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8-OHDPAT effects upon cocaine unconditioned and conditioned behaviors A role for drug stimulus effects

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Abstract

The effects of the 5-HT_{1A} agonist, (±)-8-hydroxy-dipropylaminotetralin (8-OHDPAT) upon the unconditioned and conditioned behavior induced by cocaine were assessed in rats. Separate groups ($n=7$) received saline, cocaine (10 mg/kg), 8-OHDPAT (0.2 mg/kg), or 8-OHDPAT (0.2 mg/kg) plus cocaine (10 mg/kg) for eight treatment sessions (two per week) in which the rats were tested for 20 min in an open-field. On the eighth treatment session, cocaine enhanced locomotion and rearing but decreased grooming. 8-OHDPAT also decreased grooming and, when given in combination with cocaine, enhanced locomotion but attenuated cocaine-induced rearing. The two 8-OHDPAT groups differed substantially from each other and from the cocaine group in terms of locomotion during the drug treatment phase. Subsequently, all groups received a series of conditioning tests in which they received saline, 0.1, 0.2, or 0.4 mg/kg 8-OHDPAT prior to testing. Groups which had received either 8-OHDPAT or cocaine prior to the conditioning tests exhibited equivalent conditioned effects on the saline conditioning test. When conditioning tests were conducted with 8-OHDPAT, however, only the group which had previously received the combined 0.2 mg/kg 8-OHDPAT plus cocaine treatment exhibited a conditioned response and this effect only occurred at the 0.2 8-OHDPAT dose level. These observations indicate the important influence of the stimulus properties of drugs for the study of drug conditioning and for understanding drug interactions with cocaine. Published by Elsevier Science Inc.

Keywords: Cocaine; Conditioning; Open-field; 8-OHDPAT; Locomotion

1. Introduction

Following the initial observations of Pavlov (1927), it has been well established that a variety of physiological and behavioral effects of drugs can become conditioned to drug-associated situational cues. Although the formulation of situational cues as conditioned stimuli (CS) and drug treatments as unconditioned stimuli (US) readily falls into a Pavlovian conditioning framework, the conditioned response (CR) has sometimes been problematic. A detailed analysis (Eikelboom and Stewart, 1982) of the drug unconditioned response (UR) in terms of central effects induced by a drug US has indicated that the drug UR can, in some cases, be a central compensatory response to the peripheral effects of the drug US. This functional distinction between a drug US, which involves central mechanisms directly, and a drug US, which involves central compensatory mechanisms, appears to have clarified instances where the CR is opposite to the observed drug effect UR. With this reformulation of the drug US, drug conditioning with respect to what is conditioned is in accord with Pavlov's (1928) stimulus substitution theory of conditioning.

For many years, Pavlovian conditioning of drug effects was focused exclusively upon drug-induced autonomic system responses (Broadbent and Cunningham, 1996; Eikelboom and Stewart, 1982; Pavlov, 1927; Schwarz-Stevens and Cunningham, 1993). With the emergence of behavioral pharmacology, the behavioral domain of Pavlovian drug conditioning has been expanded to include drug-induced hormonal (De Vries et al., 1998), motoric and reward effects (Carey and Damianopoulos, 1994; Pickens and Dougherty, 1971; Stewart and Eikelboom, 1987). One of the earliest reports of the conditioning of drug-induced locomotor stimulation was that of amphetamine-induced hyperlocomotion (Tilson and Rech, 1973). Many subsequent reports have confirmed this basic observation and extended it to other psychostimulant drugs (Barr et al.,

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1983; Beninger and Hahn, 1983; Herz and Beninger, 1987; Stewart and Druhan, 1993). Drug conditioning is now well recognized to be of importance in the treatment of drugs of abuse such as cocaine in that clinical studies (Childress et al., 1988; Ehrman et al., 1992; Newlin, 1992) have shown that stimuli associated with drug taking can act as CS and evoke drug-like effects. In addition to functioning as US, drugs can also acquire CS properties.

The use of drugs as CS is well established in a variety of classical conditioning paradigms (Carey, 1989, 1991; Greeley et al., 1984; Jarbe et al., 1981; Lal and Bennet, 1989; Martin, 1983; Revusky, 1985; Revusky and Reilly, 1990a,b; Siegel, 1977, 1988). One approach has involved the use of drugs such as pentobarbital (Revusky, 1985) as a CS for lithium (Li)-induced taste aversion. In addition, drugs used as a CS have been documented in classical conditioning paradigms involving CR responses ranging from hypothermia (Taukulis, 1986), anxiogenic responses (Taukulis, 1996) to morphine withdrawal (Siegel, 1988). These studies have validated the efficacy of the stimulus properties of drugs to be able to function as CS in classical conditioning paradigms where the UR is induced by another drug. We have been able to show that drug–drug interactions mediated by Pavlovian conditioning extend to motoric behavior elicited by psychomotor stimulant drugs. Using the rotation behavior model, we found that the cue properties of sodium pentobarbital or scopolamine (Carey, 1989, 1991) could acquire the rotation response properties of apomorphine. After pentabarbitol or scopolamine was paired with an apomorphine treatment which induced rotation behavior, each of these two drugs acquired the capacity to elicit rotation when given alone without apomorphine. This effect was not observed in animals given the same drug exposure but unpaired to apomorphine. Subsequently, we have demonstrated (Carey et al., 1999) in an intact animal model that drugs as diverse as the 5-HT_{1A} partial agonist, buspirone and the *N*-methyl-D-aspartate (NMDA) noncompetitive antagonist, dizocilpine (MK-801) can acquire CS properties for cocaine. In the present study, we extend the investigation of conditioned drug interactions with cocaine to include a drug treatment that not only provides drug stimuli but also alters the UR effects of cocaine. In this study, we use the 5-HT_{1A} agonist, (±)-8-hydroxy-dipropylaminotetralin (8-OHDPAT), which has well-established discriminative stimulus effects (Glennon, 1986; Schreiber and De Vry, 1993; Schreiber et al., 1995) and can also modify the locomotor stimulant effect of cocaine (Carey et al., 2001; De La Garza and Cunningham, 2000). Interestingly, 8-OHDPAT enhances cocaine-induced horizontal hyperactivity but decreases vertical hyperactivity. In this study, we undertook to determine whether these 8-OHDPAT induced alterations in the cocaine UR became expressed as conditioned drug responses in conditioning tests and, in addition, whether the cocaine stimulant effects may become conditioned to the 8-OHDPAT discriminative stimulus cues.

