mixed four-factor analyses of variance (ANOVA) using drug treatment conditions as between factors, and test session and blocks of 10 min within each session as repeated measures. Significant interactions were analyzed with additional ANOVAs performed on individual session and/or block data followed by Newman–Keuls post-hoc tests. Mean activity counts of the groups on the sensitization test (Session 11) were analyzed using mixed three-factor ANOVAs.

3. Results

3.1. Experiment 1: 7-OH-DPAT and cocaine

3.1.1. Training sessions — distance traveled

The mean distance traveled by the four groups across the four 10-min time blocks on Sessions 1, 4, and 7 are displayed in Fig. 1. As may be seen in Fig. 1, cocaine produced a progressive increase in horizontal locomotor activity with repeated administration [cocaine effect, $F(1,28) = 227.92$, $P < .0001$, and Cocaine × Session interaction, $F(6,168) = 7.05$, $P < .0001$]. In contrast, 7-OH-DPAT suppressed activity in both vehicle- and cocaine-treated rats on the first 10-min block of each session [7-OH-DPAT × Block, $F(3,84) = 46.59$, $P < .0001$]. However, although the 7-OH-DPAT-induced suppression of activity did not significantly change across sessions in vehicle-treated rats, the activity of rats injected with both 7-OH-DPAT and cocaine significantly increased across sessions [7-OH-DPAT × Cocaine × Block interaction, $F(3,84) = 5.55$, $P < .002$, and 7-OH-DPAT × Cocaine × Session × Block interaction, $F(18,504) = 1.96$, $P = .0105$]. Thus, while concurrent 7-OH-DPAT treatments produced an initial inhibition of cocaine-induced activity across sessions, it did not prevent the development of behavioral sensitization.

3.1.2. Sensitization test — distance traveled

The results of the cocaine challenge on horizontal locomotor activity are shown in Fig. 2. As may be seen the left

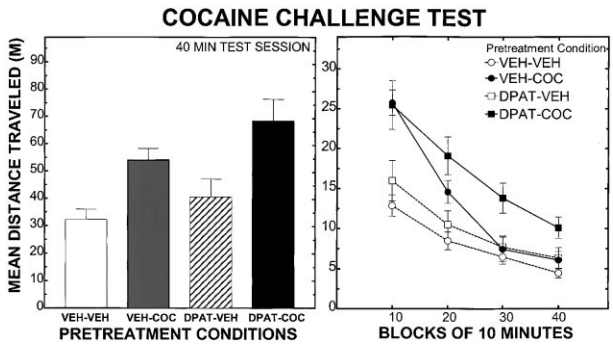

Fig. 2. Mean ± S.E.M. distance traveled after a cocaine challenge injection (10.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or cocaine (15.0 mg/kg, COC). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

panel, rats previously treated with cocaine with or without 7-OH-DPAT were significantly more active after the cocaine challenge injection than rats previously treated with either 7-OH-DPAT alone or vehicle [cocaine effect, $F(1,28) = 17.74$, $P = .0002$]. However, as may be seen in the right panel of Fig. 2, this effect varied across blocks [Cocaine × Block interaction, $F(3,84) = 16.59$, $P = .0001$; 7-OH-DPAT × Cocaine × Block interaction, $F(3,84) = 3.60$, $P = .0169$]. Indeed, subsequent analysis of the triple interaction with a Newman–Keuls post-hoc test indicated that cocaine-pretreated rats were significantly more active than the vehicle control rats only on Blocks 1 and 2 [P's < .05]. In contrast, the rats pretreated with 7-OH-DPAT alone did not differ significantly from the vehicle control rats on any block [P's > .05]. More important, the rats previously treated with both 7-OH-DPAT and cocaine were significantly more active than the other three groups on Blocks 2, 3, and 4 [P's < .05]. Thus, concurrent treatments with 7-OH-DPAT and cocaine during the training sessions appeared to prolong the effectiveness of the cocaine challenge treatment.

