ABSTRACT

Cocaine is a nonselective monoamine reuptake inhibitor that is widely abused. Useful pharmacotherapies for cocaine dependence may include substitution medications that produce cocaine-like effects but have a slower onset and longer duration of action. Accordingly, the present study examined the effects of the long-acting, nonselective monoamine reuptake inhibitor indatraline in assays of cocaine discrimination and cocaine self-administration that have been used to evaluate other candidate treatment medications. In rhesus monkeys trained to discriminate cocaine (0.4 mg/kg) from saline, indatraline (0.1–1.0 mg/kg) produced a dose- and time-dependent substitution for cocaine. The effects of 1.0 mg/kg indatraline peaked after 30 min and lasted up to 24 h. In monkeys trained to self-administer 0.032 mg/kg/injection cocaine and food pellets during alternating daily sessions of cocaine and food availability, indatraline (0.0032–0.032 mg/kg/injection) maintained lower rates of responding than cocaine. Repeated treatments with indatraline (0.1–0.56 mg/kg/day) for 7 days produced dose-dependent and sustained decreases in cocaine self-administration across a broad range of cocaine doses (0.0032–0.1 mg/kg/injection), and the highest dose of indatraline (0.56 mg/kg/day) nearly eliminated cocaine-maintained responding. However, doses of indatraline that decreased cocaine self-administration also usually decreased rates of food-maintained responding and produced behavioral stereotypies and trends toward weight loss and mild anemia. These findings suggest that although indatraline may decrease cocaine-taking behavior in rhesus monkeys, it also produces undesirable side effects that may limit its clinical utility in the treatment of cocaine dependence.

Effects of the Long-Acting Monoamine Reuptake Inhibitor Indatraline on Cocaine Self-Administration in Rhesus Monkeys

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Cocaine abuse continues to be a serious public health problem, and there are currently no uniformly effective medications for its treatment (Mendelson and Mello, 1996). The utility of oral methadone as a substitution medication for the treatment of opioid dependence (Ball and Ross, 1991) suggests that a relatively slow-onset, long-acting cocaine-like medication may be useful in the treatment of cocaine abuse. Cocaine nonselectively blocks the reuptake of dopamine, norepinephrine and serotonin (Koe, 1976), and compelling evidence suggests that the effects of cocaine on dopaminergic systems are especially important in mediating its high abuse potential (Koob and Bloom, 1988). Consequently, selective dopamine reuptake inhibitors are currently being evaluated as one class of potential substitution medications for the treatment of cocaine abuse (Mansbach and Balster, 1993; Glowa et al., 1995a; Nader et al., 1997; Dworkin et al., 1998). However, the effects of cocaine on norepinephrine and serotonin reuptake may also contribute to cocaine’s abuse-related effects (Terry et al., 1994; Callahan and Cunningham, 1995; Spealman, 1995). As a result, it may also be instructive to assess the utility of nonselective monoamine reuptake inhibitors as potential substitution medications.

Indatraline (also known as Lu 19–005) is one such nonselective monoamine reuptake inhibitor (Bogeso et al., 1985; Hyttel and Larsen, 1985). In vitro, indatraline blocked the reuptake of dopamine, norepinephrine, and serotonin into rat brain synaptosomes with similar nanomolar potencies (Hyttel and Larsen, 1985). In vivo, indatraline produced behavioral effects suggestive of enhanced dopaminergic, noradrenergic, and serotonergic activity in mice, and these effects were all observed at similar indatraline doses (Arnt et al., 1985). Indatraline also substituted for a high dose of cocaine in rats trained to discriminate between a low and a high dose of cocaine, which demonstrates that indatraline produces cocaine-like discriminative stimulus effects (Kleven and Koek, 1998). In comparison with cocaine, however, indatraline has been reported to have a slow onset and long duration of action (Arnt et al., 1985; Rosenzweig-Lipson et al., 1992). In squirrel monkeys, for example, i.m. administration of either cocaine or indatraline produced a dose-dependent increase in response rates maintained under fixed interval schedules of operant responding. However, the effects of cocaine peaked after approximately 10 min and diminished after 1 h, whereas the effects of indatraline peaked after
Subjects

chemistry.

