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Behavioral Effects of Selective Dopaminergic Compounds in Rats Discriminating Cocaine Injections[1]

JEFFREY M. WITKIN, DAVID E. NICHOLS, PHILIP TERRY and JONATHAN L. KATZ

Drug Development Group (J.M.W., P.T., J.L.K.), Psychobiology Laboratory, Preclinical Pharmacology Branch, National Institute on Drug Abuse Addiction Research Center, Baltimore, Maryland and Department of Medicinal Chemistry and Pharmacognosy (D.E.N.), School of Pharmacy and Pharmacal Sciences, Purdue University, West Lafayette, Indiana

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ABSTRACT

The involvement of dopamine receptor subtypes in the discriminative stimulus effects of cocaine was evaluated by the ability of a series of compounds selective for D_1 or D_2 dopamine receptors to produce discriminative stimulus effects comparable to cocaine. Male, Sprague-Dawley rats were trained to discriminate 10 mg/kg of cocaine HCl from saline in a two-lever discrimination procedure. Inhibitors of catecholamine and serotonin reuptake (GBR 12909, WIN 35,428 and mazindol), *d*-amphetamine, and the nonselective dopamine agonist, apomorphine, produced dose-dependent increases in cocaine-appropriate responding and fully substituted for cocaine. Cocaine methiodide, a charged, quaternary cocaine analog, did not substitute for cocaine. Neither pentobarbital, haloperidol nor SCH 23390 produced cocaine-like behavioral activity. Both D_1- and D_2-selective agonists with diverse structures partially substituted for cocaine, producing from 40 to 80% cocaine-appropriate responses. The D_1 agonists studied were SKF 38393 and stereoisomers, SKF 75670 and CY 208–243. Dihydrexidine, a full D_1 agonist for induction of adenylate cyclase activity, also only partially substituted for cocaine. The peripherally acting D_1 agonist, fenoldopam, produced predominantly saline-appropriate responding that was unrelated to dose. The D_2 agonists tested were pergolide, quinpirole, (−)-NPA, RU 24213, N-0434 and N-0437. The D_2 antagonist haloperidol did not block the discriminative stimulus effects of cocaine. In contrast, the D_1 antagonist SCH 23390 reduced the discriminative stimulus effects of cocaine by a maximum of 50%. These results suggest that both D_1 and D_2 receptors may play a role in the discriminative stimulus effects of cocaine but that stimulation of either dopamine receptor subtype alone is not sufficient.

Increased levels of cocaine abuse and associated escalations in toxicity have expanded efforts to identify mechanisms of action of cocaine relevant to its abuse. The development of effective pharmacological treatment strategies is dependent upon critical advances in this area. In preclinical assessments, animal subjects can be trained readily to report the presence or absence of cocaine (*cf.* Colpaert, 1986). Once behavior is under the discriminative stimulus control of cocaine, substitution or antagonism by compounds with specified mechanisms of action can be evaluated to help elucidate the pharmacological mechanisms of action of cocaine. Prior investigations have suggested a role for dopaminergic neurotransmission in the discriminative stimulus effects of cocaine (McKenna and Ho, 1980; Wood and Emmett-Oglesby, 1987).

The existence of multiple dopamine receptor subtypes in the central nervous system (*cf.* Kebabian and Calne, 1979; Clark and White, 1987) and compounds which interact selectively with these sites (*cf.* Waddington and O'Boyle, 1989) has permitted direct investigation of the role of dopamine receptors in the behavioral effects of cocaine (see Johanson and Fischman, 1989; Woolverton and Kleven, 1988 for reviews). Recent findings that blockade of either D_1 or D_2 dopamine receptors selectively altered behavioral, cardiovascular or toxic effects of cocaine have illustrated the potential differential involvement of subtypes of the dopamine receptor in specific pharmacological actions of cocaine (Kleven *et al.*, 1988; Katz and Witkin, 1991; Schindler *et al.*, 1991; Witkin *et al.*, 1989, 1991). For example, Witkin *et al.* (1989) reported protection against the lethal effects of cocaine by the D_1 antagonist SCH 23390 but not by the D_2 antagonist haloperidol.

The involvement of dopamine receptor subtypes in the discriminative stimulus effects of cocaine remains uncertain due to the weak or inconsistent results obtained with both selective dopamine agonist and antagonist treatments. The discriminative stimulus effects of cocaine have been reported to be only partially blocked by antagonists of D_2 receptors (*cf.* Colpaert *et al.*, 1978; Callahan *et al.*, 1990; Jarbe, 1978). For example, in rats trained to discriminate injections of cocaine from saline, haloperidol produced only a 40% reduction in the control of behavior by cocaine (Colpaert *et al.*, 1978). Other reports have shown that D_2 antagonists are either inactive or produce blockade of the discriminative stimulus effects of cocaine that is not dose-related (Barrett and Appel, 1989). Likewise, whereas the

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[1] Animals used in this study were maintained in facilities fully accredited by the American Association for the Accreditation of Laboratory Animal Care and the studies were conducted in accordance with the Guide for Care and Use of Laboratory Animals provided by the National Institutes of Health and adopted by National Institute on Drug Abuse.

D_1 antagonist SCH 23390 has been reported to block the discriminative stimulus effects of cocaine (Kleven *et al.*, 1988, 1990), Barrett and Appel (1989) and Callahan *et al.* (1991) showed that antagonism was neither complete nor dose-dependent. In contrast, some authors have reported shifts in dose functions for the behavioral effects of cocaine by both D_1 and D_2 antagonists (Kleven *et al.*, 1990; Spealman, 1990). Interpretation of these data are complicated further because large, behaviorally active doses of the antagonists are typically required for antagonist activity (*cf.* Callahan *et al.*, 1991; Katz and Witkin, 1991; Woods *et al.*, 1987).