2. Method

2.1. Animals

Naive male Sprague–Dawley rats from Taconic Farms (Germantown, NY), 4 months old and weighing approximately 400 g at the start of the experiments were used. Upon arrival, the animals were housed in individual $48 \times 27 \times 20$ cm clear polycarbonate cages in a climate-controlled room at 22–24 °C with a 12-h dark/light cycle. During the first week after arrival, all animals were handled and weighed daily for 7 days. During the second week, the animals received three injections (ip) of 0.9% saline (1.0 ml/kg) in order to acclimate the animals to the injection procedure. All experiments occurred during the 12-h light cycle (6:00 a.m.–6:00 p.m.).

2.2. Drugs

Cocaine hydrochloride (Sigma, St. Louis, MO) was dissolved in sterile distilled H_2O to a concentration of 10 mg/ml. 8-OH DPAT (RBI/Sigma, Natick, MA) was dissolved in sterile distilled H_2O to a concentration of 0.1, 0.2, or 0.4 mg/ml. All injections were administered intraperitoneally.

2.3. Apparatus

All of the behavioral tests were conducted in square open-field compartments which were $60 \times 60 \times 45$ cm. Closed-circuit video cameras (RCA TC7011U) were mounted 50 cm above the open-field enclosures. All signals were analyzed by a video tracking system, the Videomex-V from Columbus Instruments (Columbus, OH), and the data were imported into a PC compatible computer. The walls of the chamber were white and the floor of the open-field was covered by plain white paper which was changed after each animal. Masking noise (80 dB) was provided by a white noise generator (San Diego Instruments, San Diego, CA) and was turned on immediately prior to placement of the animal in the test chamber and turned off upon removal from the test chamber. Testing was conducted under conditions of red light illumination to avoid the aversive quality of white light and to enhance the contrast between the subject and background as well as to reduce the animal's shadow. The animal's head was blackened with a nontoxic marker and the camera only tracked this feature of the rat's body. During each session, data were collected every 2.5 min by the computer. Dot matrix printers (Epson FX-286e) were placed outside the test rooms and were connected to the image analyzers by a parallel cable and the computer screen tracings of the animal's movement were printed out every 2.5 min. The complete test procedure was conducted automatically without the presence of the experimenter in the test room. In addition, a VHS VCR was also connected to each camera to video tape selected sessions. The videotapes of the last cocaine treatment

session and all conditioning tests were scored for behaviors not detected by the image analyzer: rearing (the two front limbs raised off the floor) and grooming behavior (facial and flank grooming). Two experimenters uninformed of the drug treatments scored the videotapes for rearing and grooming.

2.4. Behavioral testing

Initially, all animals underwent 10 days of daily handling including 3 days of saline injections to acclimate the animals to manipulation and injection procedures. Next, all animals were given two 10-min tests in the test environment in order to form groups which were statistically equivalent with respect to the dependent variable of locomotion distance. Four days after the completion of the matching protocol, the four matched groups ($n=7$) received eight 20-min tests (two per week, spaced 3 or 4 days apart) in which spontaneous behavior was recorded. Prior to each test, all animals received two injections. The first was administered in the homecage 20 min prior to testing in the open-field and the second was administered immediately prior to placement in the open-field. The treatment groups were saline–saline, saline–cocaine (10 mg/kg), 8-OHDPAT (0.2 mg/kg)–saline, 8-OHDPAT (0.2 mg/kg)–cocaine (10 mg/kg). The first treatment specified in each pair was the one administered in the homecage and the second treatment was the one administered immediately prior to placement in the open-field. These treatment sessions served as the acquisition phase designed to establish a conditioned drug response to test environment cues and/or to the drug-generated cues which preceded and overlapped with the cocaine treatment (i.e., 0.2 mg/kg 8-OHDPAT). Subsequently, tests for conditioning were conducted. There was one conditioning test per week. In addition, each week the animals received their original (i.e., acquisition) drug treatments in order to maintain the CR. Subsequent conditioning tests were tests with 8-OHDPAT. The saline–saline test was designed to assess possible cocaine conditioning to test environment cues and the 8-OHDPAT tests were designed to assess possible cocaine conditioning to 8-OHDPAT drug cues. The 8-OHDPAT was administered in the homecage 20 min before testing and the second injection was saline which was administered immediately before testing. The animals received one of three dose levels of 8-OHDPAT (0.1, 0.2, or 0.4 mg/kg) spaced 1 week apart. The order of the 8-OHDPAT conditioning tests was 0.2, 0.4, and 0.1 mg/kg. Three days after each 8-OHDPAT conditioning test, the animals were given their acquisition treatments in which they received either saline–saline, saline–cocaine (10 mg/kg), 0.2 mg/kg 8-OHDPAT/saline, or 0.2 mg/kg 8-OHDPAT–cocaine (10 mg/kg). These treatments were administered in order to maintain conditioning. After the completion of the three 8-OHDPAT conditioning tests, the animals received a final conditioning test with 0.2 mg/kg 8-OHDPAT.