study examined the effects of repeated treatment with saline cocaine was studied for 7 consecutive days. It became appar-

bination of a treatment dose of indatraline and a unit dose of indatraline (0.1–0.56 mg/kg/day) during availability of dif-

edly with noncontingent administration of either saline or indatraline on cocaine self-administration and concurrent available for self-administration. To examine the effects of tion) for the maintenance dose of cocaine as the drug solution mg/kg/injection) or indatraline (0.0032–0.032 mg/kg/injec-

reinforcing effects of indatraline and cocaine were compared by substituting different unit doses of cocaine (0.001–0.1 mg/kg/injection) or indatraline (0.0032–0.032 mg/kg/injec-

for the maintenance dose of cocaine as the drug solution available for self-administration. To examine the effects of indatraline on cocaine self-administration and concurrent food-maintained responding, monkeys were treated repeat-
edly with noncontingent administration of either saline or indatraline (0.1–0.56 mg/kg/day) during availability of dif-

cerent cocaine doses (0.0032–0.1 mg/kg/injection). Each com-

bination of a treatment dose of indatraline and a unit dose of cocaine was studied for 7 consecutive days. It became appar-

tent during these drug self-administration studies that re-

peated administration of indatraline produced stereotypic behaviors in some monkeys. To evaluate potential toxic ef-

ccts associated with chronic indatraline treatment, a final study examined the effects of repeated treatment with saline or indatraline (0.32–1.0 mg/kg/day for 7 days) on overt be-

havior, body weight, and indices of hematology and blood chemistry.

Materials and Methods

Subjects

A total of 12 rhesus monkeys (Macaca mulatta) were subjects in three studies designed to to characterize the behavioral pharmacol-

gy of indatraline. Four male rhesus monkeys were studied in the drug discrimination experiments, two female and three male rhesus monkeys were studied in the drug self-administration experiments, and one female and two male monkeys were studied in observational studies. All monkeys had an experimental history involving the evaluation of dopaminergic and/or opioid compounds in assays of drug discrimination, drug self-administration, or thermal nocicep-

tion. Monkeys weighed 5.4 to 11.4 kg and were maintained on a diet of multiple vitamins, fresh fruit, and Lab Diet Jumbo Monkey bis-

cuits (PMI Feeds, Inc., St. Louis, MO). In addition, monkeys in drug discrimination and drug self-administration studies could receive 1-g banana-flavored pellets (Precision Primate Pellets Formula L/l Ba-

nana Flavor; P. J. Noyes Co., Lancaster, NH) during daily operant sessions (see below). Water was continuously available. A 12 h light/
dark cycle was in effect (lights on from 7:00 AM to 7:00 PM).

Animal maintenance and research were conducted in accordance with the guidelines provided by the National Institutes of Health Committee on Laboratory Animal Resources. The facility was li-

censed by the United States Department of Agriculture and protocols were approved by the Institutional Animal Care and Use Committee. The health of the monkeys was periodically monitored by consulting veterinarians. Monkeys had visual, auditory, and olfactory contact with other monkeys throughout the study. Operant procedures and foraging toys provided an opportunity for environmental manipula-

Drug Discrimination Procedures

Apparatus. Each monkey was housed individually in a well-

ventilated, stainless steel chamber (56 × 71 × 69 cm). The home cages of all monkeys were modified to include an operant panel (28 × 28 cm) mounted on the front wall. Three square translucent response keys (6.4 × 6.4 cm) were arranged 2.54 cm apart in a horizontal row 3.2 cm from the top of the operant panel. Each key could be transil-

luminated by red or green stimulus lights (Superbright LEDs). In addition, three circular translucent panels (1.9 cm in diameter) were located in a vertical column below the center response key and could be transilluminated by red or green stimulus lights (Superbright L.E.D.s). The operant panel also supported an externally mounted pellet dispenser (Gerbrands, Model G5210) that delivered 1-g food pellets to a food receptacle mounted on the cage beneath the operant response panel. Operation of the operant panels and data collection were accomplished with microprocessors and software provided by Med Associates Inc. (Georgia, VT).

Discrimination Training. Drug discrimination procedures were identical with those used in our previous studies of the effects of opioids and dopamine antagonists on cocaine self-administration (e.g., Lamas et al., 1995; Negus et al., 1995, 1996). Discrimination sessions consisted of multiple cycles and were conducted 5 days/ week. Each cycle consisted of a 15-min time-out period followed by a 5-min response period. During the time-out, all stimulus lights were off and responding had no scheduled consequences. During the re-

sponse period, the right and left response keys were transillumi-

nated red or green, and monkeys could receive up to 10 food pellets by responding under a fixed ratio (FR) 30 schedule of food presenta-

for the five monkeys, the left key was illuminated green and the right key was illuminated red. For the other two monkeys, the colors of the response keys were reversed. The center key was not illuminated at any time, and responding on the center key had no scheduled consequences. If all available food pellets were delivered before the end of the 5-min response period, the stimulus lights transilluminating the response keys were turned off, and responding had no scheduled consequences for the remainder of that response period.