The use of prototypical agonists has also not been completely successful in unambiguously defining a role for either dopamine receptor subtype in the behavioral effects of cocaine. In rats discriminating 10 mg/kg of cocaine from saline, the D_2 agonist quinpirole has been reported to dose-dependently substitute for cocaine (Barrett and Appel, 1989; Callahan *et al.*, 1991) but not in rhesus monkeys (Kleven *et al.*, 1990). SKF 38393, an agonist at D_1 receptors, either partially substitutes for cocaine (Callahan *et al.*, 1990) or is completely devoid of cocaine-like discriminative stimulus effects (Barrett and Appel, 1989; Kleven *et al.*, 1990). Because compounds selective for either dopamine receptor subtype differ substantially along a number of pharmacological dimensions (*e.g.*, receptor specificity, full *vs.* partial agonists and stimulation of second messenger systems), idiosyncratic pharmacological activities of a particular compound could determine behavioral effects. If D_1 or D_2 receptors play a role in the discriminative effects cocaine, then most compounds with affinities for these receptors should produce common behavioral effects. The present investigation was therefore conducted to provide dose-response comparisons of a range of structures with high specificity for D_1 or D_2 receptors in rats discriminating cocaine.

Methods

Animals. Eighteen adult male Sprague-Dawley rats (Charles River, Wilmington, MA) maintained at 350 g in individual cages were used. All rats were experimentally naive before this study. Water was available continuously for all rats in their living cages. The rats were housed under a 12-hr light/dark cycle and experiments were conducted during the light phase.

Apparatus. Experiments were conducted in operant-conditioning test chambers (BRS/LVE, model RTC-022). Two response levers, 17 cm apart, were centered on the front panel on either side of a food pellet trough. Food (45-mg pellets, BioServe, Frenchtown, NJ) could be delivered to the central pellet trough *via* a motor-operated feeder. White lamps above the right lever and red lamps above the left lever could be illuminated simultaneously. A white lamp at the top center of the front panel provided general illumination of the chamber. Chambers were enclosed within sound- and light-attenuating enclosures and supplied with white noise to mask extraneous sounds. Depression of the lever with a downward force of 0.4 N through 1 mm was recorded as a response and produced the audible click of a relay when the houselight and the stimulus lamps over the levers were illuminated.

Cocaine discrimination. After initial training to eat food pellets, rats were trained to depress either the right or left lever. During bar-press training, depression of the appropriate lever produced a food pellet. The rats were assigned randomly to the left or right lever for initial bar-press training to counterbalance any lever biases. Lights above only the active lever were illuminated and responses on the inactive lever had no scheduled consequences. On subsequent sessions, from one to five lever presses were required for food delivery, and the lever upon which responses produced food was varied from session to session.

Subsequently, rats were trained to discriminate i.p. injections of 10.0 mg/kg of cocaine HCl from saline. After cocaine administration, responses on only one lever produced food; after saline administration, responses on the opposite lever produced food. The lever correlated with cocaine was counterbalanced across animals. Under the cocaine-saline discrimination, a 5-min timeout occurred at the beginning of the session; during timeout the chamber was dark and responding had no scheduled consequences. After the timeout period, either 20 or 30 consecutive responses on the appropriate lever were required for food presentation, depending on the rat. Responses on the alternate lever reset the response requirement. During this phase of training and maintenance of the cocaine discrimination, lights above both the right and left response levers were illuminated simultaneously. During early stages of discrimination training, the number of responses required for food delivery was sometimes reduced in order to maintain responding. Timeout periods of 20 sec followed each food delivery, and sessions lasted until 20 food pellets had been presented or 20 min, whichever occurred first.

Test sessions in which responding on either lever produced food were conducted once greater than 85% of responses occurred on the injection-appropriate lever during regular training sessions. Discriminative performances were considered stable when greater than 85% of responses before the first food pellet and during the entire experimental session occurred on the injection-correlated lever during test sessions. After this criterion had been achieved, similar test sessions were then conducted after administration of test compounds to evaluate their ability to produce responding similar to cocaine. Test sessions with 10 mg/kg of cocaine or saline were also conducted regularly during dose-effect determinations in order to establish control values against which to compare the effects of test compounds. Injections of test compounds were given no more than twice weekly and only if responding met the 85% criteria during the preceding experimental session.