3.1.3. Training sessions — rears

The effects of 7-OH-DPAT and cocaine on rearing are shown in Fig. 3. As may be seen, cocaine significantly stimulated rearing behavior compared to vehicle control rats [cocaine effect, $F(1,28) = 57.77$, $P < .0001$]. However, this cocaine-induced increase in rearing did not significantly change across sessions. In contrast, 7-OH-DPAT treatments significantly suppressed rearing across all sessions [7-OH-DPAT effect, $F(1,28) = 10.74$, $P = .0028$], with the greatest suppression occurring on the early blocks of each session [7-OH-DPAT × Block interaction, 67.00, $P < .0001$]. Concurrent 7-OH-DPAT treatments also significantly inhibited the cocaine-induced increase in rearing across sessions, particularly on the early blocks of each session [7-OH-DPAT × Cocaine × Block, $F(3,84) = 7.90$, $P < .0001$].

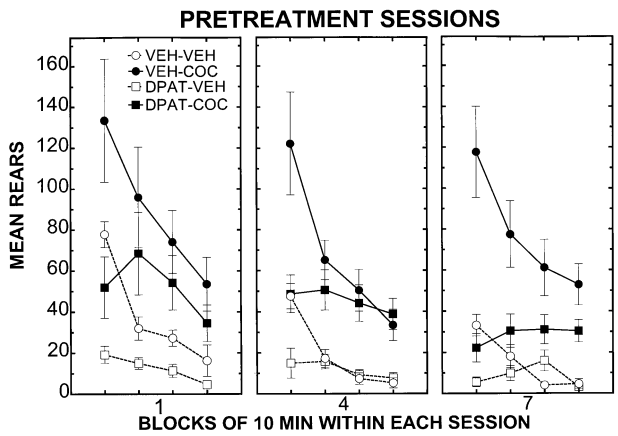

Fig. 3. Mean ± S.E.M. rears across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 15.0 mg/kg cocaine (COC).

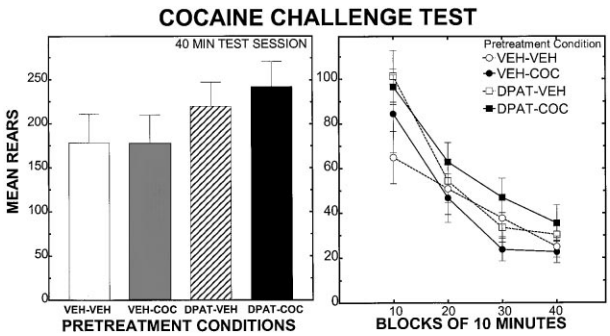

Fig. 4. Mean ± S.E.M. rears after a cocaine challenge injection (10.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or cocaine (15.0 mg/kg, COC). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

3.1.4. Sensitization test — rears

The results of the cocaine challenge treatment on rearing are shown in Fig. 4. As may be seen in the left panel, neither 7-OH-DPAT nor cocaine pretreatments significantly affected the overall number of rears after the cocaine challenge injection. However, as shown in the right panel, these treatments did appear to increase cocaine-induced rearing on the first block of this test session [7-OH-DPAT × Cocaine × Block interaction, $F(3,84) = 2.89$, $P < .05$]. Analysis of this latter interaction indicated that the two groups of 7-OH-DPAT-pretreated rats displayed significantly more rearing than the vehicle-pretreated rats on Block 1 of this session [P's < .05]. However, the rearing behavior of the two 7-OH-DPAT groups did not significantly differ from that of the cocaine-pretreated rats [P's > .05]. No significant differences in rearing were observed among the groups on Blocks 2, 3, and 4 [P's > .05]. Thus, consistent with the results of the training sessions, repeated cocaine treatments did not result in the development of sensitization to rearing. Although 7-OH-DPAT treatments suppressed rearing during the training sessions, these treatments did not decrease subsequent sensitivity to cocaine using rearing as a behavioral measure.

3.1.5. Training sessions — stereotypy score

The effects of repeated 7-OH-DPAT and cocaine treatments on stereotypic movements are shown in Fig. 5. It should be noted that this measure of stereotypy can not distinguish among various normal head and limb movements and traditional measures of drug-induced stereotypy, such as sniffing, licking, head-bobbing, etc. Thus, it primarily measures drug-induced changes in the total number of small movements per unit time. As may be seen, cocaine significantly stimulated such movements compared to vehicle control rats across all sessions and blocks [cocaine effect, $F(1,28) = 322.85$, $P < .0001$], but this increase did not change across sessions. 7-OH-DPAT significantly decreased stereotypy scores, but this inhibition decreased across days as the stereotypy scores of the vehicle control