On training days, monkeys were given an i.m. injection of either saline or 0.40 mg/kg cocaine 5 min after the beginning of each time-out period (i.e., 10 min before the response period). After ad-

ministration of saline, responding on only the green key (the saline-

appropriate key) produced food, whereas following administration of 0.40 mg/kg cocaine, only responding on the red key (the drug-appropri-

ate key) produced food. Responses on the inappropriate key reset the FR requirement on the appropriate key. Daily sessions consisted of one to five cycles, and if the training dose of cocaine was admin-

istered, it was administered only during the last cycle.

During the response period of each cycle, three dependent vari-

ables were determined: 1) percentage of injection-appropriate re-

sponding before delivery of the first reinforcer ([injection-appropriate responses emitted before 1st reinforcer / total responses emitted before 1st reinforcer] × 100); 2) percentage of injection-appropriate responding for the entire response period ([injection-appropriate responses emitted during response period / total responses emitted during response period] × 100); and 3) response rate (total responses
emitted during response period divided by total time stimulus lights were illuminated).

Monkeys were considered to have acquired cocaine discrimination when the following three criteria were met for seven of eight consecutive training sessions: 1) the percentage of injection-appropriate responding before delivery of the first reinforcer was greater than or equal to 80% for all cycles; 2) the percentage of injection-appropriate responding for the entire cycle was greater than or equal to 90% for all cycles; and 3) at least one pellet was earned during all training cycles.

**Discrimination Testing.** Once monkeys met criterion levels of cocaine discrimination, testing began. Test sessions were identical with training sessions except that responding on either key produced food, and indatraline or cocaine was administered as described below. Two series of experiments were conducted to characterize the effects of indatraline administered alone or as a pretreatment to cocaine.

In the first series of experiments, the time course of the effects of indatraline alone were determined. A single dose of indatraline (0.1–1.0 mg/kg) was administered at the beginning of the test session, and 5-min response periods began after 10, 30, 100, and 300 min. For higher doses of indatraline, additional response periods began after 24 and 48 h. In the second series of experiments, the effects of indatraline pretreatment on cocaine discrimination were determined. A single dose of 0.32 mg/kg indatraline was administered 100 min before a test session in which a cumulative cocaine dose-effect curve was determined (0.013–1.3 mg/kg). In the cumulative dosing procedure for cocaine, a single dose of cocaine was administered 5 min after the beginning of each of five consecutive cycles, and each dose increased the cumulative cocaine dose by 1/2 log units. A second cumulative cocaine dose-effect curve was determined the next day, 24 h after administration of 0.32 mg/kg indatraline.

Test sessions were usually conducted on Tuesdays, and additional test sessions were conducted on subsequent days as necessary. Test sessions were conducted only if the three criteria listed above were met during the training day immediately preceding the test day. If responding did not meet criterion levels of discrimination performance, then training was continued until criterion levels of performance were obtained for at least 2 consecutive days.

**Data Analysis.** The percentage of cocaine-appropriate responding (for the entire response period) and response rates were plotted as a function of either the time after indatraline administration (for time-course studies) or the cumulative dose of cocaine (for indatraline pretreatment studies). A percentage of cocaine-appropriate responding for a given cycle was included in the analysis only if the monkey emitted at least 30 responses during the cycle (i.e., enough responses to result in the delivery of one reinforcer). ED50 values were defined as the dose of indatraline or cocaine that produced 50% cocaine-appropriate responding, and were calculated by linear interpolation from individual subject dose-effect curves. For indatraline, ED50 values were calculated from data obtained 100 min after indatraline administration (the approximate time of peak effect), whereas for cocaine, ED50 values were calculated from cumulative dose-effect curves. Individual ED50 values were converted to their log values for calculation of means and 95% CL, then converted back to linear values for presentation.

**Drug Self-Administration**

**Apparatus.** Each monkey was housed individually in a well-ventilated stainless steel chamber (64 × 64 × 79 cm). The home cages of all monkeys were modified to include an operant panel identical with that described above for drug discrimination studies. In addition, two syringe pumps (model B5P-IE; Braintree Scientific, Braintree, MA, or model 980210; Harvard Apparatus, South Natick, MA) were mounted above each cage for delivery of saline or drug solutions through the two lumen of the i.v. catheters. Operation of the operant panels and data collection were accomplished with Apple IIGS computers located in a separate room.