Drugs. (−)-Cocaine HCl (Mallinckrodt/Nuclear, Orland, FL), Win 35,428 (Clarke *et al.*, 1973; Sterling Winthrop, Rensselaer, NY), *d*-amphetamine SO_4 (Sigma Chemical Co., St. Louis, MO), apomorphine HCl (Sigma) and pentobarbital Na (Abbott Laboratories, N. Chicago, IL) were dissolved in 0.9% NaCl. Haloperidol (Hyttel, 1983; McNeil Pharmaceuticals, Spring, House, PA), SCH 23390 (Iorio *et al.*, 1983; Hyttel, 1983; Research Biochemicals, Inc., Wayland, MA), mazindol (Javitch *et al.*, 1984), CY 208–243 (benzergoline, Foote *et al.*, 1988; Sandoz Pharmaceuticals, Hanover, NJ), pergolide mesylate (Arnt *et al.*, 1988; LY127809, Eli Lilly and Company, Indianapolis, IN), N-0434 (van der Weide *et al.*, 1986; Research Biochemicals) and N-0437 (van der Weide *et al.*, 1986; Nelson Research Laboratories) were dissolved in distilled water with minimal acidification required for solution. The following compounds were dissolved in distilled water in which mild heating and sonication were often required: cocaine methiodide (Shriver and Long, 1971; National Institute on Drug Abuse, Baltimore, MD, synthesized by Research Triangle Institute), GBR 12909 (Heikkila and Manzino, 1984; Research Biochemicals, Inc.), SKF 38393 and isomers of SKF 38393 (Setler *et al.*, 1978; O'Boyle *et al.*, 1989; Research Biochemicals, Inc.), SKF 75670 (Arnt *et al.*, 1988; O'Boyle *et al.*, 1989; Smith Kline and French Laboratories, Philadelphia, PA), dihydrexidine (*trans*-10,11-dihyroxy-5,6,6a,7,8,12b-hexahydrobenzo[*a*]phenanthridine, synthesized as described by Brewster *et al.*, 1990), fenoldopam mesylate (Hahn *et al.*, 1982; SKF 82526-J, Smith Kline and French Laboratories), Ru 24213 (N-*n*-propyl-N-phenylethyl-*p*-(3-hydroxyphenyl)-ethylamine), Euvard *et al.*, 1979; Roussel), quinpirole (Titus *et al.*, 1983; Research Biochemicals) and (−)-NPA (Arnt *et al.*, 1988; Research Biochemicals). Doses are expressed in terms of the drug forms listed above. Drugs were generally injected i.p. in a volume of 1 ml/kg immediately before experimental sessions (sessions began with a 5-min timeout which served as a pretreatment time). During drug interaction experiments, haloperidol or SCH 23390 were given 30 min before saline or 10 mg/kg of cocaine. Both i.p. and s.c. routes of administration were tested with SCH 23390 based upon previous work (Arnt *et al.*, 1988; Callahan *et al.*, 1991; Nielsen and Jepsen, 1985). Drug doses were given in a mixed sequence in at least six rats. The

order of drug determinations differed across the three groups of six rats.

Data analysis. Experimental events and data collection were controlled by a PDP 11/73 computer operating under SKED behavioral software (State Systems, Inc.). Rates of responding and the percentage of responses on the cocaine-appropriate lever were obtained for individual animals. Dose-effect functions were determined by averaging determinations across at least six rats. The percentage of responses on the cocaine lever was not considered reliable if the overall rate of responding was reduced by 85% or greater in individual animals; therefore, these data were not used in the calculation of the dose-response curves for discriminative stimulus effect. Higher doses of most test compounds therefore consisted of estimates of response distributions in less than the full complement of animals tested. However, results from at least three animals were required for construction of this portion of the dose-response curve.

Evaluation of the maximal efficacy of the test compounds was ensured by administering a full range of doses for each compound and drug combination. Where quantitative estimates of drug effects were calculated, dose-effect functions were analyzed using data from the linear portion of the curves using standard bioassay analysis of variance techniques (Finney, 1964; Snedecor and Cochran, 1967). Statistical evaluation of whether the effects of individual doses differed from the effects of the training stimulus conditions, *i.e.*, either saline or 10 mg/kg of cocaine, was based upon a conservative criterion in which drug effects greater than 20% cocaine responses were considered significantly different than the effects of saline; drug effects greater than 80% cocaine responses were considered to be similar to 10 mg/kg of cocaine.

Results

Control performance. Once behavior stabilized under the cocaine-saline discrimination, control of behavior by drug injections was maintained with a high degree of accuracy. During the course of the experiment in which the effects of cocaine and saline were assessed, saline produced 2.9 ± 2.9% and 10 mg/kg of cocaine produced 96.0 ± 3.4% cocaine lever responses (mean ± S.D.). Dose-effect functions for cocaine were relatively steep with transition from predominantly saline- to cocaine-appropriate responding occurring across doses within one order of magnitude (fig. 1, top panel, □). The ED_{50} value for cocaine was estimated to be 1.5 mg/kg (1.08–2.20 mg/kg; 95% CL, n = 12). Across these doses of cocaine, response rates were not affected appreciably (fig. 1, bottom panel, □).

Nonsubtype-selective compounds. Effects of several drugs that do not interact selectively with dopamine receptor subtypes are shown in figure 1. The long-acting cocaine analog, WIN 35,428 produced dose-dependent increases in cocaine lever responses as did the structurally distinct dopamine reuptake inhibitors mazindol and GBR 12909; these drugs fully substituted for the training dose of cocaine. In contrast, the stable charged analog, cocaine methiodide, did not produce full cocaine-appropriate responding even at doses as high as 56 times the minimally effective dose of cocaine HCl. Higher doses could not be tested due to potential toxicity. The nonselective direct dopamine agonist, apomorphine, produced full cocaine-appropriate responding. Pentobarbital did not produce effects that differed from saline at any of the doses tested. Rates of responding were not affected substantially by most of the compounds at doses that produced significant cocaine-appropriate responding. The exceptions to this were *d*-amphetamine where response rates were increased at moderate doses and apomorphine which produced dose-dependent decreases in rates of responding. For apomorphine, substitution for cocaine was obtained only at doses that markedly disrupted responding.