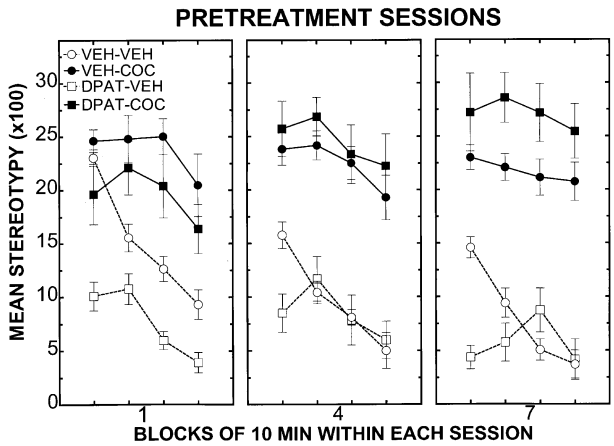

Fig. 5. Mean ± S.E.M. stereotypy counts across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 15.0 mg/kg cocaine (COC).

rats significantly decreased, particularly on the latter blocks of each session [7-OH-DPAT × Session interaction, $F(6,168) = 5.35$, $P < .0001$; 7-OH-DPAT × Block interaction, $F(3,84) = 62.59$, $P < .0001$]. Although 7-OH-DPAT suppressed stereotypy scores overall for vehicle-treated rats, the effect of 7-OH-DPAT on cocaine-induced stereotypy scores appeared to change across blocks. Indeed, a separate analysis of Session 1 indicated that the rats treated with 7-OH-DPAT and cocaine did not differ from rats treated with only cocaine on any block (P's>.05). In contrast, on Session 7, rats treated with the combination of cocaine and 7-OH-DPAT displayed significantly higher stereotypy scores on Blocks 2, 3, and 4 than rats treated with cocaine alone (P's < .05). Thus, repeated 7-OH-DPAT treatments with cocaine enhanced the stimulating effects of cocaine on stereotypic movements.

3.1.6. Sensitization test — stereotypy scores

The results of the cocaine challenge injection on stereotypy counts are shown in Fig. 6. Consistent with the training session, repeated cocaine treatments did not produce behavioral sensitization to cocaine-induced stereotypy. Although concurrent 7-OH-DPAT treatments with cocaine appeared to increase cocaine-induced stereotypy compared to vehicle treatments, this increase was not significant [7-OH-DPAT × Cocaine interaction, $F(1,28) = 2.87$, $P > .05$; 7-OH-DPAT × Cocaine × Block, $F(3,84) = 0.77$]. Thus, although concurrent 7-OH-DPAT treatments increased cocaine-induced stereotypy scores during the training sessions, this pretreatment did not significantly affect subsequent sensitivity to cocaine-induced stereotypy.

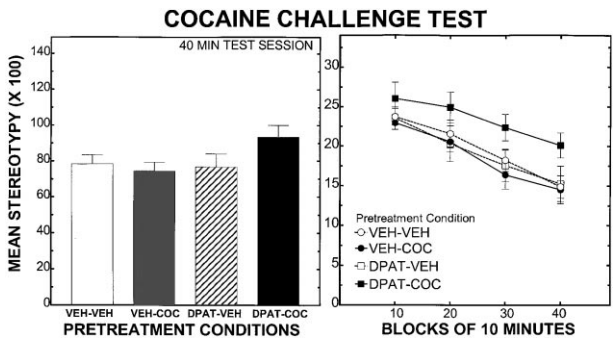

Fig. 6. Mean ± S.E.M. stereotypy counts after a cocaine challenge injection (10.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or cocaine (15.0 mg/kg, COC). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

3.2. Experiment 2: 7-OH-DPAT and apomorphine

3.2.1. Training sessions — distance traveled

The mean distance traveled by the four groups across the four 10-min blocks of Sessions 1, 4, and 7 are shown in Fig. 7. As may be seen in Fig. 7, apomorphine produced a progressively greater increase in activity across sessions [apomorphine effect, $F(1,28) = 66.54$, $P < .0001$; Apomorphine × Session interaction, $F(6,168) = 30.16$, $P < .0001$]. Consistent with Experiment 1, 7-OH-DPAT treatments produced a significant inhibition of activity compared to vehicle-treated rats on the first 10-min block of each session [7-OH-DPAT × Block interaction, $F(3,84) = 4.26$, $P = .0075$]. However, in contrast to Experiment 1, 7-OH-DPAT treatments did not significantly attenuate the stimulating effects of apomorphine. Indeed, a separate analysis of Session 7 revealed a significant main effect of apomorphine treatment [$F(1,28) = 79.22$, $P < .0001$] and a significant Apomorphine × Block interaction [$F(3,84) = 28.16$, $P < .0001$]. However, none of the effects involving 7-OH-DPAT as a factor were significant. Thus, concurrent treatments with 7-OH-DPAT did not affect either the locomotor-stimulating effects of apomorphine or the development of behavioral sensitization to apomorphine.

