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Behavioral effects of AMI-193, a 5-HT_{2A}- and dopamine D_2-receptor antagonist, in the squirrel monkey

Paul W. Czoty, Leonard L. Howell*

Yerkes Regional Primate Research Center, Emory University, Atlanta, GA 30322, USA

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Abstract

8-[3-(4-Fluorophenoxy) propyl]-1-phenyl-1,3,8-triazaspiro[4,5]decan-4-one (AMI-193) was developed as a 5-HT_{2A}-selective antagonist with in vivo activity suitable for behavioral studies. However, AMI-193 is a potent dopamine D_2-receptor antagonist with low nanomolar affinity. Accordingly, D_2-actions may contribute to its behavioral pharmacology. In the present study, the effects of AMI-193 on operant behavior were characterized in squirrel monkeys. In subjects trained under a fixed-interval (FI) schedule of stimulus termination, AMI-193 (0.003–0.01 mg/kg) dose-dependently decreased response rate. When administered in combination with cocaine (0.03–3.0 mg/kg) or the selective dopamine uptake inhibitor, GBR 12909 (0.03–3.0 mg/kg), the rate-decreasing effects of AMI-193 were reversed by both dopamine indirect agonists. In drug-discrimination experiments, AMI-193 (0.003 and 0.01 mg/kg) attenuated the discriminative-stimulus effects of cocaine. AMI-193 (0.003 and 0.01 mg/kg) also reduced response rate under a second-order schedule of i.v. self-administration of cocaine (0.1 mg/infusion). The profile of behavioral effects and drug interactions observed in the present study, in conjunction with the relatively high affinity of AMI-193 for dopamine D_2 receptors, suggests that its D_2-antagonist effects play a prominent role in the behavioral pharmacology of AMI-193. © 2000 Elsevier Science Inc. All rights reserved.

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Understanding the function of individual subtypes of brain 5-HT_2 receptors is currently hindered by the scarcity of adequate pharmacological tools [2,14]. While several drugs are available to discriminate 5-HT_2 receptors from other 5-HT receptor classes, few drugs exist that show sufficient selectivity among the members of the 5-HT_2 family [5-HT_{2A}, 5-HT_{2B}, and 5-HT_{2C}; for review see Ref. [19]]. Although 5-HT_{2B} receptor mRNA has been found in mouse [26] and human [5,25,30] brain, its function and distribution in the rat brain are not well understood [11]. Accordingly, more effort has been directed at developing compounds that show selectivity for 5-HT_{2A} and 5-HT_{2C} subtypes. Spiperone, the ligand historically used to identify 5-HT_2 receptors in rat brain [28], has been frequently used to discriminate 5-HT_{2A} from 5-HT_{2C} receptors. However, spiperone has high affinity for dopamine D_2 receptors [20], and it elicits dramatic rate-decreasing effects that can be disruptive in behavioral studies [13,16]. 8-[3-(4-Fluorophenoxy) propyl]-1-phenyl-1,3,8-triazaspiro[4,5]decan-4-one (AMI-193) is a spiperone derivative that has twice the selectivity for 5-HT_{2A} versus 5-HT_{2C} receptors compared to spiperone, and its affinity for dopamine D_2 receptors is 15-fold less than that of spiperone [20]. Its ability to serve as a functional 5-HT_{2A} antagonist in behavioral studies was demonstrated through studies in which AMI-193 blocked the discriminative-stimulus effects of the $5HT_{2A}$ agonist 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (DOM) [20]. However, the low nanomolar affinity of AMI-193 for D_2 receptors may still be sufficient to interfere with its 5-HT_{2A}-antagonist effects, particularly in behavioral studies that involve dopamine systems.

The ability of 5-HT to exert inhibitory modulation on brain dopamine systems has been well documented [10,12,18,21,31], and pharmacological studies have implicated 5-HT_2 receptors in this modulation [32,36]. Consistent with these findings, studies in squirrel monkeys have

* Corresponding author. Tel.: +1-404-727-7786; fax: +1-404-727-1266.

E-mail address: leonard@rmy.emory.edu (L.L. Howell).