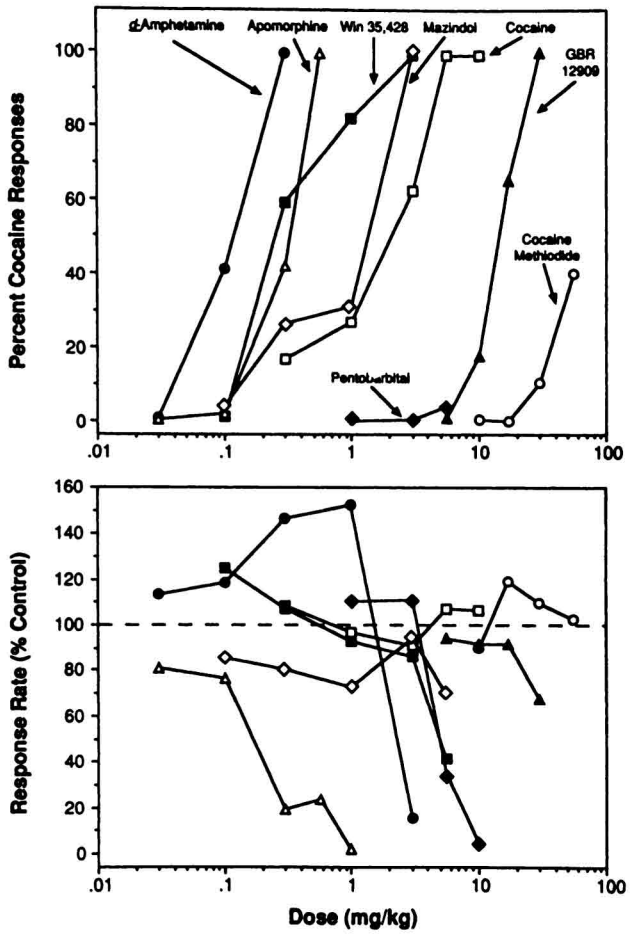

Fig. 1. Effects of cocaine, cocaine analogs, central nervous system stimulants and pentobarbital on the percentage of responses on the cocaine-correlated lever (top panel) and on rates of responding (bottom panel). Rats were trained to discriminate 10 mg/kg of cocaine from saline as described under "Methods." Each compound was administered separately immediately before experimental test sessions. Each point represents the mean effect in at least six rats (with the exception, top panel only, of 30 mg/kg of GBR 12909, n = 4; 0.56 mg/kg of apomorphine, n = 4; 3 mg/kg of mazindol, n = 5; and 5.6 mg/kg of pentobarbital, n = 3). Higher doses of *d*-amphetamine, WIN 35,428 and mazindol, all of which produced greater than 95% cocaine responses, are not shown in the top panel to avoid clutter.

D_1 agonists. Seven D_1 agonists were evaluated for their ability to engender responding on the cocaine lever (fig. 2, top panels). The 1-phenyl-3-benzazepines, SKF 38393 and SKF 75670 produced dose-related increases in responding on the cocaine lever. However, neither compound fully substituted for cocaine. SKF 75670 was the most efficacious of the D_1 agonists tested, producing effects which nearly reached cocaine levels at 3 mg/kg (78%), the highest dose in which enough responding occurred to estimate response distributions. CY 208–243 also produced dose-dependent increases in cocaine-lever responses. The maximal effect of CY 208–243 was 60%. Dihydrexidine, a phenanthridine with full D_1 agonist activity also produced partial substitution for cocaine with a maximum of 38% cocaine-appropriate responses at 3 mg/kg. Fenoldopam, a D_1 agonist with limited availability to the central nervous system upon systemic administration, produced effects that were not related to dose. Only one of the doses of fenoldopam produced effects significantly different than saline. All of the D_1 agonists tested decreased response rates at doses that produced signifi-