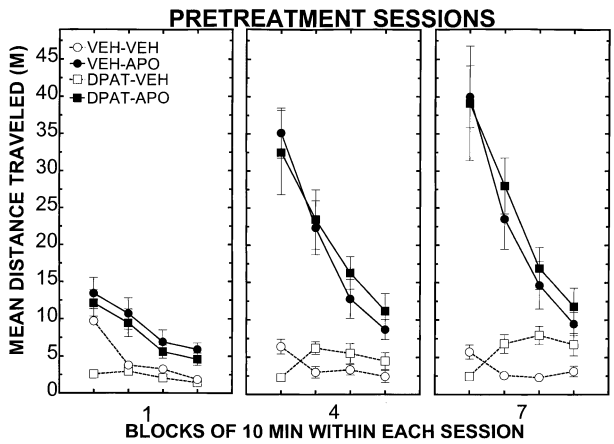

Fig. 7. Mean ± S.E.M. distance traveled across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 1.0 mg/kg apomorphine (APO).

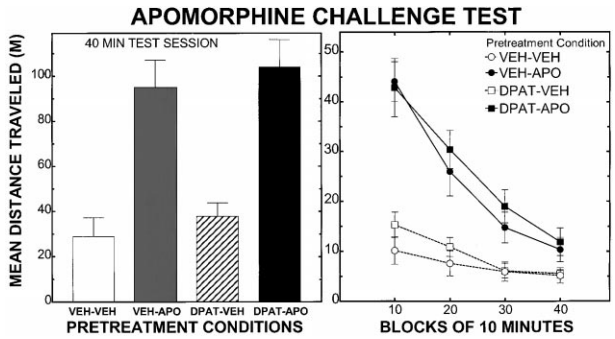

Fig. 8. Mean ± S.E.M. distance traveled after an apomorphine challenge injection (1.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or apomorphine (1.0 mg/kg, APO). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

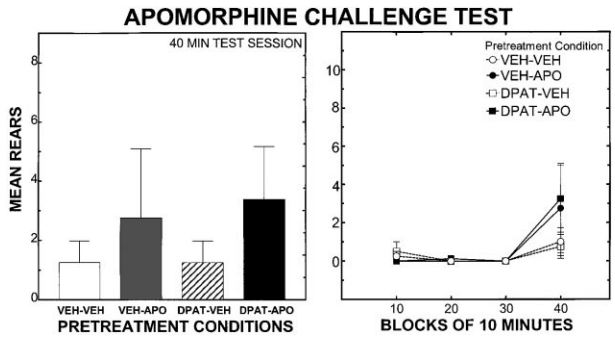

Fig. 10. Mean ± S.E.M. rears after an apomorphine challenge injection (1.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or apomorphine (1.0 mg/kg, APO). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

3.2.2. Sensitization test — distance traveled

The results of the apomorphine challenge test on distance traveled are shown in Fig. 8. As may be seen in the left panel of Fig. 8, rats previously treated with apomorphine with or without 7-OH-DPAT were significantly and equally more active after the apomorphine challenge injection than rats previously treated with only vehicle [apomorphine effect, $F(1,28) = 4.22$, $P < .0001$]. As shown in the right panel, this sensitization effect was greatest on the first block of the test session [block effect, $F(3,84) = 67.67$, $P < .0001$, and Apomorphine × Block interaction, $F(3,84) = 25.83$, $P < .0001$]. Prior treatment with 7-OH-DPAT did not significantly affect subsequent sensitivity to apomorphine for rats also pretreated with either vehicle or apomorphine. Thus, consistent with the results of the training sessions, concurrent treatments with 7-OH-DPAT did not affect the development of behavioral sensitization to apomorphine using horizontal locomotor activity as a behavioral measure.