**Surgical Procedures.** Double-lumen Silicone rubber catheters (i.d. 0.7 mm; o.d. 2.0 mm) were implanted in the jugular or femoral vein and exited in the midcascal region. All surgical procedures were performed under aseptic conditions. Monkeys were initially sedated with ketamine (5 mg/kg), and anesthesia was induced with sodium thiopental (10 mg/kg, i.v.). In addition, monkeys were treated with 0.05 mg/kg atropine to reduce salivation. Following insertion of a tracheal tube, anesthesia was maintained with isoflurane (1–1.5% in oxygen). After surgery, aspirin or acetaminophen (80–160 mg/day, p.o.) was administered for 3 days. An antibiotic, procaine penicillin G (300,000 U/day, i.m.), was administered every day for 5 days. The i.v. catheter was protected by a tether system consisting of a custom-fitted nylon vest connected to a flexible stainless steel cable and fluid swivel (Lomir Biomedical, Malone, NY). This flexible tether system permitted monkeys to move freely. Catheter patency was periodically evaluated by i.v. administration of a short-acting barbiturate, methohexital (3 mg/kg, i.v.). The catheter was considered to be patent if i.v. administration of methohexital produced a loss of muscle tone within 10 s.

**Initial Training Procedures.** Procedures for the evaluation of cocaine- and food-maintained responding were identical with those used in our previous studies of the effects of opioids and dopamine antagonists on cocaine self-administration (Negus et al., 1995, 1996, 1997). After initial shaping of key pressing behavior for food reinforcement, animals were maintained on a variable ratio (VR) schedule that was gradually increased to a VR of 16. After monkeys received at least 50 food pellets/day for 3 consecutive days under the VR 16 schedule, behavior was maintained on a second order schedule that consisted of two components, a VR and FR. After completion of a variable number of responses that averaged 16, a red lightoriginally associated with food delivery was illuminated for 1 s before the center response key (VR 16:S). The FR component was gradually increased to a terminal second order schedule response requirement of FR2 (VR 16:S) (one monkey) or FR4 (VR 16:S) (all other monkeys).

Under this terminal schedule, monkeys had to complete two or four VR components, and an average of 32 (range 26–39) or 64 responses (range 53–78) was required for the delivery of each food pellet. Each experimental day began at 9:00 AM, and there were four food sessions during each experimental day beginning at 11:00 AM, 3:00 PM, 7:00 PM, and 6:00 AM the next morning. Each session lasted for 1 h or until a maximum of 25 food pellets had been delivered, whichever occurred first.

Once monkeys received at least 50 food pellets/day for at least 3 consecutive days under the terminal second order schedule, the i.v. double-lumen catheter was implanted as described above. After recovery from surgery (at least 1 week after surgery), key pressing behavior for drug reinforcement (0.032 mg/kg/injection i.v. cocaine injections) was shaped under a series of increasing variable ratios identical with those used during training of food-maintained responding. The final second order schedule response requirement was identical for food- and drug-maintained responding (FR 2 [VR 16:S] or FR 4 [VR 16:S]). There were four cocaine sessions in each experimental day beginning at noon, 4:00 PM, 8:00 PM, and 7:00 AM the next morning (i.e., 1 h after the beginning of the food sessions). Each cocaine session lasted 1 h or until a maximum of 20 injections had been delivered, whichever occurred first.

Cocaine was delivered through one lumen of the double lumen catheter, and saline was automatically delivered through the second lumen. From 9:30 to 10:20 AM every morning, saline was delivered as a daily pretreatment at a rate of 0.1 ml/min for a total of 5.0 ml. For the remainder of the day, saline was delivered at a rate of 0.1 ml every 20 min for a total of 6.9 ml.

The conditions of food and cocaine availability were associated with different colored stimulus lights projected on the center response key of the operant response panel. The two side keys were not transilluminated during the food and drug self-administration studies, and responding on these keys had no scheduled consequences. During food sessions, the center key was transilluminated with a red
stimulus light, whereas during cocaine sessions, the center key was transilluminated with a green stimulus light. Completion of each VR component of the second order schedule was followed by a 10-s time-out period, during which the stimulus light illuminating the center response key was turned off and responding had no scheduled consequences. In addition, the appropriate colored stimulus light (red for food, green for injections) was illuminated for 1 s below the center response key on the completion of each ratio requirement and at the onset of reinforcer delivery. Room lights were extinguished during all food and drug self-administration sessions.