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suggested a prominent role for 5-HT_2 receptors in the ability of brain 5-HT systems to modulate the behavioral-stimulant and reinforcing effects of cocaine [17]. For example, behaviorally inactive doses of quipazine, a 5-HT direct agonist with high affinity at 5-HT_2 receptors, caused an insurmountable attenuation of the behavioral-stimulant effects of cocaine in a manner similar to that of the 5-HT uptake inhibitors alaproclate, citalopram, and fluoxetine [17,34]. Conversely, the nonselective 5-HT antagonist mianserin and the 5-HT_2-selective antagonists ketanserin and ritanserin increased the behavioral-stimulant effects of cocaine, whereas 5-HT_1-selective (NAN-190) and 5-HT_3-selective (MDL 72222) antagonists did not. Ritanserin administration also increased response rate in monkeys self-administering cocaine [17] or the selective dopamine uptake inhibitor, GBR 12909 [18]. The ability of serotonergic manipulations to modulate the discriminative-stimulus effects of cocaine in rats [8,9,24] and monkeys [29,34] is likewise documented.

The present experiments were conducted to characterize the behavioral pharmacology of AMI-193 in nonhuman primates. Drug interactions with dopamine indirect agonists were used to elucidate the involvement of 5-HT_{2A}- and dopamine D_2-antagonist effects of AMI-193 in vivo. Studies examined the direct rate-altering effects of AMI-193 on schedule-controlled behavior, as well as the ability of AMI-193 to modulate the behavioral-stimulant effects of cocaine and GBR 12909, and the discriminative-stimulus and reinforcing effects of cocaine. The results suggest that D_2 antagonism plays a prominent role in the behavioral pharmacology of AMI-193.

1. Material and methods

1.1. Subjects

Fourteen adult male squirrel monkeys (*Saimiri sciureus*), weighing 850–1300 g, served as subjects. Between daily experimental sessions, subjects lived in individual home cages and had access to food (Harlan Teklad monkey diet, fresh fruit, and vegetables) and water. Monkeys S122, S124, S126, S130, S135, S137, and S138 were surgically prepared with a chronically indwelling venous catheter for i.v. administration of drugs. Anesthesia was induced with Telazol and maintained with ketamine hydrochloride. Polyvinyl chloride tubing (0.38 mm internal diameter, 0.76 mm outer diameter) was inserted into the left or right femoral or external jugular vein using sterile surgical techniques. The distal end of the catheter was passed subcutaneously and exited between the scapulae. The catheter was filled with heparinized saline (20 U/0.2 ml saline) and sealed with a stainless steel obturator when not in use. A nylon-mesh jacket protected the externalized end of the catheter. All animals had served as subjects in previous experiments involving acute administration of drugs [17,18,29]. All animal use procedures were in strict accordance with the NIH "Guide for the Care and Use of Laboratory Animals" and were approved by the Institutional Animal Care and Use Committee at Emory University.

1.2. Apparatus

Experimental test sessions were conducted daily within a ventilated, sound-attenuating chamber with each subject seated in a Plexiglas chair [6]. Illumination was provided by one of three pairs of 7-W a.c. colored lights (red, white, and blue) mounted on the front of the chair just above eye level. A response lever mounted on the chair, facing the monkey, registered a response and operated an auditory feedback relay when depressed with a downward force greater than 0.2 N. The monkey's tail was held motionless in a small stock. In experiments using a stimulus-termination schedule, two brass plates rested on a shaved portion near the end of the monkey's tail. Electrode paste applied to the tail minimized changes in impedance between the tail and the brass plates when a 3-mA electric stimulus was delivered. In drug discrimination experiments, subjects faced two retractable response levers placed 10 cm apart on a horizontal plane. A food reservoir mounted on the front of the chair delivered 190-mg sucrose pellets (P.J. Noyes, Lancaster, NH) into a tray positioned between the levers. In drug self-administration experiments, the distal end of the catheter was connected via polyvinyl tubing to a syringe located outside the test chamber. The syringe was driven by a 110-V a.c. motor that was controlled by electronic circuitry to yield a precise injection volume of 0.2 ml. Sessions were conducted 5 days/week and each session lasted approximately 80 min for the fixed-interval (FI) stimulus-termination schedule, 30 min for drug-discrimination sessions, and 65 min for the second-order stimulus-termination and self-administration schedules. Continuous white noise and an exhaust fan masked external sounds during all test sessions.