3.2.3. Training sessions — rears

The effect of repeated 7-OH-DPAT and/or apomorphine treatments on rearing behavior is shown in Fig. 9. In contrast to the stimulating effects of cocaine observed in Experiment 1, apomorphine treatments almost completely abolished rearing behavior across for both sessions and blocks [apomorphine effect, $F(1,28) = 99.89$, $P < .0001$]. Consistent with Experiment 1, treatment with 7-OH-DPAT also decreased rearing compared to vehicle rats, but this 7-OH-DPAT-induced inhibition of rearing decreased across sessions and block [7-OH-DPAT × Block interaction, $F(3,84) = 32.97$, $P < .0001$; 7-OH-DPAT × Session interaction, $F(6,168) = 4.79$, $P < .001$]. Although probably due a floor effect, 7-OH-DPAT did not decrease rearing in apomorphine-treated rats [7-OH-DPAT × Apomorphine × Block interaction, $F(3,84) = 33.47$, $P < .0001$, and 7-OH-DPAT × Apomorphine × Session interaction, $F(6,164) = 4.79$, $P < .001$].

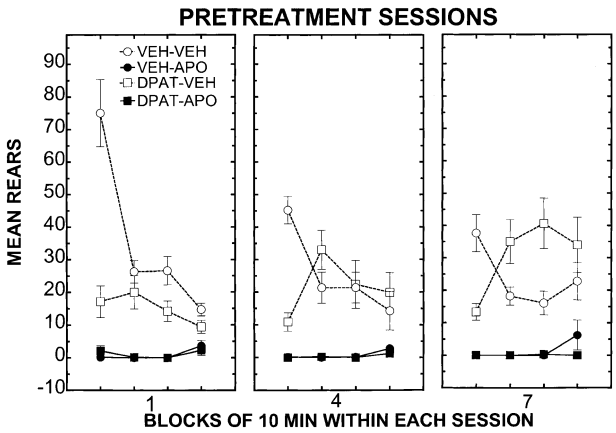

Fig. 9. Mean ± S.E.M. rears across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 1.0 mg/kg apomorphine (APO).

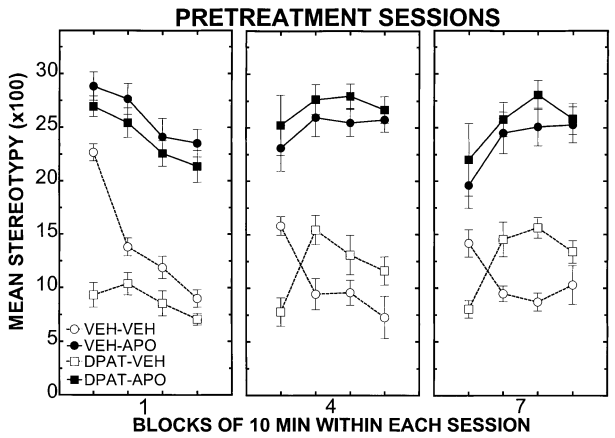

Fig. 11. Mean ± S.E.M. stereotypy counts across blocks of 10 min on Sessions 1, 4, and 7 for rats treated before each session with either vehicle (VEH) or 0.05 mg/kg 7-OH-DPAT (DPAT) and either vehicle or 15.0 mg/kg apomorphine (APO).

3.2.4. Sensitization test — rears

The results of the apomorphine challenge injection on rearing are shown in Fig. 10. As may be seen, the apomorphine challenge injection greatly suppressed rearing behavior in all pretreatment groups. Consequently, none of the effects involving either apomorphine or 7-OH-DPAT as a factor were significant.

3.2.5. Training sessions — stereotypy counts

The effects of repeated 7-OH-DPAT and apomorphine treatments on stereotypic counts on Sessions 1, 4, and 7 are displayed in Fig. 11. As may be seen, apomorphine treatments significantly increased stereotypy counts [apomorphine effect, $F(1,28)=229.22$, $P<.0001$]. Although this apomorphine-induced stimulation of stereotypy appeared to decrease across sessions, particularly on Block 1, neither the Apomorphine × Session interaction nor the Apomorphine × Session × Block interaction approached significance. Consistent with Experiment 1, 7-OH-DPAT treatments significantly decreased stereotypy on the first session compared to vehicle rats, but this inhibition decreased across both sessions and blocks [7-OH-DPAT × Session interaction, $F(6,168)=8.64$, $P<.0001$, and 7-OH-DPAT × Block interaction, $F(3,84)=12.91$, $P<.0001$]. However, concurrent 7-OH-DPAT treatments did not significantly affect stereotypy counts in apomorphine-treated rats on any block [7-OH-DPAT × Apomorphine × Block interaction, $F(3,84)=13.04$, $P<.0001$]. These findings contrast with those from Experiment 1 that indicated that repeated 7-OH-DPAT treatments with cocaine significantly increased stereotypy counts compared to cocaine alone.