Monkeys were trained until they met the following criteria for stable food and cocaine self-administration under the terminal schedule: 1) 3 consecutive days during which the number of drug injections per day differed by no more than 20% from the mean number of drug injections per day during those 3 days and there was no upward or downward trend; and 2) during the same 3 consecutive days, the mean number of both drug injections per day and food pellets per day was greater than 50.

**Drug Self-Administration Testing.** Once monkeys met the criteria for high stable levels of cocaine and food self-administration, testing began. Three series of tests were conducted to assess the effects of indatraline. The first series of experiments examined the reinforcing effects of indatraline alone in a group of three monkeys. Each dose of indatraline (0.0032–0.32 mg/kg/injection) was substituted for the maintenance dose of cocaine (0.032 mg/kg/injection) for a period of 7 days. The maintenance dose of cocaine was reinstated after each substitution test for a period of at least 4 days and until the number of reinforcers per day maintained by cocaine and food returned to baseline levels. Indatraline doses were tested in an irregular order across monkeys.

The second series of experiments examined the effects of noncontingent treatment with saline or indatraline (0.1–0.56 mg/kg/day) on food- and cocaine-maintained responding in a group of four monkeys during availability of 0.01 mg/kg/injection cocaine. This unit dose of cocaine was used for initial studies because it was the lowest dose to reliably maintain high rates of cocaine self-administration in all monkeys, and because previous studies have shown that behavior maintained by this unit dose of cocaine is especially sensitive to the effects of pretreatment compounds (Negus et al., 1995, 1996, 1997).

In this study, the effects of repeated treatments with saline and each dose of indatraline were examined for 7 consecutive test days. Saline or indatraline was administered by i.v. injection at a rate of 0.1 ml/min through the second lumen of the double lumen catheter from 9:30 to 10:20 AM each morning during the 7-day test period. At the conclusion of each daily indatraline treatment, the catheter was flushed with sterile saline in a volume that exceeded the estimated catheter dead space, and saline was delivered through the second lumen of the double lumen catheter during the remainder of the day. At the conclusion of each 7-day test period, the maintenance dose of cocaine (0.032 mg/kg/injection) and saline pretreatments were reinstated for a period of at least 4 days and until the number of reinforcers per day maintained by cocaine and food returned to baseline levels. Indatraline doses were tested in an irregular order across monkeys.

The final series of experiments examined the effects of saline or indatraline treatment (0.32–0.56 mg/kg/day) on the complete cocaine self-administration dose-effect curve (saline and 0.001–0.1 mg/kg/injection cocaine) in a group of three monkeys. Evaluation of saline and each dose of indatraline during availability of each dose of cocaine was conducted during a block of 7 consecutive test days as described above. Treatment conditions were studied in an irregular order across monkeys. One monkey exhausted all its catheter sites before the effects of the highest dose of indatraline (0.56 mg/kg/day) were examined on self-administration of 0.032 and 0.1 mg/kg/injection cocaine, so data for these combinations were obtained in only two monkeys.

**Data Analysis.** The total numbers of injections or food pellets delivered per day were determined. Data for the effects of indatraline on self-administration of 0.01 mg/kg/injection cocaine (i.e., the first series of experiments) are shown in their entirety and evaluated using a two-factor ANOVA, with indatraline dose and treatment day as the two factors. A significant ANOVA was followed by individual means comparison using the Duncan post hoc test (Winer, 1971). The criterion for significance was set at $p < .05$.

For the remaining series of experiments, data are expressed as the mean ($\pm$S.E.) numbers of injections per day and pellets per day for the 7 days of each test condition. For substitution experiments, cocaine and indatraline dose-effect curves were evaluated using one-factor ANOVAs, and post hoc tests were conducted where appropriate as described above to compare data obtained during saline substitution with data obtained during cocaine and indatraline substitution.

**Observational Studies**

**Apparatus and Procedure.** Informal observation of monkeys during the drug self-administration studies suggested that chronic indatraline treatment may have produced stereotypies and a decrease not only in operant food-maintained responding but also in consumption of daily food rations. To assess the effects of chronic indatraline treatment more systematically, a series of observational studies was conducted in a group of three monkeys. Monkeys remained in their home cages throughout the 4-week experiment and had free access to both food and water. Saline was administered i.m. at 10:00 AM each morning during the first week, and increasing doses of indatraline (0.32, 0.56, and 1.0 mg/kg/day, i.m.) were administered during the second, third, and fourth weeks, respectively. On the last day of each 7-day treatment, monkeys were videotaped for a 10-min period between 1:00 and 1:30 PM, approximately 3 h after the last injection. Monkeys were then weighed under mild ketamine sedation (3–5 mg/kg), and blood samples were collected from the saphenous vein for analysis of hematometry and blood chemistry indices.