1.3. Procedure

1.3.1. FI stimulus termination schedule

Monkeys S122, S130, S135, and S137 were trained under a FI 300-s schedule of stimulus termination [27]. Red lights illuminated the test chamber during the fixed interval and, after 300 s elapsed, the animal had 3 s to press a lever and terminate the lights that were associated with an impending electric stimulus. When the monkey pressed the lever during the limited hold and terminated the red lights, white lights were illuminated for 2 s, followed by a 60-s timeout during which the chamber was dark and responding had no scheduled consequences. In the absence of a response during the 3-s limited hold, a 3-mA stimulus was delivered once for 200 ms, followed by a 60-s timeout. A daily session consisted of 13 consecutive FI 300-s components, each followed by a 60-s timeout.

1.3.2. Drug discrimination schedule

Procedural details of drug discrimination experiments have been described previously [29]. Briefly, monkeys S86, S116, and S134 were trained to press a response lever under a FR 30 schedule of food reinforcement. During training, subjects received either 0.3 mg/kg cocaine or saline i.m. 10 min presession. Sessions began with red and blue lights illuminated and both levers extended. Responses on the lever associated with the presession injection were reinforced with a food pellet on an FR 30 schedule, while responses on the other lever were not reinforced. Each pellet delivery was accompanied by illumination of white lights for 15 s and retraction of the levers, and was followed by a 60-s timeout. A daily session consisted of 30 FR 30 components. AMI-193 pretreatment or substitution experiments were conducted on Tuesday and Friday provided that the previous two sessions had produced greater than 90% correct responding. During pretreatment sessions, AMI-193 was administered 30 min presession and cocaine (0.03–1.0 mg/kg) was administered 10 min presession. During substitution sessions, AMI-193 was administered 30 min presession. In either case, a food pellet was delivered after the first FR 30 completed on either lever. The session ended after 1 FR had been completed or after 30 min had expired.

1.3.3. Second-order drug self-administration schedule

Monkeys S124, S126, and S138 were trained under a second-order FI 900-s (FR 20:s) schedule of i.v. drug delivery. Red lights illuminated the chamber during the 900-s fixed interval, and every 20th response (FR 20) changed the lights from red to white for 2 s. After 900 s elapsed, the animal had 300 s to complete an FR 20 and receive a drug injection. When the monkey completed an FR 20 during the limited hold, white lights were illuminated for 15 s, followed by a 60-s timeout during which the chamber was dark and responding had no scheduled consequences. If an FR 20 was not completed during the 300-s limited hold, no drug was delivered and a 60-s timeout followed. A daily session consisted of four consecutive FI 900-s components, each followed by a 60-s timeout.

1.3.4. Second-order stimulus-termination schedule

Monkeys S91, S98, S104, and S130 were trained under a second order, FI 900-s (FR 20:s) schedule of stimulus termination. Red lights illuminated the chamber during the 900-s fixed interval, and every 20th response (FR 20) changed the lights from red to white for 2 s. After 900 s elapsed, the animal had 20 s to complete an FR 20 and terminate the red lights that were associated with the impending electrical stimulus. When the monkey completed an FR 20 during the limited hold and terminated the red lights, white lights were illuminated for 15 s, followed by a 60-s timeout during which the chamber was dark and responding had no scheduled consequences. If an FR 20 was not completed during the 20-s limited hold, a 3-mA stimulus was delivered once for 200 ms, followed by a 60-s timeout. A daily session consisted of four consecutive FI 900-s components, each followed by a 60-s timeout.

1.4. Drug administration

AMI-193 was administered i.m. as an injection in the thigh muscle in a volume of 0.4–0.8 ml 30 min presession. In drug interaction studies using the FI 300 schedule, cocaine (0.03–3.0 mg/kg) or GBR 12909 (0.03–3.0 mg/kg) was subsequently administered i.v. using a cumulative-dosing procedure similar to that described by Kelleher and Goldberg [22] and Wenger [37]. A complete dose–effect curve was established in a single session by injecting graded doses i.v. during the timeouts preceding component 2, 5, 8, and 11 [15]. Typically, drug experiments were conducted on Tuesday and Friday and saline (control) was administered on Thursday. In studies using the second-order schedule of drug self-administration or stimulus termination, saline was administered as a pretreatment on Tuesday, Wednesday, and Thursday, and AMI-193 (0.003 and 0.01 mg/kg) was administered on Tuesday, Wednesday, and Thursday the following week. Each drug dose or drug combination was studied at least twice in each monkey, and experiments were conducted in a quasi-random order to preclude changes in drug sensitivity due to order of administration.