2.5. Statistical analyses

Two-way analysis of variance (ANOVA) was used to analyze the behavioral data to determine the group effects, repeated treatment effects, as well as the interaction between variables. Subsequently, to make more specific comparisons, one-way ANOVAs were used. In order to make specific group comparisons, post hoc Duncan's multiple range tests were performed. $P<.05$ was used as the criterion for statistical significance.

3. Results

In order to assess conditioned drug effects, it is necessary to identify the unconditioned drug response. In comparing the four treatment groups over the eight acquisition sessions, there was a statistically significant treatment effects upon locomotion distance [$F(3,24)=25.1$, $P<.001$]. The Treatment drug × Group interaction was not significant [$F(3,24)=1.3$, $P>.05$]. The treatment group means and S.E.M.s for locomotion distance in meters (m) per session over the eight sessions were 32.4 ± 2.1, 33.0 ± 2.2, 53.7 ± 2.3, and 81.9 ± 2.8 for the saline, 8-OHDPAT, cocaine, and 8-OHDPAT plus cocaine groups, respectively. A comparison of group means using Duncan's Multiple Range test indicated that the 8-OHDPAT plus cocaine group had a higher mean distance score than all other groups ($P<.01$) and that the cocaine group had higher locomotor distance scores ($P<.01$) than the saline and 8-OHDPAT groups which did not differ from each other ($P>.05$). In order to more completely characterize the unconditioned drug effect of cocaine and 8-OHDPAT, the locomotion distance, rearing and grooming scores are presented (Fig. 1) for the eighth treatment session. As can be seen in Fig. 1, the locomotor stimulant effects of cocaine were quite evident on the eighth treatment day. The group differences in locomotor distance were statistically significant [$F(3,24)=31.2$, $P<.001$] and group comparisons using Duncan's Multiple Range test indicated that the 8-OHDPAT plus cocaine group had statistically higher scores than all other groups ($P<.01$). Also, the cocaine group had statistically higher scores than the saline and 8-OHDPAT groups ($P<.01$) which did not differ from each other ($P>.05$). There was a statistically significant treatment effect on rearing behavior [$F(3,24)=3.2$, $P<.05$]. As can be seen in the middle panel of Fig. 1, the cocaine treatment enhanced rearing behavior. The 8-OHDPAT treatment, however, attenuated the effect of cocaine upon rearing activity. As is shown in the bottom panel of Fig. 1, grooming was suppressed by both cocaine and 8-OHDPAT [$F(3,24)=15.4$, $P<.001$].

The results of the conditioning tests in which groups were pretreated either with saline or 8-OHDPAT injections are summarized in Fig. 2 in a dose–response format. The top panel presents the locomotion distance scores, the