Videotapes were later scored by an observer blind to the treatment condition using a scoring system modified from that described by Farfel et al. (1992) for analysis of the unconditioned behavioral effects of chronic cocaine. Specifically, each 10-min observation period was divided into 1-min bins, and the presence or absence of the following four behavioral signs was recorded for each 1-min bin: grooming stereotypies (repetitive and unusually frequent grooming of or picking at skin or hair), buccal stereotypies (repetitive and unusually frequent movements of the tongue or mouth), visual checking (rapid, continuous shifts of visual field resulting from repetitive eye and/or head movements), and speared legs (an unusual posture in which the monkey sits on the perch or floor of the cage with legs spread apart and turned outward). Thus, the maximum score that could be obtained for each behavioral sign during the 10-min observation period was “10,” and the maximum total score that could be obtained across the four signs was “40.”

**Data Analysis.** A one-factor, repeated measures ANOVA was used to evaluate the effects of indatraline on behavioral signs, body weight, and indices of hematometry and blood chemistry. A significant ANOVA was followed by individual means testing using the Duncan post hoc test (Winer, 1971). The level of significance was set a priori at $p > .05$.

**Drugs**

Cocaine HCl was obtained in crystalline form from the National Institute on Drug Abuse (National Institutes of Health, Bethesda, MD), and purity was certified to be greater than 98%. Indatraline HCl (also known as Lu 19–005) was purchased from Research Biochemicals International (Natick, MA). All drugs were dissolved in sterile saline, filter-sterilized using a 0.22-μm Millipore filter and stored in pyrogen-free vials. In the drug discrimination studies and behavioral observation studies, all drugs were administered i.m. in a volume of 0.1 to 2.0 ml. In the drug self-administration experiments,
cocaine and indatraline were delivered i.v in a volume of 0.1 ml/injection.

**Results**

**Cocaine-Like Discriminative Stimulus Effects of Indatraline.** Indatraline administered alone produced a dose- and time-dependent substitution for the discriminative stimulus effects of cocaine in monkeys trained to discriminate 0.4 mg/kg cocaine from saline (Fig. 1, top left). The lowest dose of indatraline (0.1 mg/kg) produced primarily saline-appropriate responding throughout the observation period. A higher dose of indatraline (0.32 mg/kg) produced a maximum of approximately 50% cocaine-appropriate responding after 300 min, but monkeys responded almost exclusively on the saline-appropriate key after 24 h. A dose of 1.0 mg/kg indatraline substituted completely for cocaine in all four monkeys. These cocaine-like discriminative stimulus effects peaked after 30 min and lasted 24 h. Monkeys responded primarily on the saline-appropriate key after 48 h. The mean ED$_{50}$ value (95% CL) for indatraline in producing cocaine-appropriate responding was 0.24 mg/kg (0.084–0.67 mg/kg) at the 100-min time point. Indatraline had little effect on response rates (Fig. 1, bottom left) at the doses and times tested.

Figure 1 (right) shows the effects of indatraline pretreatment on the cocaine dose-effect curve. Cocaine alone (0.013–1.3 mg/kg) produced a dose-dependent increase in cocaine-appropriate responding. The ED$_{50}$ (95% CL) for cocaine was 0.15 mg/kg (0.087–0.26 mg/kg), and complete substitution was observed at the training dose of 0.4 mg/kg cocaine. A dose of 0.13 mg/kg cocaine produced a slight increase in response rates, and higher cocaine doses decreased response rates. Pretreatment with indatraline (0.32 mg/kg, 100 min) increased levels of cocaine-appropriate responding produced by low doses of cocaine (0.013–0.13 mg/kg) and produced a small leftward shift in the cocaine dose-effect curve for response rates. These effects diminished after 24 h, although a small leftward shift in the cocaine discrimination dose-effect curve was still evident. ED$_{50}$ values for cocaine after indatraline pretreatment could not be determined, because three monkeys responded exclusively on the cocaine-appropriate key 100 min after indatraline pretreatment, and one monkey responded exclusively on the cocaine-appropriate key after 24 h.