1.5. Data analysis

Response rate maintained by the FI 300-s schedule of stimulus termination was computed by dividing the total number of responses in a component by the total time the red light was present. Mean control rate was determined for each monkey by averaging response rate for all saline (control) sessions. Mean rate of responding during cumulative-dosing studies was determined for each monkey by averaging data from selected individual FI components following administration of an i.v. dose. Although three components followed the infusion of each dose, only the last two were used to determine the mean response rate to compensate for the time needed for absorption and distribution of the drug. Mean response rate maintained by the second-order FI 900-s schedule of stimulus termination was determined for individual monkeys by averaging response rates from all FI components of a session. For both stimulus-termination schedules, group data were derived and expressed as the mean ± SEM for the group based on the percentage of change in the rate of responding during drug sessions compared to sessions in which saline was administered. In drug discrimination experiments, percent responding on the cocaine lever for individual animals was computed by dividing the number of responses on the cocaine-associated lever by the total

responses during the test session and multiplying by 100. Data are expressed as group means ± SEM. Mean rate of responding during drug self-administration studies was determined for individual monkeys by averaging response rate in the presence of the red light during all components of a session. Data are presented as mean response rate ± SEM during three consecutive sessions when a particular pretreatment dose was administered, expressed as a percentage of the mean response rate when saline was administered as a pretreatment for 3 consecutive days during the previous week. A repeated-measures analysis of variance with Tukey's post hoc multiple comparisons was used to determine the statistical significance of drug treatment conditions. Statistical significance was accepted at the 95% level of confidence ($p < 0.05$).

1.6. Drugs

The drugs studied were AMI-193 (generously supplied by Dr. Richard A. Glennon, Department of Medicinal Chemistry, Virginia Commonwealth University, Richmond, VA), cocaine HCl (National Institute on Drug Abuse, Rockville, MD) and GBR 12909 (Research Biochemicals, Natick, MA). AMI-193 and cocaine were dissolved in 0.9% saline, and doses were determined in terms of the salts. GBR 12909 was dissolved in a small volume of 95% alcohol and emulphor, and diluted with 0.9% saline.

2. Results

2.1. FI stimulus-termination schedule

Rates and patterns of lever-pressing were characteristic of typical performance engendered by FI schedules in squirrel monkeys. Response rates were low at the beginning of each FI and increased as the interval progressed. In a group of three monkeys, mean ± SEM rate of responding following saline administration was 0.71 ± 0.16 responses/s. Time-course analyses of intra-session response rates revealed that the effects of AMI-193 were consistent over the 13 FI components in each session. Thus, data were collapsed across the 13 components of each session. The i.m. administration of 0.003 mg/kg AMI-193 had no significant effect on response rates, while 0.01 mg/kg AMI-193 significantly ($p < 0.001$) decreased response rates (Fig. 1, top). When administered i.v. using a cumulative-dosing procedure, cocaine (0.03–3.0 mg/kg) produced a characteristic inverted U-shaped dose–effect curve (Fig. 1, top). When cocaine was administered following pretreatment with AMI-193, the rate-decreasing effects of AMI-193 were reversed. Also, there was a trend toward shifting the cocaine dose–effect curve to the right at the highest dose of AMI-193, but the interaction was not statistically significant. In a separate group of three monkeys, response rate was significantly decreased by 0.003 mg/kg AMI-193