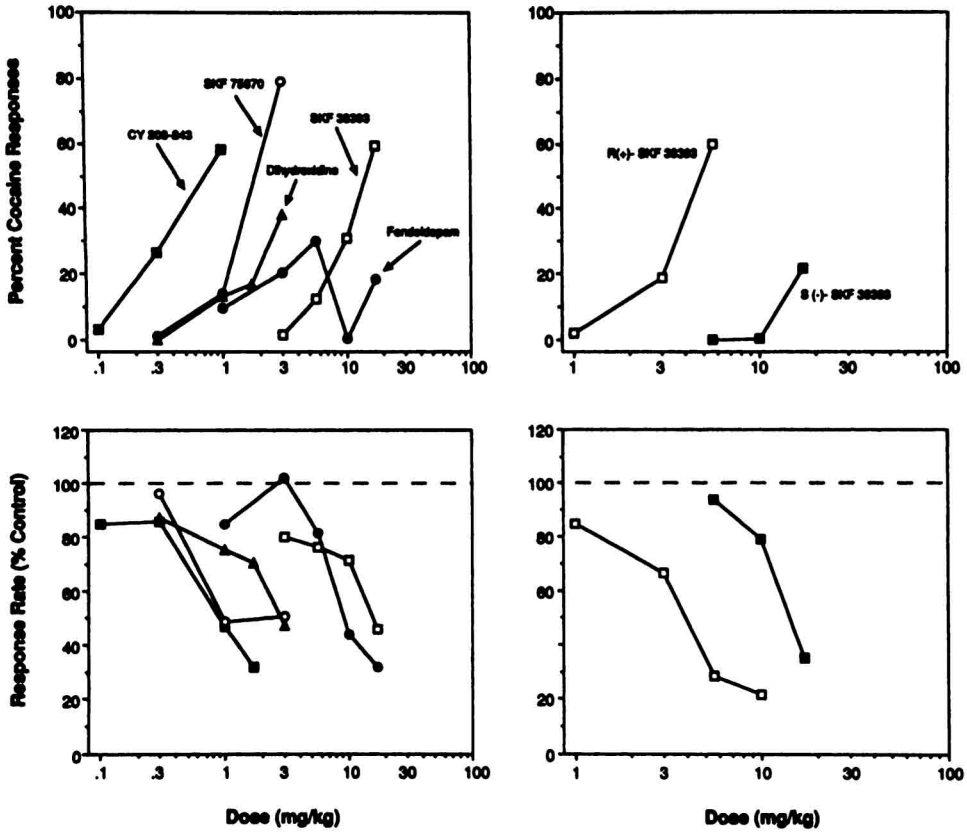

Fig. 2. Effects of D_1 agonists on the percentage of responses on the cocaine-correlated lever (top panel) and on rates of responding (bottom panel). Other details as in figure 1. Each point represents the mean effect in at least six rats (with the exception, top panel only, of 17 mg/kg of SKF 38393, $n = 4$; 10 mg/kg of (+)-SKF 38393, $n = 5$; 17 mg/kg (−)-SKF 38393, $n = 5$; 1 mg/kg of CY 208–243, $n = 3$; 17 mg/kg of fenoldopam, $n = 3$; and 3 mg/kg of SKF 75670, $n = 4$).

cant effects, although in most cases this reduction was not substantial (fig. 2, bottom panels).

The discriminative stimulus effects of SKF 38393 were stereospecific (fig. 2, right panel). The (−)-enantiomer had minimal activity even at the highest doses tested. The (+)-isomer with the D_1 receptor activity of the isomeric pair was more efficacious and about 6 times more potent than the inactive enantiomer (fig. 2, right panel). Both isomers of SKF 38393 produced dose-dependent reductions in rates of responding.

D_2 agonists. Six D_2 agonists were tested for their ability to produce effects comparable to cocaine (fig. 3, top panel). All of the compounds produced dose-related increases in the percentage of cocaine lever responses. These compounds produced between 40 and 70% responding on the cocaine-correlated response lever. N-0434 and N-0437 produced effects that differed significantly from saline across a wider dose range than observed with the other D_2 agonists. All of the D_2 agonists produced dose-related decreases in rates of responding (fig 3., bottom panel). Responding was suppressed markedly by all of the compounds at doses that produced significant increases in the percentage of cocaine responses with the exception of N-0434 and N-0437 in which partial substitution for cocaine was observed at doses that only decreased responding modestly.

Effects of haloperidol. When given 30 min before administration of 10 mg/kg of cocaine, haloperidol (0.01–0.3 mg/kg) did not significantly attenuate the discriminative stimulus effects of cocaine (fig. 4, top panel, filled symbols). At the highest dose of haloperidol (0.3 mg/kg), the percentage of responses on the cocaine-appropriate lever was 87%. Haloperidol alone produced discriminative stimulus effects that were not significantly different from saline (fig. 4, top panel, unfilled symbols). In the presence of cocaine, the dose-effect function for the rate-suppressant effects of haloperidol was shifted to the right (fig. 4, bottom panel).

Effects of SCH 23390. The D_1 antagonist, SCH 23390, decreased cocaine appropriate responding by 50% (0.03 and 0.1 mg/kg) when given by the i.p. route (fig. 5, top left, filled symbols). At higher doses of SCH 23390 (0.17–0.3 mg/kg), the discriminative stimulus effects of the training dose of cocaine (10 mg/kg) were not altered. Thus, a biphasic function described the interaction of SCH 23390 and cocaine, administered i.p. When SCH 23390 was given alone at a high dose (0.1 mg/kg), the percentage of cocaine responses was significantly greater than saline (fig. 5, top left, unfilled symbols). Responding was not decreased appreciably by doses of SCH 23390 that were effective in blocking the discriminative stimulus effects of cocaine and in partially substituting for cocaine when given alone (fig. 5, bottom left).

Like i.p. administration, s.c. administration of SCH 23390 produced only a 50% maximal inhibition of the discriminative-stimulus effects of cocaine. SCH 23390 was about 10 times more potent by the s.c. than the i.p. route (see also Callahan *et al.*, 1991).

However, in contrast to the i.p. route, the percentage of responses on the cocaine lever was not enhanced when SCH 23390 was given alone by the s.c. route (fig. 5, top right). Rates of responding were not altered appreciably by the lowest dose of SCH 23390 (0.003 mg/kg) that was effective as an antagonist of the discriminative stimulus effects of cocaine (fig. 5, bottom right).

Discussion

The present study examined the role of dopamine receptors in the discriminative stimulus effects of cocaine. The full sub-