**Self-Administration of Cocaine and Indatraline.** Figure 2 shows the effects of substituting saline, cocaine alone (0.001–0.1 mg/kg/injection), or indatraline alone (0.0032–0.032 mg/kg/injection) for the maintenance dose of 0.032 mg/kg/injection cocaine in monkeys trained to respond for i.v. cocaine injections and food. Saline substitution maintained an average of 23.9 (±9.7) injections/day during the 7-day test period, and monkeys earned an average of 95.4 (±1.4) food pellets/day. Cocaine substitution produced an inverted U-shaped dose-effect curve for drug self-administration. Doses of 0.0032 to 0.1 mg/kg/injection cocaine maintained more injections per day than saline (p < .05), and peak rates of self-administration were maintained by the maintenance dose of 0.032 mg/kg/injection cocaine (79.5 ± 0.3 injections/day). Cocaine self-administration across the dose

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**Fig. 1.** Effects of indatraline administered alone (left) or as a pretreatment to cocaine (right) in monkeys trained to discriminate 0.4 mg/kg cocaine from saline. Abscissae (left), time after indatraline administration in minutes (10–300) or hours (24–48). Abscissae (right), cumulative dose cocaine in mg/kg. Ordinates (top), percentage of cocaine-appropriate responding. Ordinates (bottom), response rate in responses per second. All points show mean data (±S.E.) from four monkeys except for “% Cocaine Responding” for the following points in the upper right: 1.3 mg/kg cocaine alone, n = 1; 0.32 mg/kg indatraline (100 min) + 0.4 mg/kg cocaine, n = 3; 0.32 mg/kg indatraline (100 min) + 1.3 mg/kg cocaine, n = 2; and 0.32 mg/kg indatraline (24 h) + 1.3 mg/kg cocaine, n = 3. The remaining monkeys did not respond under these conditions, and a “% Cocaine Responding” could not be determined.
range tested did not significantly alter the number of food pellets earned per day.

Indatraline substitution also produced an inverted U-shaped self-administration dose-effect curve, but across the dose range tested, the numbers of injections per day during indatraline substitution were not significantly different from during saline substitution. The highest rates of self-administration were maintained by a dose of 0.01 mg/kg/injection indatraline (43.2 ± 10.4 injections/day), and one monkey earned an average of 63.9 injections/day during this treatment condition. In contrast to the effects of cocaine substitution, the number of pellets earned per day decreased dose dependently during indatraline substitution. Monkeys earned significantly fewer pellets per day during 0.032 mg/kg/injection indatraline substitution than during saline substitution. In addition, stereotypes were observed in two of the three monkeys during indatraline availability, and substitution of the highest dose of 0.032 mg/kg/injection indatraline had to be terminated after 3 days in one monkey because of severe oral stereotypes. These findings suggest that, although monkeys self-administered relatively few indatraline injections per day, the self-administered doses of indatraline were behaviorally active.

**Effects of Indatraline Treatment on Cocaine Self-Administration.** Figure 3 shows the numbers of injections per day and pellets per day when a unit dose of 0.01 mg/kg/injection cocaine was available for self-administration and monkeys were treated with saline or indatraline (0.1–0.56 mg/kg/day) daily for 7 days. During saline treatment, monkeys earned nearly maximal numbers of injections and pellets per day. Indatraline produced a dose- and time-dependent decrease in both cocaine- and food-maintained responding. Treatment with 0.32 mg/kg/day indatraline significantly decreased the number of pellets per day ($p < .05$), and 0.56 mg/kg/day indatraline significantly decreased the numbers of both pellets and cocaine injections per day ($p < .01$). The effects of indatraline on cocaine self-administration increased significantly during the 7 days of treatment ($p < .01$), and cocaine self-administration was nearly eliminated after 7 days of treatment with 0.56 mg/kg/day indatraline. The effects of indatraline on food-maintained responding did not change significantly over time. However, there was a trend toward recovery of food-maintained responding during the least few days of treatment with 0.56 mg/kg/day indatraline, when cocaine self-administration was maximally decreased.