3.2.6. Sensitization test — stereotypy counts

The results of the apomorphine challenge test are shown in Fig. 12. Overall, as shown in the left panel, the various pretreatment combinations did not significantly affect apomorphine-induced stereotypy counts. However, as shown in the right panel, prior treatments with apomorphine with or without 7-OH-DPAT did attenuate apomorphine-induced stereotypy counts on the first block of this test session [Apomorphine × Block interaction, $F(3,84)=13.75$, $P<.0001$]. This finding is consistent with the tendency observed during the training sessions of declining stereotypy counts with repeated apomorphine treatments. This finding suggests that the stereotypy-inducing effects of apomorphine may undergo some tolerance with repeated apomorphine administration. 7-OH-DPAT treatments, however, did not influence this effect.

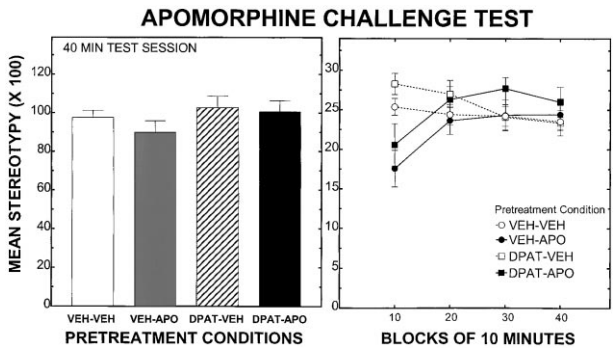

Fig. 12. Mean ± S.E.M. stereotypy counts after a cocaine challenge injection (10.0 mg/kg) for rats previously treated subchronically with either vehicle (VEH) or 7-OH-DPAT (0.05 mg/kg, DPAT) and either vehicle or apomorphine (1.0 mg/kg, APO). The left panel represents the total session activity, and the right panel presents the same data as a function of four 10-min blocks within the session.

4. Discussion

Consistent with previous research, the low dose of 7-OH-DPAT used in the present study significantly suppressed both horizontal (distance traveled) and vertical (rearing) activity, as well as stereotypic small movements (Geter-Douglas et al., 1997; Mattingly et al., 1996). Furthermore, repeated treatments with 7-OH-DPAT alone did not induce behavioral sensitization and did not increase subsequent behavioral sensitivity to either apomorphine or cocaine (Mattingly et al., 1996). In contrast, cocaine acutely stimulated all measures of motor behavior and resulted in behavioral sensitization to the horizontal locomotor-activating effects of cocaine with repeated treatment. Like cocaine, apomorphine stimulated horizontal locomotion and stereotypy, and produced behavioral sensitization with repeated treatment (Mattingly et al., 1988a,b). However, unlike cocaine, apomorphine nearly completely abolished rearing behavior. The differential effect of apomorphine, cocaine, and 7-OH-DPAT on rearing behavior suggests that cocaine-induced increases in rearing may not be mediated by dopaminergic receptor stimulation. Indeed, dopamine receptor stimulation alone appears to directly inhibit this behavior.

When combined with cocaine, 7-OH-DPAT significantly attenuated cocaine-induced increases in horizontal locomotion and rearing. Although 7-OH-DPAT initially suppressed cocaine-induced increases in stereotypy, concurrent 7-OH-DPAT treatments produced an enhancement of cocaine-induced stereotypy with repeated treatments (cf. Fig. 11). Consistent with these findings, the D_3-preferring receptor agonists, PD128907 and quinpirole, have been reported to attenuate the locomotor-activating effects of amphetamine (DeBoer et al., 1997; Furmidge et al., 1991), and the D_3-preferring receptor antagonist, U99194A, has been reported to enhance the stimulating effects of amphetamine (Waters et al., 1993). The effects of these dopamine D_3-preferring ligands on amphetamine-induced hyperactivity have been attributed to a preferential effect on dopamine autoreceptors (e.g., Furmidge et al., 1991). It has been assumed, for example, that concurrent stimulation of dopamine autoreceptors may attenuate amphetamine-induced hyperactivity by reducing the stimulant-induced increase in extracellular dopamine (e.g., Furmidge et al., 1991). However, as noted previously, it has also been argued that the locomotor

