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Buspirone Fails to Affect the Discriminative Stimulus Effects of Cocaine

DARION RAPOZA[1]

Drug Abuse Research Center, Pritzker School of Medicine, The University of Chicago, Chicago, IL 60637

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RAPOZA, D. *Buspirone fails to affect the discriminative stimulus effects of cocaine.* PHARMACOL BIOCHEM BEHAV **45**(1) 179–183, 1993.—Rats were trained to discriminate 4.0 mg/kg cocaine from saline in a two-lever, food-reinforced drug discrimination paradigm. Cocaine (0.5–8.0 mg/kg, IP) produced a dose-related increase in cocaine-appropriate responding, with the training dose of 4.0 mg/kg being the lowest dose that met criterion (>90% cocaine-appropriate responding over the entire session) for substitution. Pretreatment with buspirone (2.0–16 mg/kg, IP) did not attenuate the discriminative stimulus properties of 4.0 mg/kg cocaine at doses up to those that caused complete suppression of responding (16 mg/kg, IP). In contrast, combinations of 0.12 mg/kg haloperidol with 4.0 mg/kg cocaine decreased cocaine-appropriate responding from 100 to 65% while suppressing response rate to 50% of the response rate seen with the 4.0-mg/kg dose of cocaine alone. Thus, behaviorally active doses of buspirone failed to attenuate the discriminative stimulus effects of cocaine in a sensitive behavioral paradigm.

Cocaine Dopamine Buspirone Haloperidol Drug discrimination Behavior Rat

EVIDENCE gathered using drug discrimination techniques has implicated dopamine as a neurotransmitter that may mediate the discriminative stimulus effects of cocaine. Indirect dopamine agonists have been found to substitute for cocaine (2–4,8–10,22), and dopamine antagonists partially antagonize the discriminative stimulus effects of cocaine (2,3,22). In contrast, a wide variety of nondopaminergic agonists (e.g., adrenergic, serotonergic, and cholinergic) do not substitute for cocaine (4,8,14,17,18,22), nor do compounds that are antagonists at these other receptor types block its discriminative stimulus effects (1,16,22).

Buspirone is an atypical anxiolytic that has been found to have properties in common with the dopamine antagonists. It binds to dopamine receptors (29) and, like the dopamine antagonist haloperidol, enhances firing of dopaminergic neurons in the substantia nigra and reverses inhibition of these neurons caused by apomorphine, amphetamine, or dopamine (23). Behaviorally, both buspirone and dopamine antagonists disinhibit punished responding, increase activity in a two-compartment exploratory task (25), attenuate apomorphine-induced stereotypy (28), and attenuate the discriminative stimulus effects of apomorphine (19). While it has been suggested that the antipunishment (31) and discriminative stimulus effects (21) of buspirone are mediated by the 5-HT_{1A} receptor, buspirone's actions at the D_2 receptor may also have behavioral relevance. Previous research has indicated that the discriminative stimulus effects of apomorphine are primarily mediated through D_2 receptors, as these effects are mimicked by other D_2 agonists and blocked by D_2 antagonists (6,27,35,36). Buspirone has been reported to attenuate the discriminative stimulus effects of apomorphine in a manner consistent with competitive antagonism, causing dose-dependent decreases in apomorphine-appropriate responding, as well as a parallel shift to the right in the dose–response function for apomorphine (19). To the extent that dopamine mediates the discriminative stimulus effects of cocaine, the pharmacological actions of buspirone described above suggest that buspirone administration might also block the discriminative stimulus of cocaine. This study was therefore undertaken to assess the ability of buspirone to antagonize the discriminative stimulus effects of cocaine in rats.

METHOD

Animals and Apparatus

Eight experimentally naive male Sprague-Dawley rats (Holtzman Co., Madison, WI) were used as subjects. Rats were maintained at 80 ± 5% of their initial free-feeding body weights by restricting food intake: Individual target weights ranged from 260–310 g. They were individually housed in stainless steel cages in a room maintained at 24°C and on a 12 L : 12 D cycle (light 7:00 a.m.–7:00 p.m.). In addition to the 45-mg food pellets (P.J. Noyes Co., Lancaster, NH) delivered during the experimental sessions, diet was supplemented with Teklad 4% Mouse and Rat Diet. Water was continuously available except during experimental sessions.

[1] Current address: Department of Neurosurgery, Box 3807, Duke University Medical Center, Durham, NC 27710.