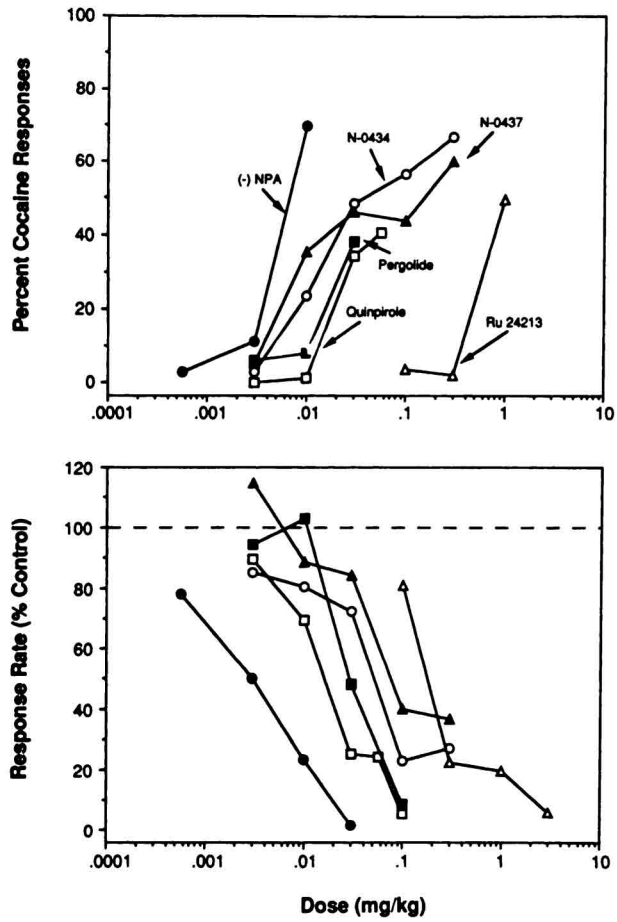

Fig. 3. Effects of D_2 agonists on the percentage of responses on the cocaine-correlated lever (top panel) and on rates of responding (bottom panel). Other details as in figure 1. Each point represents the mean effect in at least six rats (with the exception, top panel only, of 0.056 mg/kg of quinpirole, $n = 3$; 0.03 mg/kg of pergolide, $n = 5$; 0.01 mg/kg of (−)-NPA, $n = 5$; 0.1 and 0.3 mg/kg of N-0434, $n = 4$ and 3, respectively; 0.1 and 0.3 mg/kg of N-0437, $n = 3$; and 1 mg/kg of Ru 24213, $n = 3$).

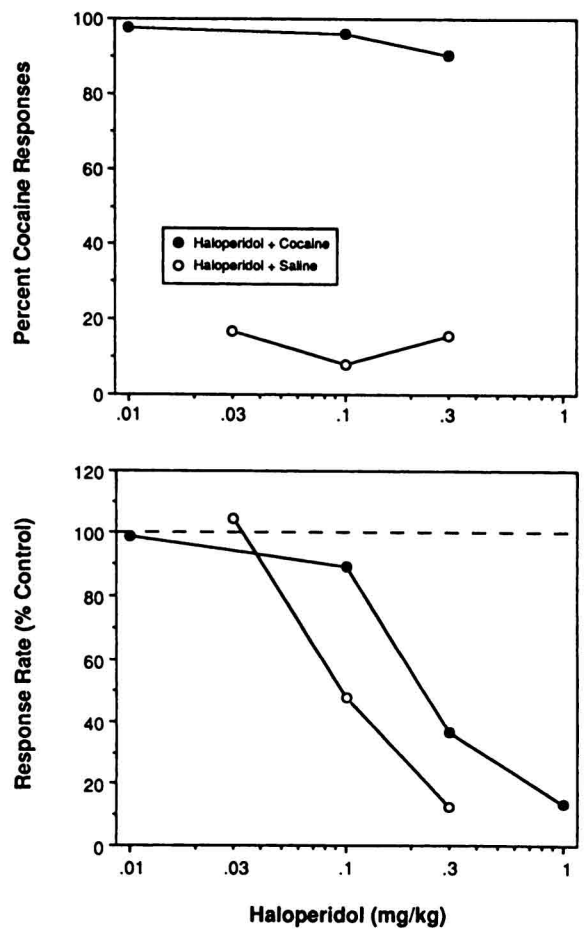

Fig. 4. Effects of the D_2 antagonist haloperidol alone and in conjunction with 10 mg/kg of cocaine. Haloperidol was administered 30 min before 10 mg/kg of cocaine or saline as described under "Methods." Each point represents the mean effect in at least six rats (with the exception, top panel only, of 0.1 and 0.3 mg/kg of haloperidol alone, $n = 5$ and 4, respectively, and 0.3 mg/kg of haloperidol with cocaine, $n = 4$).

stitution for cocaine by the dopamine reuptake inhibitors, GBR 12909 and mazindol, suggests a role for dopamine in the discriminative stimulus effects of cocaine (see also comparable findings in Kleven *et al.*, 1990). Data suggesting that inhibition of other neurotransmitter reuptake systems are not critical to the discriminative stimulus effects of cocaine was provided by Kleven *et al.* (1990) in which inhibitors of norepinephrine and serotonin uptake did not substitute for cocaine. Additional data from the present study indicated that stimulation of postsynaptic dopamine receptors is involved in the discriminative stimulus effects of cocaine. These data include the full substitution of apomorphine and the partial substitution of the selective D_1 and D_2 agonists.

The partial substitution of compounds with high specificity for D_1 or D_2 receptors provide evidence that stimulation of either D_1 or D_2 receptors alone cannot fully account for the pharmacological activity of cocaine. Previous investigations have provided mixed results with the prototypic agonists, SKF 38393 and quinpirole. Barrett and Appel (1989) and Callahan *et al.* (1991) reported that quinpirole fully substituted, whereas SKF 38393 only partially substituted for cocaine in rats. Quinpirole but not SKF 38393 only partially substituted for cocaine in rhesus monkeys (Kleven *et al.* 1990). The observations in the present study with structurally diverse agonists supports the conclusion that separate stimulation of either D_1 or D_2 receptors is not sufficient to engender cocaine-like discriminative stimulus effects, and therefore would probably not fully reproduce the subjective effects of cocaine in humans.