suppressive effects of D_3-preferring agonists are mediated exclusively by stimulation of postsynaptic D_3 receptors, which unlike D_1- and D_2-type postsynaptic receptors, are inhibitory with respect to locomotor activity (Svensson et al., 1994a,b; Waters et al.). Either of these mechanisms could account for the inhibitory effect of 7-OH-DPAT on acute cocaine-induced hyperactivity in the present study.

In contrast to the inhibitory effect of 7-OH-DPAT on cocaine-induced hyperactivity, concurrent 7-OH-DPAT treatments did not significantly affect apomorphine-induced changes in horizontal locomotion, stereotypy, or rearing. This differential effect of 7-OH-DPAT treatments on cocaine- and apomorphine-induced hyperactivity may be related to their different mechanisms of action. As noted previously, unlike cocaine, high doses of apomorphine are thought to increase locomotor activity by directly increasing stimulation of dopamine D_1- and D_2-type postsynaptic receptors independent of changes in synaptic dopamine levels. If so, then an autoreceptor-mediated reduction in dopamine release resulting from 7-OH-DPAT treatments may not be expected to affect apomorphine-induced increases in postsynaptic receptor stimulation (Khroyan et al., 1999), and therefore, locomotor activity. On the other hand, it has been hypothesized that concurrent 7-OH-DPAT treatments may suppress various behavioral effects of apomorphine by increasing the ratio of postsynaptic D_3 receptor stimulation to D_1 and/or D_2 receptor stimulation (Khroyan et al., 1999). Although 7-OH-DPAT should have increased D_3 postsynaptic receptor stimulation in the current study, this did not reduce the locomotor-activating effects of apomorphine. Thus, taken together, the suppressive effect of 7-OH-DPAT on cocaine-induced but not apomorphine-induced hyperactivity appears more consistent with increased autoreceptor stimulation rather than postsynaptic D_3 receptor stimulation. However, additional neurochemical data are clearly needed to help clarify these findings. For example, whether systemic treatments with low doses of 7-OH-DPAT actually reduce cocaine-induced increases in extracellular dopamine has apparently not been determined.

It should be noted that these findings contrast with those of a previous study (Khroyan et al., 1999), which reported some different effects of 7-OH-DPAT on apomorphine- and cocaine-induced activity and stereotypy. Although the exact reasons for this discrepancy are not clear, there were a number of methodological differences between the studies. For example, in the previous study (Khroyan et al., 1999), concurrent 7-OH-DPAT treatments did not attenuate cocaine-induced increases in horizontal locomotor activity. However, in contrast to the present study, a higher dose of 7-OH-DPAT was administered intraperitoneally with cocaine that did not suppress horizontal locomotor activity when administered alone (Khroyan et al., 1999). Moreover, in the current study, 7-OH-DPAT had its greatest effect on cocaine-induced activity within the first 10-min block. In the previous study, only total activity over a 40-min period was reported (Khroyan et al., 1999). Any of these, as well as other differences between the two studies, may be responsible for the discrepant findings.

As discussed previously, concurrent treatments with low doses of 7-OH-DPAT have been reported to attenuate the development of CPP to cocaine, amphetamine, and morphine (DeFonseca et al., 1995; Khroyan et al., 1998, 1999). In contrast, concurrent 7-OH-DPAT treatments do not attenuate the development of CPP to apomorphine (Khroyan et al., 1999). These findings seem congruent with the differential acute effects of 7-OH-DPAT on cocaine- and apomorphine-induced horizontal locomotor activity in the present study. That is, 7-OH-DPAT attenuated cocaine-induced but not apomorphine-induced increases in activity. Consistent with the current study, the 7-OH-DPAT attenuation of CPP was attributed to the stimulation of dopamine autoreceptors by 7-OH-DPAT, resulting in an attenuation of drug-induced increases in the synaptic levels of dopamine (Khroyan et al., 1999). Taken together with the current results, these findings suggest that the locomotor-activating and rewarding effects of cocaine and apomorphine may be mediated by similar mechanisms.