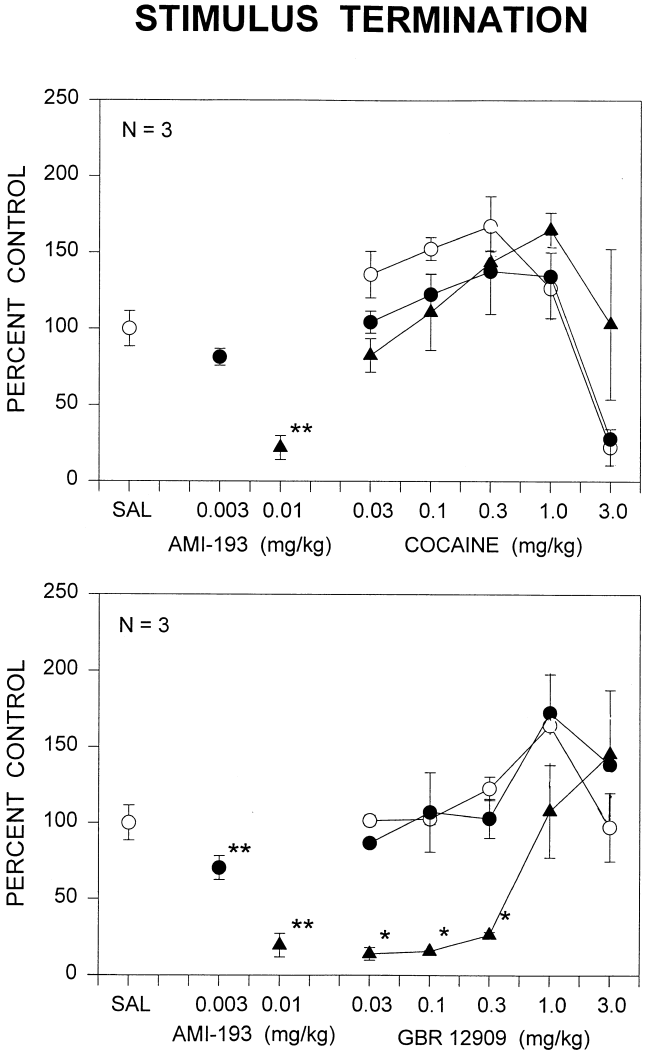

Fig. 1. Effects of cocaine (0.03–3.0 mg/kg; top) and GBR 12909 (0.03–3.0 mg/kg, bottom) administered alone and in combination with AMI-193 (0.003 and 0.01 mg/kg) on mean ± SEM response rates maintained by a FI 300-s schedule of stimulus termination in two groups of three monkeys. The effects of AMI-193 administered alone are shown with the filled symbols to the left of the cocaine and GBR 12909 dose–effect curves. Abscissae: dose, log scale. Ordinates: mean ± SEM response rates expressed as a percentage of control rates obtained when saline (SAL) was administered. * Significant difference ($p < 0.05$) from response rate obtained after administration of GBR 12909 alone. * Significant difference ($p < 0.05$) from control rates obtained when saline was administered.

($p < 0.05$) and 0.01 mg/kg AMI-193 ($p < 0.01$). When administered i.v. using a cumulative-dosing procedure, the selective dopamine uptake inhibitor, GBR 12909 (0.03–3.0 mg/kg), produced a characteristic inverted U-shaped dose–effect curve (Fig. 1, bottom). When GBR 12909 was administered following pretreatment with AMI-193, the rate-decreasing effects of AMI-193 were reversed. As seen with cocaine, there was a trend toward shifting the GBR 12909 dose–effect curve to the right at the highest dose of AMI-193, but the interaction was not statistically significant.

2.2. Drug discrimination schedule

Substitution with different doses of cocaine (0.03–1.0 mg/kg) engendered increases in responding on the cocaine-associated lever in a dose-related manner (Fig. 2). The 0.3 mg/kg training dose occasioned 100% responding on the cocaine-associated lever, whereas saline occasioned 99.1% responding on the saline-associated lever. When administered alone 30 min presession, 0.003 and 0.01 mg/kg AMI-193 occasioned responding almost exclusively on the saline-associated lever (< 5% cocaine-appropriate responding). Response rates for AMI-193 alone did not differ significantly from those obtained following the training dose of cocaine. When administered in combination with cocaine, AMI-193 antagonized the discriminative-stimulus effects of cocaine, shifting the cocaine dose–effect curve to the right. Response rates obtained during drug-interaction experiments were not significantly different from rates obtained when respective doses of cocaine were administered alone. Statistical analysis revealed a significant main effect of drug pretreatment ($p < 0.01$) and a significant interaction ($p < 0.001$). Doses of cocaine (0.01 and 0.3 mg/kg) which engendered 81.3% and 100.0% cocaine-appropriate responding, respectively, elicited only 13.4% and 61.3% cocaine-appropriate responding when administered in combination with 0.01 mg/kg AMI-193.