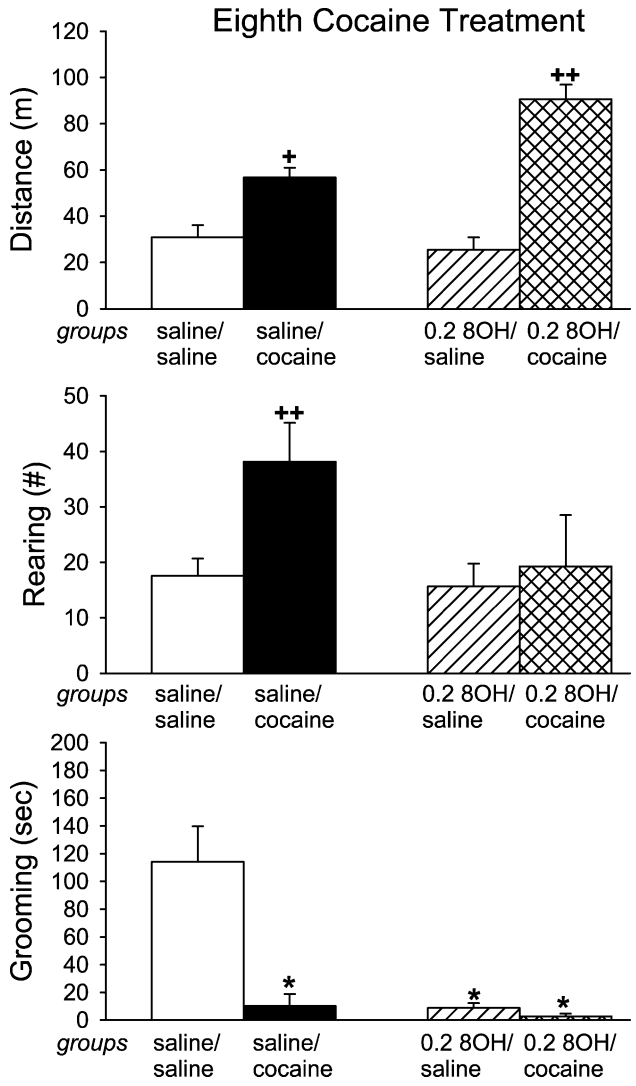

Fig. 1. Means and S.E.M.s for distance (top), rearing (middle), and grooming (bottom) on the (eighth) drug treatment session conducted 3 days prior to the start of conditioning tests. The four treatment groups (saline–saline, saline–10 mg/kg cocaine, 0.2 mg/kg 8-OHDPAT–saline, and 0.2 mg/kg 8-OHDPAT–10 mg/kg cocaine) received two injections. The first injection was administered in the homecage 20 min before testing and the second injection was administered immediately before testing. $^{++}$ Denotes scores higher than all other groups ($P<.05$). $^{+}$ Denotes scores higher than the two noncocaine groups ($P<.05$). * Denotes scores lower than the saline group ($P<.05$).

middle panel the rearing scores, and the bottom panel the grooming duration scores. The statistical analysis for each response measure indicated statistically significant dose effects [$F(3,24)=34.7$, $F(3,24)=33.5$, and $F(3,24)=64.3$, $P<.001$ for distance, rearing, and grooming, respectively]. Statistically significant group differences were only obtained for the distance measure [$F(3,24)=6.6$, $P<.01$]. However, there were statistically significant ($P<.05$) Group × Dose level interactions for the distance, rearing, and grooming response measures [$F(9,72)=2.5$, $F(9,72)=2.1$, and $F(9,72)=2.2$, respectively]. In order to identify the dose levels at which statistically significant group differences occurred, one-way ANOVAs were performed at each dose level. For distance, the zero or saline conditioning test was statistically significant [$F(3,24)=6.3$, $P<.01$]. Using Duncan's Multiple Range test to identify which group differences were significant, it was found that for the saline–cocaine, 0.2 mg/kg 8-OHDPAT–saline, and 0.2 mg/kg 8-OHDPAT–cocaine groups had higher distance scores than the saline–saline groups. There also were statistically significant group differences for the 0.2-mg/kg 8-OHDPAT conditioning test [$F(3,24)=7.9$, $P<.01$]. The differences evident at the 0.2-mg/kg 8-OHDPAT conditioning test were selective for the 0.2-mg/kg 8-OHDPAT–cocaine group. Individual group comparisons using Duncan's Multiple Range test indicated that the 0.2-mg/kg 8-OHDPAT–cocaine group had higher distance scores than all other groups ($P<.01$), which did not differ from each other ($P>.05$). None of the group differences obtained on the conditioning tests with 0.1 and 0.4 mg/kg 8-OHDPAT were statistically significant ($P>.05$). In assessing the results on

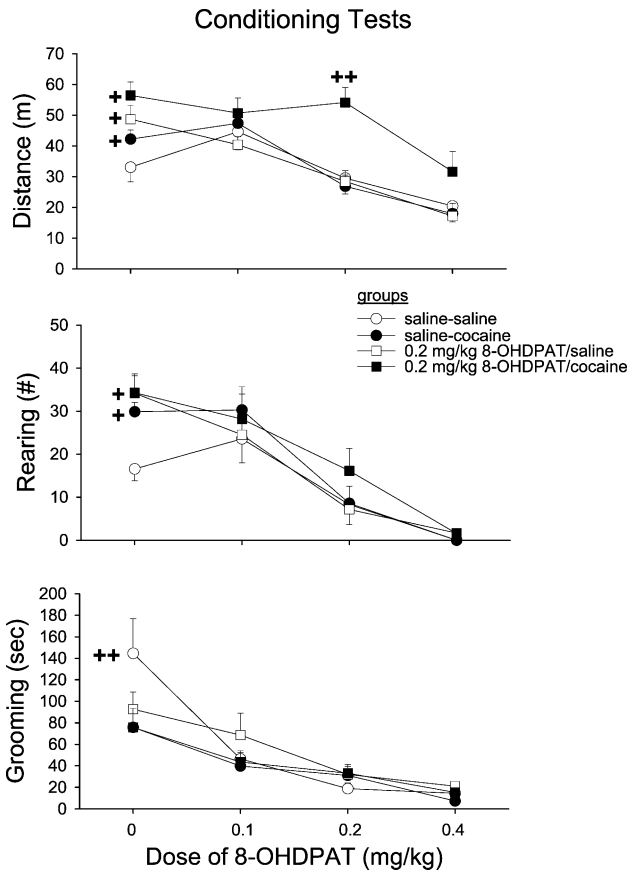

Fig. 2. Means and S.E.M.s for distance (top), rearing (middle), and grooming (bottom) scores obtained on four conditioning tests spaced 1 week apart in which all groups received pretreatments of either 0 (saline), 0.1 mg/kg 8-OHDPAT, 0.2 mg/kg 8-OHDPAT, or 0.4 mg/kg 8-OHDPAT 20 min prior to a 20-min test. Previously, the groups had received a series of saline–saline, saline–cocaine (10 mg/kg), 0.2 mg/kg 8-OHDPAT–saline, or 0.2 mg/kg 8-OHDPAT–cocaine (10 mg/kg) treatments. $^{+}$ Denotes $P<.05$ versus the saline–saline group. $^{++}$ Denotes $P<.05$ versus all other groups.

the conditioning tests for rearing (middle panel), only the saline test yielded a statistically significant effect [$F(3,24) = 5.8$, $P < .01$]). A comparison of the groups on the saline conditioning test using Duncan's Multiple Range test indicated that the cocaine, 8-OHDPAT, and the 8-OHDPAT plus cocaine groups all had higher rearing scores than the saline group. A generally similar statistical result was obtained for the grooming results in that the only one-way ANOVA which reached statistical significance was the saline conditioning test [$F(3,24) = 6.8$, $P < .01$]. Similar to the rearing results, the cocaine and the 8-OHDPAT groups were statistically different ($P < .05$) from the saline group.