Although concurrent treatments with 7-OH-DPAT attenuated the cocaine-induced increase in horizontal activity during the pretreatment sessions, this concurrent treatment did not attenuate the development of behavioral sensitization to cocaine. Indeed, rats previously treated with both 7-OH-DPAT and cocaine displayed a more prolonged increase in horizontal locomotor activity after the cocaine challenge injection than rats previously treated with only cocaine. Thus, if anything, 7-OH-DPAT enhanced the development of behavioral sensitization to cocaine. In contrast, 7-OH-DPAT did not affect the development of behavioral sensitization to apomorphine.

At present, the mechanism responsible for this 7-OH-DPAT-induced enhancement in the development of behavioral sensitization to cocaine is not clear. One possible explanation is that concurrent 7-OH-DPAT treatments with cocaine enhanced the development of autoreceptor subsensitivity. Autoreceptor subsensitivity has been proposed as an initial event in the cellular cascade associated with the development of behavioral sensitization to psychomotor stimulants (see Henry et al., 1989; Henry and White, 1995; Wolf et al., 1993). According to this hypothesis, sensitization may involve at least two cellular alterations in the mesocorticolimbic dopamine system: an initial and transient decrease in the sensitivity of D_2-type autoreceptors followed by a long-lasting increase in the sensitivity of D_1-type postsynaptic receptors. Thus, the 7-OH-DPAT enhancement of cocaine sensitization may be due to a greater decrease in autoreceptor sensitivity. In a previous study (Mattingly et al., 1996), however, basal dopamine synthesis was not affected by repeated 7-OH-DPAT treatments, suggesting that synthesis-modulating autoreceptors do not become subsensitive with repeated 7-OH-DPAT

treatments (Mattingly et al., 1996). Of course, the regulation of synthesis-modulating autoreceptors may differ from impulse- or release-modulating autoreceptors (Rowlett et al., 1997), and acute 7-OH-DPAT treatments have been reported to have a greater effect on release-modulating than synthesis-modulating autoreceptors (Gainetdinov et al., 1996). Although repeated apomorphine treatments also result in the development of autoreceptor subsensitivity (Rowlett et al., 1991, 1997), as noted before, the expression of sensitization to apomorphine may be less affected by changes in dopamine release than indirect dopamine agonists, such as cocaine.

In conclusion, the current findings indicate that concurrent treatment with the putative dopamine D_3-preferring receptor agonist 7-OH-DPAT has differential effects on the acute and chronic effects of the indirect dopamine agonist, cocaine, and the direct dopamine agonist, apomorphine. Although 7-OH-DPAT acutely attenuated the locomotor-activating effects of cocaine, concurrent treatments with 7-OH-DPAT and cocaine enhanced the development of behavioral sensitization. In contrast, concurrent treatments with 7-OH-DPAT did not affect either the acute motor effects of apomorphine or the development of behavioral sensitization to apomorphine. Although more research is needed, the differential effects 7-OH-DPAT on the acute and chronic effects of cocaine and apomorphine appear more consistent with an explanation based upon dopamine autoreceptor stimulation than postsynaptic D_3 receptor stimulation. Although the dose of 7-OH-DPAT used in the present study is thought to be selective for dopamine D_3 receptors (Gainetdinov et al., 1996; Levant et al., 1996), whether or not the current findings with 7-OH-DPAT are mediated exclusively by D_3 receptor stimulation is not clear. As noted previously, based upon research with D_3 ''knock-out'' mice, it has been suggested that D_3 receptors do not function as autoreceptors (Koeltzow et al., 1998). Although evidence from these genetically altered mice must be interpreted with caution due to potential developmental compensations (Shafer and Levant, 1998), if accurate, the current findings with 7-OH-DPAT may involve dopamine D_2 rather than D_3 autoreceptors. Finally, to the extent that behavioral sensitization is a valid model of drug craving, the current results suggests that concurrent treatments with 7-OH-DPAT may enhance rather than attenuate the development of craving. On the other hand, since 7-OH-DPAT treatment alone did not increase subsequent behavioral sensitivity to either cocaine or apomorphine, it is possible that 7-OH-DPAT treatments during drug withdrawal will not maintain or enhance drug craving.

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