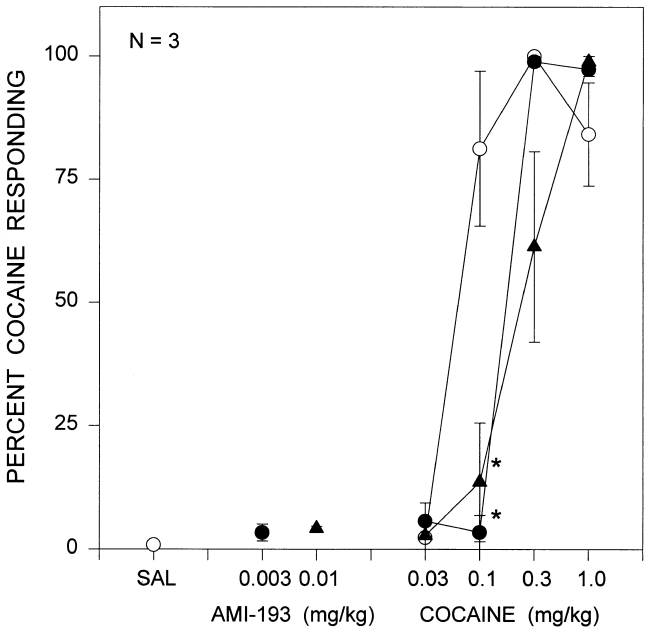

Fig. 2. Discriminative-stimulus effects of cocaine (0.03–1.0 mg/kg) and AMI-193 (0.003 and 0.01 mg/kg) in a group of three monkeys trained to discriminate 0.3 mg/kg cocaine and saline (SAL) under a FR 30 schedule. Percent responding on the cocaine lever was computed by dividing the number of total responses on the cocaine-associated lever by the total responses during the test session, and multiplying by 100. Abscissa: dose, log scale. Ordinate: mean ± SEM percent cocaine-lever responding. * Significant difference ($p < 0.05$) from percent cocaine responding obtained after the corresponding dose of cocaine administered alone.