In order to determine the reliability of the effect observed in the conditioning test with 0.2 mg/kg 8-OHDPAT, the groups received a second conditioning test with 0.2 mg/kg 8-OHDPAT. As was the case in Fig. 2, there was a statistically significant group difference in the 0.2-mg/kg 8-OHDPAT conditioning test for distance [$F(3,24) = 6.4$, $P < .01$]. Comparisons of group means using Duncan's Multiple Range test indicated that the 8-OHDPAT plus cocaine group had higher distance scores than all other groups ($P < .01$), which did not differ from each other ($P > .05$). There were no statistically significant group differences for rearing or grooming [$F(3,24) = 1.6$, $P > .05$ and $F(3,24) = 1.5$, $P > .05$, respectively]. Fig. 3 presents the result from this second conditioning test in which all groups were treated with 0.2 mg/kg 8-OHDPAT 20 min prior to testing and saline immediately before placement in the test environment.

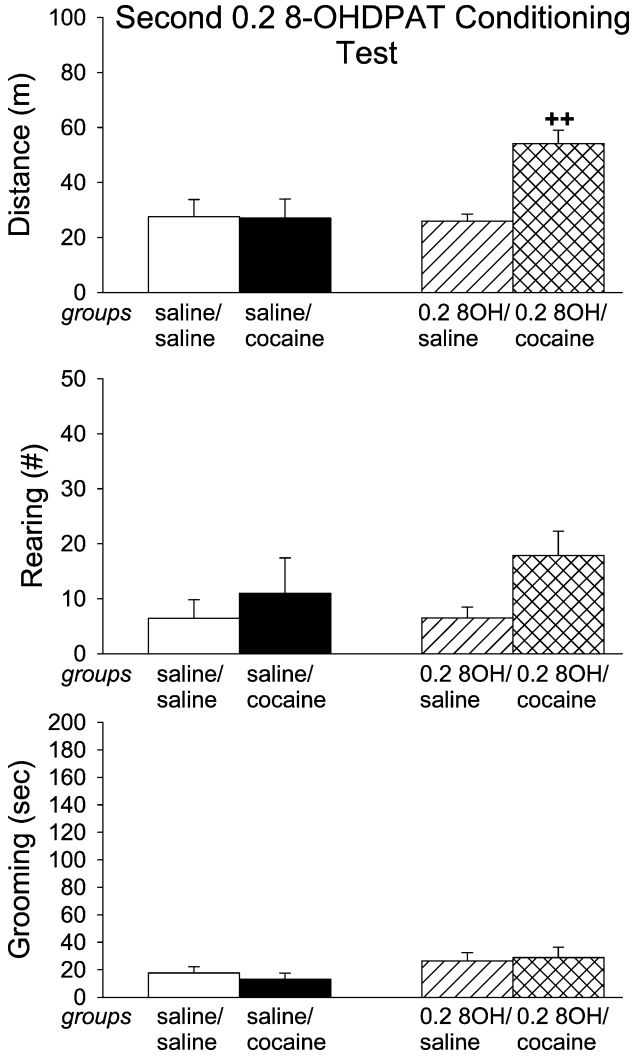

Fig. 3. Means and S.E.M.s for distance (top), rearing (middle), and grooming (bottom) on a repeat conditioning test in which all groups received 8-OHDPAT (0.2 mg/kg) 20 min before testing. All groups were given saline immediately prior to placement in the open-field. $^{++}$ Denotes scores higher than all other groups ($P < .05$).

4. Discussion

Cocaine induced the expected effect upon behavior by enhancing horizontal as well as vertical activity and reducing grooming behavior. In tests for conditioning, the cocaine-conditioned behavioral responses to test environment cues generally matched the cocaine-induced behavioral responses albeit to an attenuated extent. These findings are consistent with the literature (Damianopoulos and Carey, 1994) and agree with the Pavlovian conditioning principle that the CR be similar to the UR but of lesser magnitude.