Additional support for this conclusion can be gleaned from studies with selective dopamine antagonists. The D_2 antagonist haloperidol did not alter the discriminative stimulus effects of cocaine in the present study (see also Barrett and Appel, 1989). Other studies have also indicated that haloperidol generally does not fully block the discriminative stimulus effects of cocaine (see Introductory section). In contrast, the D_1 antagonist SCH 23390 consistently attenuated the discriminative stimulus effects of cocaine by a maximum of 50%. Moreover, partial blockade of the discriminative stimulus effects of cocaine was achieved at doses of SCH 23390 that were devoid of significant behavioral effects when given alone. However, the conclusion that D_1 receptors are preferentially involved in the discriminative control of behavior by cocaine is tenuous. D_1 agonists do not consistently substitute for cocaine across studies (see above) and SCH 23390 also binds to serotonin-2 and serotonin-1C receptors in addition to its selectivity as a D_1 antagonist (Hyttel, 1978; Nicklaus *et al.*, 1988). SCH 23390 has

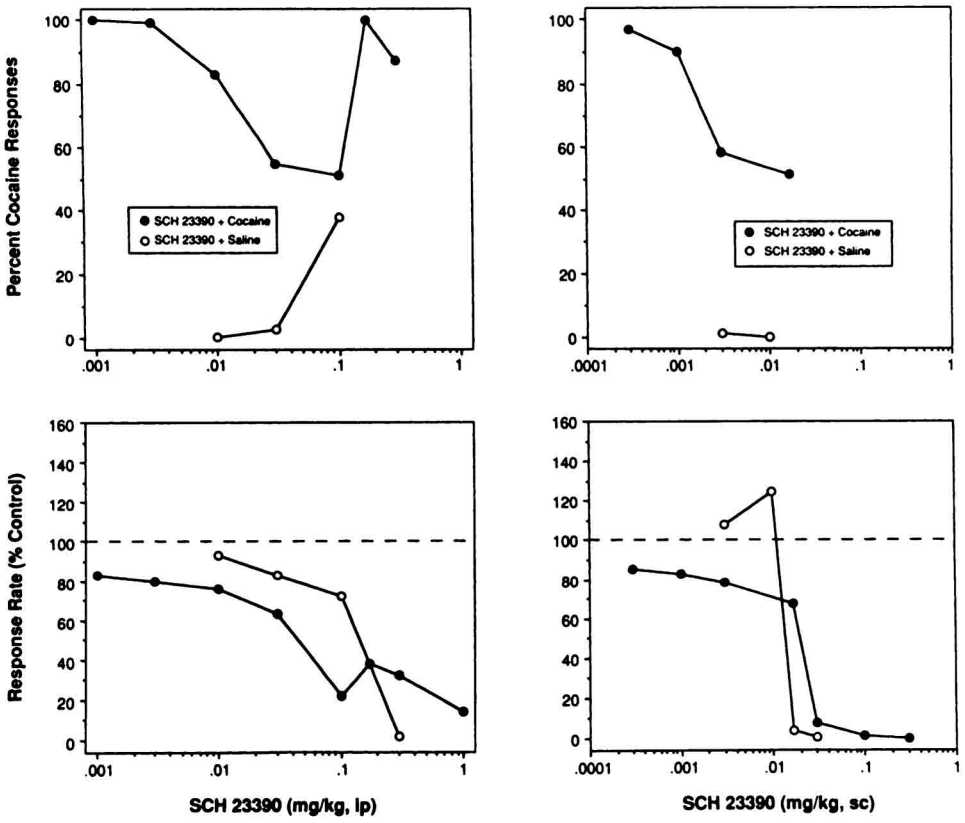

Fig. 5. Left panels, effects of i.p. administration of the D_1 antagonist SCH 23390 alone and in conjunction with 10 mg/kg of cocaine. SCH 23390 was administered 30 min before either 10 mg/kg of cocaine or saline as described under "Methods." Each point represents the mean effect in at least six rats (with the exception, top panel only, of 0.1 mg/kg of SCH 23390 alone, $n = 3$ and 0.3 mg/kg of SCH 23390 with cocaine, $n = 5$). Right panels, effects of s.c. administration of the D_1 antagonist SCH 23390 alone and in conjunction with 10 mg/kg of cocaine. SCH 23390 was administered 30 min before either 10 mg/kg of cocaine or saline as described under "Methods." Each point represents the mean effect in at least six rats (with the exception, top panel only, of 0.017 mg/kg of SCH 23390 with cocaine, $n = 5$).

also been shown to block behavioral effects of D_2 agonists (*cf.* Clark and White, 1987).

By the i.p. route, SCH 23390 displayed a U-shaped dose-effect function against cocaine, with higher doses providing less protection than moderate doses. A similar finding was reported by Barrett and Appel (1989). The partial substitution of SCH 23390 at higher doses may be related to this finding. Partial substitution of SCH 23390 has also been reported under quinpirole-saline discriminations (Williams *et al.*, 1990; Whethersby and Appel, 1986).

Although correlational data have suggested that some of the behavioral effects of cocaine related to its abuse may be related to increased availability of postsynaptic dopamine (Bergman *et al.*, 1989; Ritz *et al.*, 1987; Spealman *et al.*, 1989), the present findings suggest that the involvement of either D_1 or D_2 receptors in the discriminative control of behavior by cocaine may be limited. However, the functional interaction of dopamine receptor subtypes (*cf.* Waddington and O'Boyle, 1989) may have some bearing on this conclusion. Nonetheless, a coherent experimental framework within which to cast such dynamics is not forthcoming despite detailed experimental efforts. Drug combination studies with D_1 and D_2 compounds by Williams *et al.* (1990) emphasize the complexity of these interactions, present even with agents with restricted pharmacologies. Directly pertinent to the present problem, SKF 38393 did not alter the discriminative stimulus effects of cocaine (Callahan *et al.*, 1991). Thus, the possible interactive effects of stimulation of D_1 and D_2 receptors in the discriminative stimulus effects of cocaine remains to be determined.