Four identical operant chambers for rats (Ralph Gerbrands Co., Model D-1) were used. In each chamber, two response levers were mounted on one of the walls and a food receptacle was located between them. Each chamber was illuminated from the onset of an experimental session by a single 6-W light located on the wall opposite the levers. Extraneous noise was diminished by enclosing each chamber in an insulated picnic chest and operating ventilation fans mounted on the outside of each chest. An AIM-65 microcomputer (Dynatem Corp., Irvine, CA), connected to a custom-designed input-output interface (ERH Electronics, Delton, MI) located in an adjacent room controlled stimulus events and recorded lever presses.

Procedure

Animals were assigned to one of the four experimental chambers. In two chambers, the right lever was designated as the saline-appropriate lever and the left lever was designated as the drug-appropriate lever. In the other two chambers, the reverse assignments were made. Training sessions were conducted once a day, 6 days/week. Rats were pretreated with IP injections of either 4.0 mg/kg cocaine or saline and placed in the darkened chambers. After a 10-min time-out period, the houselight was illuminated and the 15-min session began. One food pellet was available for every response recorded on the injection-appropriate lever. Responses on the inappropriate lever were counted but had no other programmed consequence. Injection-appropriate lever responding was shaped by successive approximation. Training sessions were scheduled according to a double-alternation sequence in which two sessions of saline pretreatments alternated with two sessions of 4.0 mg/kg cocaine pretreatments. Once food-maintained responding had been established under each of the training conditions, the response requirement on either lever was gradually increased such that under terminal conditions every 30th consecutive response (fixed-ratio 30) on the lever appropriate to the injection resulted in the delivery of a food pellet. The double-alternation training sequence continued until a rat met the following criteria for stimulus control of responding. First, within each session at least 80% of the responses before delivery of the first food pellet and at least 90% of the responses over the entire session had to have occurred on the injection-appropriate lever. Second, the within-session criteria must have been met in seven of the last eight as well as three of the last three consecutive sessions.

Once a rat had met the criteria for stimulus control, test sessions were conducted every third session using a pretreatment sequence of 4.0 mg/kg cocaine, saline, test, saline, cocaine, test as long as performance in the training sessions between tests remained at or above the criterion for stimulus control. If a rat's performance fell below criterion levels during the intervening training sessions, it was returned to the double-alternation training sequence until discrimination was again at or above criterion levels. Occasionally, a lever preference developed and was corrected by increasing the frequency of nonpreferred lever correct sessions.

Test sessions were identical to training sessions except food was available for responding on either lever. Two types of test sessions were used. In substitution tests, saline, cocaine (0.5–16.0 mg/kg) or haloperidol (0.125 mg/kg, the dose used in antagonism experiments) were tested for their ability to substitute for the training dose of cocaine. Only those doses of cocaine necessary to generate a dose-response function that extended from less than 10% drug-appropriate responding to greater than 90% drug-appropriate responding over the entire session were tested. Saline or doses of cocaine were administered 10 min before the session, while haloperidol was administered 60 min presession in conjunction with a saline injection 10 min prior to the session. In antagonism tests, doses of buspirone (2.0–16 mg/kg 20 min presession) or haloperidol (0.125 mg/kg 60 min presession, which preliminary studies suggested would attenuate the discriminative stimulus effects of cocaine) were administered in conjunction with 4.0 mg/kg cocaine administered 10 min prior to the session. The 4.0-mg/kg dose of cocaine used in antagonism studies was the lowest dose of cocaine tested that met criterion for engendering cocaine-appropriate responding. The highest dose of buspirone that did not completely suppress responding was determined for each rat. The time intervals between the injections of cocaine, buspirone, and haloperidol and the test sessions were selected from the literature (2,16,24). Some tests were conducted twice, preceded once by a drug training session and once by a saline training session. Each test condition was tested in at least five rats.

Data Analysis

The percent of the total responses that occurred on the drug-appropriate lever and the rate of responding on both levers during test sessions were calculated for each rat. Where two determinations were made, the results were averaged for use in further analysis. If a rat failed to receive a food pellet in any test session, the data for that session were not included in the percent drug-appropriate responses calculation but were included in the response rate determination. The number of sessions from the first training session to the first session in which the criteria for discriminative stimulus control of behavior were met was determined for each rat and recorded as sessions to criteria. For each test condition comprising the substitution tests, the mean and SEM were calculated for the group for both the percent drug-appropriate responses and the response rate measure. For antagonism tests, the percent of total responses that occurred on the drug-appropriate lever was calculated for each rat. A paired *t*-test for repeated measures was used to compare group data of percent drug-appropriate responses occasioned in test sessions by 4.0 mg/kg cocaine alone to the percent drug-appropriate responses occasioned by the combination of cocaine and antagonist.