Pretreatment of animals with the 5-HT_{1A} agonist, 8-OHDPAT substantially modified the unconditioned locomotor responses induced by cocaine. Given alone, the 8-OHDPAT did not affect locomotion distance and by the eighth treatment did not affect rearing. When given in combination with cocaine, however, 8-OHDPAT attenuated the cocaine-induced increase in rearing behavior and enhanced cocaine-induced horizontal locomotion. In that both 8-OHDPAT and cocaine suppressed grooming behavior, any possible additivity was obscured by a possible "floor" effect. 8-OHDPAT is a full 5-HT_{1A} agonist (Barrett et al., 1994; Callahan and Cunningham, 1997; Cornfield, 1991). While 8-OHDPAT can directly stimulate 5-HT_{1A} postsynaptic receptors, it also stimulates 5-HT_{1A} autoreceptors. As a consequence, the effects of 8-OHDPAT can be complex depending upon dose level. That is, lower doses may primarily activate autoreceptors and, thereby, inhibit 5-HT activity whereas higher doses by directly activating 5-HT_{1A} postsynaptic receptors can induce motoric syndromes associated with high levels of serotonergic activation (Sternbach, 1991). In the present study, the 0.2-mg/kg dose of 8-OHDPAT administered intraperitoneally did not modify locomotion but did suppress grooming behavior. When given in combination with cocaine, however, the 8-OHDPAT treatment enhanced horizontal locomotion. Possibly, the increase in horizontal locomotion in animals given the combined 8-OHDPAT plus cocaine treatment was simply a response reorganization effect in which an 8-OHDPAT suppression of rearing permitted more horizontal locomotion. Alternatively, the enhanced horizontal locomotion may represent the additive

effects of stimulation of the 5-HT_{1A} receptors induced directly by 8-OHDPAT and indirectly by increased 5-HT stimulation induced by cocaine (Ritz et al., 1990). The fact that the higher dose of 8-OHDPAT (0.4 mg/kg) did not increase horizontal locomotion, however, argues against this possibility.

The effects of 8-OHDPAT on unconditioned locomotion and rearing behavior, with or without cocaine, did not transfer into corresponding CRs. 8-OHDPAT-treated animals with or without cocaine exhibited equivalent horizontal and vertical activity in the conditioning tests and these activity levels were reliably higher than the saline treatment group. Surprisingly, the mean locomotion distance and rearing scores on the saline conditioning test were overall higher for the 8-OHDPAT–saline group than for the cocaine group. These effects of 8-OHDPAT on the saline conditioning test, however, were not consistent with the 8-OHDPAT unconditioned drug-induced response effects and, therefore, are not consistent with the basic tenet of Pavlovian conditioning either as conditioned drug responses or conditioned compensatory drug responses. One likely possibility is that the 8-OHDPAT treatments blocked habituation or that the habituation to the test environment was 8-OHDPAT state dependent. In either case, the net result would be for the test environment to be relatively more novel for the 8-OHDPAT treatment groups versus the saline group when tested under saline conditions. From this perspective, both 8-OHDPAT groups would be expected to exhibit increases in exploratory behavior relative to the habituated saline animals. In several previous studies (Carey and Gui, 1997; Damianopoulos and Carey, 1994), we have shown that the effects of cocaine in conditioning tests using open-field behavior are not readily explicable in terms of antihabituation effects.

While the saline tests in the present study were not consistent with conditioned drug behavior in the animals treated with 8-OHDPAT, evidence consistent with a cocaine-conditioned locomotor response was observed in animals which had received the combined 8-OHDPAT–cocaine treatment when the tests were conducted with 8-OHDPAT alone. First of all, this cocaine-conditioned effect associated with 8-OHDPAT was closely linked to the dose level of 8-OHDPAT used in the paired treatment with cocaine. Specifically, the cocaine effects conditioned to the 8-OHDPAT cues were only observed at the 0.2-mg/kg dose level which had been paired with cocaine. In addition to the dose selectivity in 8-OHDPAT functioning as a CS there was behavioral selectivity. That is, the 8-OHDPAT selectively enhanced horizontal activity when given in combination with cocaine and this was the only behavioral response which was reliably enhanced relative to other groups in the 8-OHDPAT dose–response conditioning tests.

The effects of the 8-OHDPAT treatment given alone or in combination with cocaine on unconditioned behavior are consistent with previous reports (De La Garza and Cunningham, 2000; Carey et al., 2001). To date, however, the impact of the 8-OHDPAT treatment upon conditioned drug effects has not been studied. In this initial evaluation of 8-OHDPAT effects upon conditioned drug effects, the substantial contribution of the stimulus properties of 8-OHDPAT (Schreiber and De Vry, 1993; Schreiber et al., 1995; Glennon, 1986) appeared to become manifested. That is, the 8-OHDPAT treatment response effects observed during the cocaine treatment phase were not consistent with the behavioral response patterns observed in the saline conditioning test. In the cocaine treatment phase, the 8-OHDPAT treatment given alone did not affect locomotion distance as compared to the saline group, whereas the 8-OHDPAT group given cocaine exhibited an increase in locomotion greater than the cocaine group. Consequently, the two 8-OHDPAT groups had highly disparate levels of locomotion during the drug treatment phase. Thus, the response effects generated by the 8-OHDPAT treatments during the drug treatment phase do not provide a basis to interpret the behavior observed in the saline conditioning test. On the other hand, the stimulus effects of 8-OHDPAT appear to provide a way to account for the locomotor behavior effects observed in the 8-OHDPAT groups both in the saline and 8-OHDPAT conditioning tests. A drug stimulus way to interpret these observations is to consider that the 8-OHDPAT treatments generated drug stimuli which combined with environmental cues to create a stimulus complex in which the behavior occurred. The removal of the drug cues during the saline conditioning test would result in the test environment being more novel for the 8-OHDPAT-treated animals than for the saline control group. As a consequence of this greater novelty, the 8-OHDPAT animals tested with saline would be expected to engage in more exploratory locomotor behaviors than the well-habituated saline group. When tested with the 8-OHDPAT cues, the 8-OHDPAT plus cocaine-treated animals would have the full complement of cocaine-associated cues restored. The restoration of cocaine-associated cues permits the occurrence of the cocaine-conditioned behavior in the group which had received cocaine in combination with 8-OHDPAT. This consideration of 8-OHDPAT as generating drug stimulus cues, which together with the test environment cues comprise the stimulus complex in which the animals behave, is also relevant to the behavior of the saline versus cocaine-treated animals in the 8-OHDPAT conditioning tests. In all tests with 8-OHDPAT, the behavioral responses of these two groups were essentially the same. If the 8-OHDPAT treatments were viewed simply as inducing response effects, then one might expect that the 8-OHDPAT interaction with the cocaine CR effects to maintain a cocaine CR differential. This latter result was not observed. The equivalence between saline and cocaine groups, however, is compatible with an 8-OHDPAT drug stimulus plus drug response effect in that the 8-OHDPAT drug stimulus would serve to create a new stimulus complex which would mask the cues to which the cocaine had been conditioned. As a consequence of this drug stimulus modification of the cocaine CS, the cocaine CR effect is eliminated. Therefore, the saline and cocaine groups