Despite differences in structure and pharmacological activity, all of the D_1 agonists behaved similarly in this assay as did those compounds studied with D_2 agonist activity. In rats with 6-hydroxydopamine lesions of the corpus striatum, both D1 and D2 agonists induce rotation. The correlation between the potency of the compounds to induce rotation and the discriminative stimulus effects of cocaine was positive ($r = 0.832$, $P < .05$; rotation data from Winkler and Weiss, 1989; seven compounds in common with the present study). Although only a few compounds overlap with the present data, the potency of drugs to inhibit [^{3}H]SCH 23390 or [^{3}H]spipirone binding is also related positively to the potency in producing partial substitution for the discriminative stimulus effects of cocaine (binding data from Arnt *et al.*, 1988). Thus, the behavioral effects of the selective dopamine compounds studied here appears to be related to *in vivo* and *in vitro* activity at the dopamine receptor subtypes.

The common effects of the D_1 agonists and of the D_2 agonists in the present study is interesting when viewed against the differential pharmacological effects of the compounds within each drug class. D_1 agonists stimulate adenylate cyclase with different degrees of efficacy relative to dopamine. For example, SKF 38393 and SKF 75670 produce 69 and 33%, respectively, of the maximal dopamine-induced stimulation (*cf.* Arnt *et al.*, 1988). The newly synthesized D_1 agonist dihydrexidine fully stimulates rat striatal adenylate cyclase (Brewster *et al.*, 1990). However, despite this full efficacy, dihydrexidine did not substitute fully for the discriminative stimulus effects of cocaine. In fact, SKF 75670, which only weakly stimulates cyclase activity, had the greatest efficacy in substituting for cocaine. Unlike other D_2 agonists, (−)-NPA and pergolide, also stimulate adenylate cyclase and induce oral stereotypies (Arnt *et al.*, 1988; Wong and Reid, 1980). Again, despite the unique pharmacological profile for these D_2 agonists, they both induced only a partial substitution for cocaine which was similar to the other D_2 agonists. The only major difference observed across compounds within a drug class was with the D_2 agonists N-

0434 and N-0437. These potent drugs produced significant discriminative stimulus effects across a range of doses that did not substantially alter rates of responding. The other D_2 agonists only produced significant discriminative effects at doses that disrupted ongoing rates of responding.

The lack of substitution of cocaine methiodide, a permanently charged molecule which cannot readily penetrate the central nervous system, for cocaine provided evidence that peripheral receptors are not critical to the discriminative stimulus effects of cocaine. McKenna and Ho (1980) reported similar findings but, in their study, only 20 mg/kg of cocaine methiodide was investigated. However, cocaine methiodide is about 10 times less potent than cocaine in its noradrenergic and local anesthetic properties (Shriver and Long, 1971; Tessel *et al.*, 1978) and 30 times less potent than cocaine in binding to the dopamine transporter (Ritz *et al.*, 1990). Thus, sufficient doses of the compound need to be investigated to fully compare its effects to those of cocaine. In the present study, cocaine methiodide did not substitute for cocaine when given up to 56 mg/kg, a dose 56 times higher than the minimal effective dose of cocaine. The D_1-selective compound fenoldopam, which penetrates the central nervous system in a limited manner, was less efficacious than the other D_1 agonists investigated. These data provide further evidence for the importance of central mechanisms in the discriminative stimulus effects of cocaine (*cf.* Wood *et al.*, 1986).

The stereospecificity of the discriminative stimulus effects of cocaine has been demonstrated previously. Katz *et al.* (1990) showed that (+)-cocaine did not substitute for cocaine in doses that were 60-fold higher than the ED_{50} for the (−)-enantiomer. In the present study, *S*-(−)-SKF 38393, a compound relatively devoid of affinity for D_1 receptors, was without appreciable efficacy in substituting for cocaine. In contrast, the active *R*-(+)-isomer was both more potent and displayed marked efficacy when substituted for cocaine. Although more potent than (±)-SKF 38393, *R*-(+)-SKF 38393 displayed equivalent efficacy indicating that the (−)-enantiomer did not inhibit maximal efficacy of the racemate.

In summary, results of the present investigation suggest that the discriminative control of behavior by cocaine in rats is not an exclusive consequence of D_1 or D_2 receptor stimulation. Although data exist to suggest that D_1 and D_2 receptors may have a differential role in other behavioral and cardiovascular effects of cocaine, these effects are often small, dependent upon the intrinisic activity of the antagonist, or highly specific to the behavioral or physiological parameter (Schindler *et al.*, 1991; Katz and Witkin, 1991; Witkin *et al.*, 1991). Additional pharmacological mechanisms of action of the behavioral effects of cocaine are likely to be uncovered. Nonetheless, the ability of SCH 23390 to alter some of the behavioral effects of cocaine, to antagonize its discriminative and reinforcing effects and to attenuate its lethality (*cf.* Johanson and Fischman, 1989; Kleven *et al.*, 1988, 1990; Katz and Witkin, 1991; Witkin *et al.*, 1989, 1991) suggests that D_1 selective compounds should be evaluated clinically as potential prophylactic agents in certain aspects of cocaine abuse and toxicity.

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Send reprint requests to: J. M. Witkin, Ph.D., Psychobiology Laboratory, NIDA ARC, P.O. Box 5180, Baltimore, MD 21224.