Drugs

Cocaine HCl (National Institute on Drug Abuse, Rockville, MD) and buspirone (Bristol-Myers, Evansville, IN) were dissolved in 0.9% saline. Haloperidol (McNEILAB, Inc., Fort Washington, PA) was obtained in solution and diluted to the desired concentration in the solvent provided by the supplier. All injections were 1.0 ml/kg IP and the concentrations were varied appropriately.

RESULTS

Rats acquired the cocaine-saline discrimination with a mean number of sessions to criterion of 42. Cocaine (0.5–8.0 mg/kg) engendered a dose-related increase in cocaine-appropriate responding, with the training dose of 4.0 mg/kg being the lowest dose that met criterion (>90% drug-appropriate responding over the entire session) for substitution (Fig. 1). The mean response rate was 2.0 responses per second under both the saline and 4.0-mg/kg cocaine test conditions. Higher doses of cocaine engendered 100% drug-appropriate responding and decreased response rates.

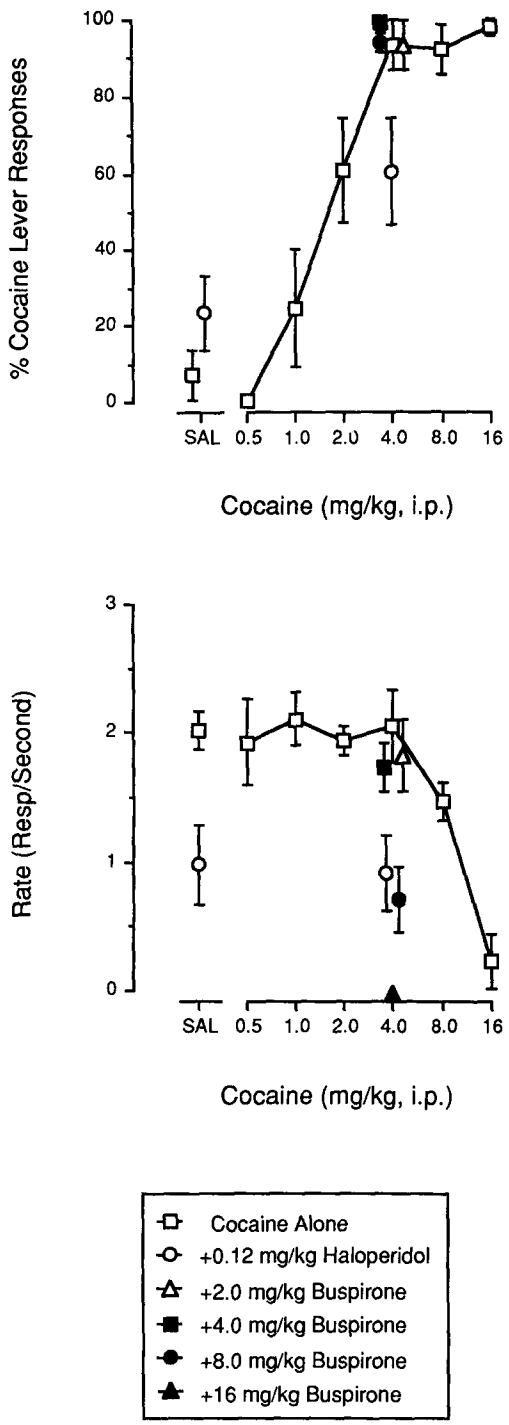

FIG. 1. Dose-response functions for the discrimination of cocaine (top) and the effects of cocaine on response rate (bottom) in a fixed-ratio 30, food-reinforced, drug discrimination paradigm. The effects of combining buspirone (2.0–16 mg/kg) or haloperidol (0.125 mg/kg) with 4.0 mg/kg cocaine were tested. Bars represent the SEM of each point. In those cases in which error bars are not shown, SEMs are contained within the point. Points plotted above saline and the 4.0-mg/kg cocaine dose were separated for visual clarity.

Administration of buspirone (2.0–8.0 mg/kg) in combination with 4.0 mg/kg cocaine (the training dose) did not alter the percent of cocaine-lever responding. Response rates were suppressed to 38% ($p = 0.003$) of control level by combinations of 8.0 mg/kg buspirone and 4.0 mg/kg cocaine, and responding was completely suppressed by combinations of 16 mg/kg buspirone and 4.0 mg/kg cocaine (Fig. 1).

In contrast, combinations of 0.12 mg/kg haloperidol and 4.0 mg/kg cocaine decreased drug-appropriate responding from 100 to 65% ($p = 0.04$). This treatment suppressed response rate to 50% ($p = 0.007$) of the response rate seen with the training dose of cocaine (Fig. 1).