would be expected to exhibit equivalent response effects to the 8-OHDPAT treatments.

While the preponderance of drug behavior studies using open-field behavior are devoted to the UR effects of the drug treatment, the possible contribution of stimulus effects of the drug treatments is uncertain because the stimulus effects are not manifested in the behavioral responses. The incorporation of conditioning tests appear to provide an opportunity to assess the possible stimulus effects of the drug treatments by permitting the opportunity to detect effects on saline conditioning tests which are inconsistent with the behavioral effects obtained in the tests of unconditioned drug response effects. Since the stimulus effects of drug treatments typically occur at dose levels below the response effects (Carey et al., 1999; Zajaczkowski et al., 1996), even dose levels of drugs which are subthreshold for inducing locomotor response effects, along with dose levels of the same drug which suppress locomotion, may generate increased activity levels relative to saline-treated animals in a saline conditioning test. Seemingly, the removal of the drug stimulus would have an antihabituation effect as compared to the saline treatment group. The present study also suggests that the stimulus properties of drugs offer a novel approach to blocking cocaine CS effects and, therefore, may have relevance to the development of pharmacological treatments for blunting cocaine craving evoked by cocaine CS.

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References

Barr GA, Sharpless NS, Cooper S, Schiff SR, Paredes W, Bridger WH. Classical conditioning, decay and extinction of cocaine-induced hyperactivity and stereotypy. Life Sci 1983;33:1341–51.

Barrett JE, Zhang L, Gleeson S, Gamble EH. Anxiolytic and antidepressant mechanisms of 5-HT1A drugs in the pigeon: contributions form behavioral studies. Neurosci Biobehav Rev 1994;18:73–83.

Beninger RJ, Hahn BL. Pimozide blocks establishment but not expression of amphetamine-produced environment-specific conditioning. Science 1983;220:1304–6.

Broadbent J, Cunningham CL. Pavlovian conditioning of morphine hyperthermia: assessment of interstimulus interval and CS–US overlap. Psychopharmacology (Berlin) 1996;126:154–64.

Callahan PM, Cunningham KA. Modulation of the discriminative stimulus properties of cocaine: comparison of the effects of fluoxetine with 5-HT_{1A} and 5-HT_{1B} receptor agonists. Neuropharmacology 1997;36:373–81.

Carey RJ. Stimulant drugs as conditioned and unconditioned stimuli in a classical conditioning paradigm. Drug Dev Res 1989;16:305–15.

Carey RJ. Pavlovian conditioning between co-administered drugs: elicitation of an apomorphine-induced antiparkinsonian response by scopolamine. Psychopharmacology (Berlin) 1991;104:463–9.

Carey RJ, Damianopoulos EN. Conditioned cocaine induced hyperactivity: an association with increased medial prefrontal cortex serotonin. Behav Brain Res 1994;62:177–85.

Carey RJ, Gui J. A simple and reliable method for the positive identification of Pavlovian conditioned cocaine effects in open-field behavior. J Neurosci Methods 1997;73:1–8.

Carey RJ, Damianopoulos EN, DePalma G. Issues in the pharmacological modification of cocaine conditioning: evidence that the stimulus properties of drugs can interact with contextual cues to activate or inactivate cocaine conditioned stimuli. Behav Brain Res 1999;101:189–206.

Carey RJ, DePalma G, Damianopoulos EN. Cocaine and serotonin: a role for the 5-HT1A receptor site in the mediation of cocaine stimulant effects. Behav Brain Res 2001;126:127–33.

Childress AR, McLellan AT, Ehrman R, O'Brien CP. Classically conditioned responses in opioid and cocaine dependence: a role in relapse? NIDA Res Monogr Ser 1988;84:25–43.

Cornfield LJ. Intrinsic activity of enantiomers of 8-hydroxy-2-(di-*n*-propylamino) tetralin and tis analogs at 5-hydroxytryptamine 1A receptors that are negatively coupled to adenylate cyclase. Mol Pharmacol 1991;39:780–1.

Damianopoulos EN, Carey RJ. A new method to assess Pavlovian conditioning of psychostimulant drug effects. J Neurosci Methods 1994;53:7–17.