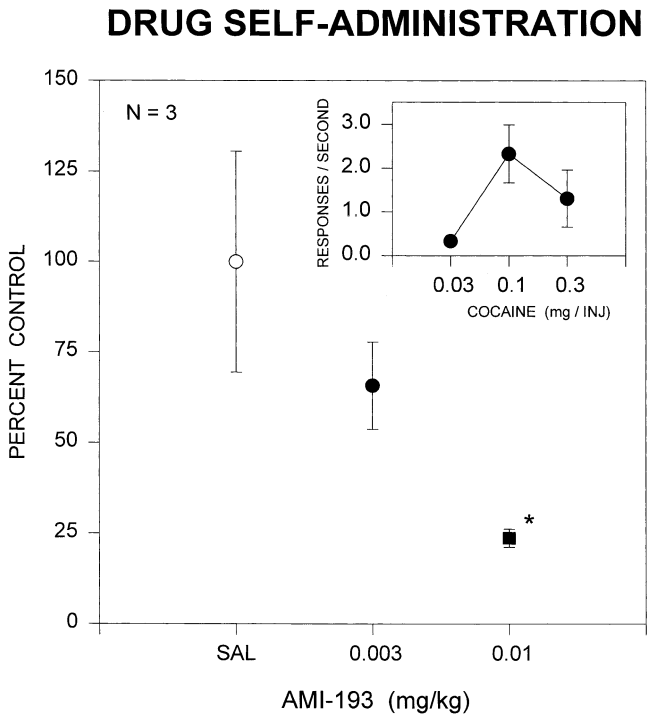

Fig. 3. Mean ± SEM rate of responding maintained by a second-order FI 900-s (FR 20:s) schedule of i.v. self-administration of cocaine (0.1 mg/injection) after pretreatment with AMI-193 (0.003 and 0.01 mg/kg) in a group of three monkeys. Abscissa: dose, log scale. Ordinates: mean ± SEM response rate expressed as a percent of control response rates obtained when cocaine was self-administered after saline pretreatment. Open symbol indicates mean ± 95% confidence limits for cocaine self-administration following saline pretreatment (control). * Significant difference ($p < 0.05$) when compared to control response rate. Inset: mean rate of responding maintained by i.v. injection of cocaine (0.03–0.3 mg/injection) under an FI 900-s (FR 20:s) schedule in a group of three monkeys. Data for each cocaine dose were derived from at least 15 consecutive sessions. Abscissae: dose, log scale. Ordinates: mean ± SEM response rate expressed as responses per second.

2.3. Second-order drug self-administration schedule

Rates and patterns of responding were characteristic of typical performance engendered by second-order schedules of drug self-administration in squirrel monkeys. During each component, responding commenced following a variable delay and was maintained by the brief presentation of white lights after the completion of each FR 20. A steady, high rate of lever-pressing characterized the FR 20 components. Mean ± SEM rate of responding at the maintenance dose (0.1 mg/infusion) of cocaine was 2.33 ± 0.66 responses/s. Substitution with two other doses (0.03 and 0.3 mg/infusion) resulted in a decrease in response rate,

leading to an inverted U-shaped dose–effect curve typical of stimulants such as cocaine (Fig. 3, inset). Pretreatment with 0.01 mg/kg AMI-193 decreased response rates for the maintenance dose of cocaine (0.1 mg/infusion) compared to days during which saline was given as a pretreatment (Fig. 3). No trend toward progressively increasing or decreasing response rates was observed across each 3-day pretreatment with AMI-193. Although the lower dose of AMI-193 decreased mean response rates by approximately 35%, the effect was not statistically significant. However, pretreatment with the higher dose of AMI-193 significantly decreased response rates to less than 25% of control values ($p < 0.01$).

2.4. Second-order stimulus-termination schedule

Rates and patterns of lever-pressing maintained under a second-order schedule of stimulus termination were similar to those obtained under the second-order schedule of cocaine self-administration. Mean ± SEM rate of responding following saline administration was 0.87 ± 0.24 responses/s. There was a significant ($p < 0.001$) main effect of pretreatment with 0.003–0.01 mg/kg AMI-193, such that response rate decreased in a dose-dependent manner (Fig. 4).

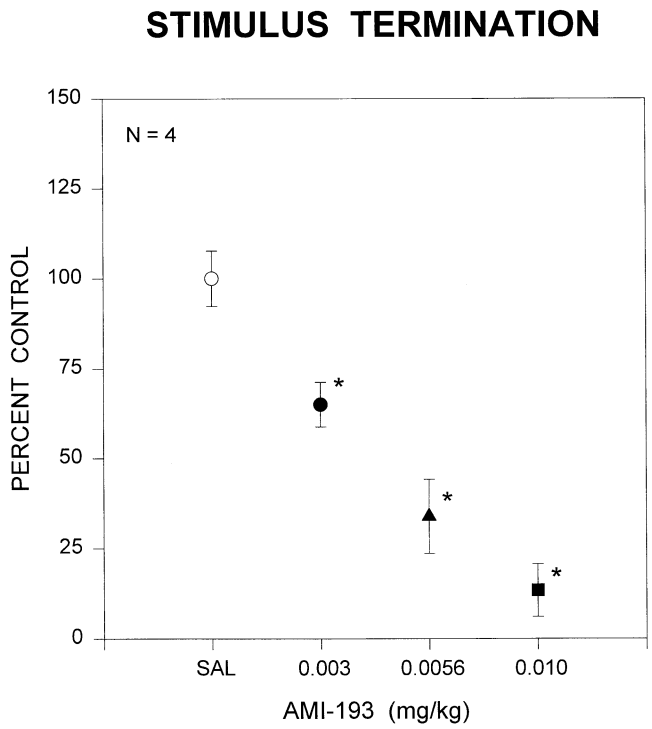

Fig. 4. Mean ± SEM rate of responding maintained by a second-order FI 900-s (FR 20:s) schedule of stimulus termination after pretreatment with AMI-193 (0.003–0.01 mg/kg) in a group of four monkeys. Open symbol indicates mean ± 95% confidence limits for stimulus termination performance following saline pretreatment (control). Otherwise, as in Fig. 3.