DISCUSSION

Buspirone did not attenuate the discriminative stimulus effects of 4.0 mg/kg cocaine IP at doses up to those that caused complete suppression of responding. Methodological issues cannot account for the failure of buspirone to block the discriminative stimulus effects of cocaine. The preparation was sensitive to the discriminative stimulus effects of cocaine because the discrimination was stable during the training sessions and dose related during test sessions. Further, the buspirone doses tested were shown to be behaviorally active by their suppression of response rate: Higher doses of buspirone could not be tested because they completely suppressed responding. Administration of the D_2 antagonist haloperidol attenuated the discriminative stimulus effects of cocaine, a finding that is consistent with the literature (2,3,18). Thus, the drug-discrimination paradigm used in the present experiment was sensitive to the ability of test compounds to attenuate the discriminative stimulus effects of cocaine.

One possible explanation of why buspirone did not antagonize the discriminative stimulus effects of cocaine is that it does not have sufficient antidopaminergic activity. Like dopamine receptor antagonists, buspirone can enhance firing of dopaminergic neurons in the substantia nigra and reverse the inhibition of those neurons caused by dopamine, the indirect dopamine agonist amphetamine, or the direct dopamine receptor agonist apomorphine (23). However, the similarity between buspirone and other dopamine antagonists is limited in this regard. The maximal increase in cell firing in the zona compacta of the substantia nigra caused by the D_2 antagonist haloperidol (0.5–1.0 mg/kg) is 20% while the maximal enhancement caused by buspirone (1.0–2.0 mg/kg) approaches 95%, and neither D_1 agonists nor antagonists affect dopamine release in this region (23). Other findings call into question the efficacy of buspirone as an antidopaminergic agent. Buspirone can mimic the ability of the dopamine receptor agonist apomorphine to induce contralateral rotation in rats that have been unilaterally lesioned with 6-hydroxydopamine in the substantia nigra and reverse the catalepsy induced by dopaminergic antagonists (26). The dose-response function for the release of punished responding by buspirone is unaffected by either a dopamine agonist or antagonist (30). In addition, chronic buspirone administration does not result in the increases in dopamine receptors in in vitro binding assays that are seen after chronic administration of other dopamine antagonists (15).

There is evidence that buspirone interacts with the serotonergic neurotransmitter system. For instance, the serotonin$_{1A}$ (5-HT$_{1A}$) receptor ligand 8-OH-DPAT substituted for buspirone in pigeons trained to discriminate buspirone from saline, while dopaminergic agonists, a dopaminergic antagonist, a benzodiazepine, and a serotonin$_{1B}$ receptor ligand produced

no buspirone-appropriate responding (21). In an antipunishment paradigm, the release of punished responding caused by buspirone was attenuated by the serotonin agonist MK-212, potentiated by the serotonin antagonist cyproheptadine, and mimicked by 8-OH-DPAT (31). In binding studies, buspirone has been found to bind to serotonin$_{1A}$ sites with high affinity (31). These findings suggest that the behavioral effects of buspirone are mediated by the 5-HT$_{1A}$ receptor. Thus, it is possible that the behavioral actions of buspirone in the drug discrimination paradigm are mediated by pharmacological activity involving a nondopaminergic (5-HT$_{1A}$) rather than a dopaminergic neurotransmitter system. The position that activation of serotonergic receptors does not mediate the discriminative stimulus effects of cocaine is supported by findings that the serotonergic antagonists methysergide and cyproheptadine fail to attenuate the discriminative stimulus effects of cocaine (4).

While a lack of sufficient antidopaminergic activity may underlie buspirone's failure to block the discriminative stimulus effects of cocaine, it should be noted that antidopaminergic agents in general may not completely block the behavioral effects of cocaine as these effects may not be exclusively mediated by dopamine. Although there is evidence suggestive of a role for dopamine in the discriminative stimulus properties of cocaine, the evidence is not conclusive. Direct dopamine agonists and some compounds that increase dopaminergic tone only partially substitute for cocaine, and, as in the present study, dopamine antagonists only partially block the discrimination of cocaine (4,18,22,33). Other compounds (phencyclidine, opiate agonists, an anticholinergic, a local anesthetic, and several monoamine oxidase inhibitors) have also been shown to substitute partially or completely for cocaine (4,5). Although each of these compounds is capable of increasing dopaminergic tone at the synapse (7,11–13,20), there are some instances of partial substitution for cocaine by drugs that do not have dopaminergic effects (clonidine, nicotine, propanolol, and β-phenethylamine (4,8,9,32,34). The implications of the latter studies are not well understood, although they raise the possibility that the discriminative stimulus properties of cocaine are not mediated exclusively by dopamine.

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