De La Garza R, Cunningham KA. The effects of the 5-hydroxytrypamine 1A agonist 8-hydroxy-2-(di-*n*-propylamino)tetralin on spontaneous activity, cocaine-induced hyperactivity and behavioral sensitization: a microanalysis of locomotor activity. J Pharmacol Exp Ther 2000;292:610–7.

De Vries AC, Taymans SE, Sundstrom JM, Pert A. Conditioned release of corticosterone by contextual stimuli associated with cocaine is mediated by corticotropin-releasing factor. Brain Res 1998;786:39–46.

Ehrman RN, Robbins SJ, Childress AR, O'Brien CP. Conditioned responses to cocaine-related stimuli in cocaine abuse patients. Psychopharmacology (Berlin) 1992;107:523–9.

Eikelboom R, Stewart J. Conditioning of drug-induced physiological responses. Psychol Rev 1982;89:507–28.

Glennon RA. Discriminative stimulus properties of the 5-HT1A agonist 8-hydroxy-2-di-*n*-propylaminotetralin (8-OH DAT). Pharmacol, Biochem Behav 1986;25:135–9.

Greeley J, Lee DA, Polos CX, Capel H. Alcohol is an effective cue in the conditional control of tolerance to alcohol. Psychopharmacology (Berlin) 1984;83:159–62.

Herz RS, Beninger RJ. Comparison of the ability of (+)-amphetamine and caffeine to produce environment-specific conditioning. Psychopharmacology (Berlin) 1987;92:365–70.

Jarbe TU, Sterner U, Hjerpe C. Conditioning of an interoceptive drug stimulus to different exteroceptive contexts. Psychopharmacology (Berlin) 1981;73:23–6.

Lal H, Bennet DS. Drugs as interoceptive stimuli. Drug Dev Res 1989;16:397–464.

Martin GE. The effects of pairings of pentobarbital with lithium chloride on the capacity of pentobarbital to maintain a conditioned aversion. Behav Neural Biol 1983;39:145–8.

Newlin DB. A comparison of drug conditioning and craving for alcohol and cocaine. In: Galanter M, editor. Recent developments in alcoholism. Alcohol and cocaine: similarities and differences, vol. 10. New York: Plenum, 1992. pp. 147–64.

Pavlov IP. Conditioned reflex London: Oxford University Press, 1927.

Pavlov IP. Lectures on conditioned reflexes, vol. 1. New York: International Publishers, 1928.

Pickens R, Dougherty J. Conditioning of the activity effects of drugs. In: Thompson T, Schuster C, editors. Stimulus properties of drugs. New York: Appleton-Century-Crofts, 1971. pp. 39–50.

Revusky S. Drug interactions measured through taste aversion procedures with an emphasis on medical implications. Ann NY Acad Sci 1985;443:250–71.

Revusky S, Reilly S. Dose effects on heart rate conditioning when pentobarbital is the CS and amphetamine is the US. Pharmacol, Biochem Behav 1990a;36:933–6.

Revusky S, Reilly S. When pentobarbital is the conditioned stimulus and amphetamine is the unconditioned stimulus, conditioning depends on the type of conditioned response. Behav Neurosci 1990b;104:693–703.

Ritz MC, Cone EJ, Kuhar MJ. Cocaine inhibition of ligand binding at dopamine, norepinephrine and serotonin transporters: a structure–activity study. Life Sci 1990;46:635–45.

Schreiber R, De Vry J. Studies on the neuronal circuits involved in the discriminative stimulus effects of 5-hydroxytryptamine1A receptor agonists in the rat. J Pharmacol Exp Ther 1993;265:572–9.

Schreiber R, Lefebvre de Ladonchamps B, Monneyron S, Millian MJ. A drug discrimination analysis of the actions of novel serotonin1A receptor ligands in the rat using the 5-HT1A receptor agonist, 8-hydroxy-2-(di-*n*-propylamino)tetralin. J Pharmacol Exp Ther 1995;275:822–31.

Schwarz-Stevens KS, Cunningham CL. Pavlovian conditioning of heart rate and body temperature with morphine: effects of CS duration. Behav Neurosci 1993;107:1039–48.

Siegel S. Morphine tolerance acquisition as an associative process. J Exp Psychol 1977;3:1–13.

Siegel S. State-dependent learning and morphine tolerance. Behav Neurosci 1988;10:228–32.

Sternbach MD. The serotonin syndrome. Am J Psychiatry 1991;148:705–13.

Stewart J, Druhan JP. Development of both conditioning and sensitization of the behavioral activating effects of amphetamine is blocked by the non-competitive NMDA receptor antagonist, MK-801. Psychopharmacology (Berlin) 1993;110:125–32.

Stewart J, Eikelboom R. Conditioned drug effects. New York: Plenum, 1987. pp. 1–57.

Taukulis HK. Conditioned hyperthermia in response to atropine associated with a hypothermic drug. Psychopharmacology (Berlin) 1986;90:327–31.

Taukulis HK. Pavlovian conditioning to a diazepam cue with yohimbine as the unconditioned stimulus. Neurobiol Learn Mem 1996;65:223–32.

Tilson HA, Rech RA. Conditioned drug effects and absence of tolerance to D-amphetamine induced motor activity. Pharmacol, Biochem Behav 1973;1:149–53.

Zajaczkowski W, Moryl E, Papp M. Discriminative stimulus effects of the NMDA receptor antagonists MK-801 and CGP 37849 in rats. Pharmacol, Biochem Behav 1996;55:163–8.