3. Discussion

AMI-193 was developed as a 5-HT_{2A}-selective antagonist with in vivo activity suitable for behavioral studies. It is highly selective for 5-HT_{2A} (K_i = 2 nM) versus 5-HT_{2C} (K_i = 4300 nM) receptors, and its ability to serve as a functional 5-HT_{2A} antagonist in behavioral studies was demonstrated in its blockade of the discriminative-stimulus effects of the 5-HT_{2A} agonist DOM [20]. Antagonism of D_2 receptors appeared to play no role in the interaction of AMI-193 with DOM, but DOM lacks dopaminergic effects [13]. Given that AMI-193 has comparable affinity for dopamine D_2 (K_i = 3 nM) and 5-HT_{2A} receptors, its dopamine antagonist effects may contribute significantly to the behavioral pharmacology of AMI-193.

In the present study, the effects of AMI-193 on operant behavior were characterized in nonhuman primates. Drug interactions with cocaine and the selective dopamine uptake inhibitor GBR 12909 were used to elucidate the involvement of 5-HT_{2A}- and dopamine D_2-antagonist effects of AMI-193 in vivo. AMI-193 had pronounced rate-decreasing effects when administered alone that were reversed by both dopamine indirect agonists. The higher dose of AMI-193 also tended to shift the cocaine and GBR 12909 dose–effect curves rightward, although the latter effects were not statistically significant. These results contrast with those reported previously for the 5-$HT_{2A/2C}$ antagonists ritanserin and ketanserin. Both antagonists enhanced the behavioral-stimulant effects of cocaine [17] and GBR 12909 [18] in squirrel monkeys under identical protocols. The pronounced behavioral-depressant effects of AMI-193 administered alone and the ability of cocaine and GBR 12909 to reverse the rate-decreasing effects of AMI-193 are consistent with the pharmacological profile of D_2 antagonists reported previously in squirrel monkeys [4,15,16,33].

Consistent with its ability to modify the behavioral-stimulant effects of cocaine and GBR 12909, AMI-193 antagonized the discriminative-stimulus effects of cocaine in a manner similar to D_2-receptor antagonists. In drug-discrimination experiments, AMI-193 shifted the dose–effect curve for cocaine to the right. The rightward shift elicited by AMI-193 is characteristic of D_2 antagonist effects on the discriminative-stimulus effects of cocaine in rats [1] and monkeys [23,35]. However, 5-HT_{2A} antagonist effects cannot be ruled out since both ketanserin and ritanserin have been shown to attenuate the discriminative-stimulus effects of cocaine in a similar manner [29]. Lastly, pretreatment with AMI-193 dose-dependently decreased response rate in monkeys trained to self-administer cocaine intravenously. Again, this result is more characteristic of a D_2 antagonist effect in rats [7] and monkeys [3,38] than it is of a 5-HT_2 antagonist such as ritanserin, which selectively increased response rate in squirrel monkeys self-administering cocaine [17]. The effects of AMI-193 on cocaine self-administration were not selective in that the

same doses of AMI-193 also dose-dependently decreased response rate maintained by an identical schedule of stimulus termination. Hence, the rate-decreasing effects of AMI-193 were observed under two different operant schedules of stimulus termination (FI 300-s; second-order FI 900-s/FR 20:s) and under two schedules with identical parameters maintained by different reinforcers (cocaine infusion; stimulus termination).

In summary, AMI-193 had pronounced rate-decreasing effects that were reversed by two dopamine indirect agonists. AMI-193 also attenuated the discriminative-stimulus effects of cocaine, and decreased response rate on a second-order schedule of intravenous cocaine self-administration. The profile of behavioral effects and drug interactions observed in the present study, in conjunction with the relatively high affinity of AMI-193 for D_2 dopamine receptors, suggests that D_2 antagonism plays a prominent role in the behavioral pharmacology of AMI-193.

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