![Fig. 2. Self-administration of saline, cocaine (0.001–0.1 mg/kg/injection), or indatraline (0.0032–0.032 mg/kg/injection) and concurrent responding for food. Abscissae, unit dose cocaine or indatraline in mg/kg/injection. Points above “Sal” show the effects of saline substitution. Ordinate (left), average number of injections per day (maximum = 80). Ordinate (right), average number food pellets per day (maximum = 100). Each point shows mean data (±S.E.M.) for three monkeys during 7 consecutive days of treatment, except for the highest dose of indatraline (0.032 mg/kg/injection). Availability of 0.032 mg/kg/injection indatraline had to be terminated after 3 days in one monkey due to the emergence of oral stereotypes. Asterisks indicate effects significantly different from saline substitution (∗$p < .05$; ∗∗$p < .01$).](image1)

![Fig. 3. Effects of treatment with saline or indatraline (0.1–0.56 mg/kg/day) on responding maintained by cocaine (0.01 mg/kg/injection) and food. Abscissae, consecutive days of treatment. Ordinate (left), average number of 0.01 mg/kg/injection cocaine injections per day (maximum = 80). Ordinate (right), average number food pellets per day (maximum = 100). Each point shows mean data (±S.E.M.) for four monkeys.](image2)
Figure 4 shows the effects of chronic indatraline treatment (0.32 and 0.56 mg/kg/day) on the entire cocaine self-administration dose-effect curve. As in the experiments described above, each dose of indatraline in combination with a unit dose of cocaine was studied for 7 consecutive days. The lower dose of 0.32 mg/kg/day indatraline had little effect on cocaine self-administration (0.0032–0.032 mg/kg/day). The higher dose of 0.56 mg/kg/day indatraline decreased cocaine self-administration across a broad range of cocaine doses (0.0032–0.1 mg/kg/injection) and produced a downward shift in the cocaine self-administration dose-effect curve. Indatraline treatment also tended to decrease the numbers of pellets per day, although effects on food-maintained responding were variable. Finally, chronic indatraline treatment appeared to produce stereotypies and a decrease in food consumption in some monkeys, and as a result, additional studies were conducted as described below.

Other Effects of Chronic Indatraline Treatment. Table 1 shows the effects of chronic treatment with saline and increasing doses of indatraline (0.32–1.0 mg/kg/day) on overt behavior, body weight, and indices of hematology and blood chemistry. During saline treatment, visual checking, stereotypic grooming and buccal movements, and splayed legs were rarely observed. However, indatraline significantly increased the overall stereotypy score. Visual checking was observed primarily during treatment with 0.32 mg/kg indatraline, and this sign decreased in frequency during treatment with higher indatraline doses. After treatment with 1.0 mg/kg/day indatraline, all three monkeys showed continuous buccal and/or grooming stereotypies, and splayed legs were frequently observed in two of the three monkeys. Indatraline treatment also produced a dose-dependent decrease in body weight, blood hemoglobin, hematocrit, red blood cell count, and white blood cell count. The decrease in blood hemoglobin was statistically significant at the highest dose of indatraline (1.0 mg/kg/day), and changes in the other indices approached significance (p < .1; Table 1). Blood chemistry measures were not significantly affected by indatraline treatment.

Discussion

The present study examined the hypothesis that indatraline may produce cocaine-like behavioral effects and be useful as a substitution medication for the treatment of cocaine abuse. Our major findings were that indatraline produced cocaine-like discriminative stimulus effects with a slow onset and a long duration of action, and repeated treatment with indatraline produced a sustained decrease in cocaine self-administration across a broad range of cocaine doses. However, at doses that decreased cocaine self-administration, indatraline usually produced undesirable effects that could limit its clinical utility. Each of these major findings is discussed below.

Cocaine-Like Discriminative Stimulus Effects of Indatraline. Drug discrimination studies confirmed that indatraline produced a dose-dependent and complete substitution for cocaine in rhesus monkeys. The potencies of indatraline and cocaine in producing cocaine-like discriminative stimulus effects were similar, and neither compound substantially altered response rates at doses that produced complete substitution. However, indatraline had a slow onset and long duration of action relative to cocaine. For example, the effects of 1.0 mg/kg indatraline peaked after 30 min and lasted at least 24 h, whereas the effects of the training dose of cocaine (0.40 mg/kg) peaked after only 3 min and were nearly gone after 60 min (Lamas et al., 1995). These results extend previous reports that indatraline produced psychomotor stimulant effects in rats and squirrel monkeys with a potency similar to cocaine but with a slower onset and longer duration of action (Arnt et al., 1985; Rosenzweig-Lipson et al., 1992; Kleven and Koek, 1998). These behavioral findings are also consistent with in vitro studies demonstrating that indatraline binds to [3H]cocaine-labeled binding sites in monkey striatum (Madras et al. 1989) and, like cocaine, functions as a relatively nonselective monoamine reuptake inhibitor (Koe, 1976; Hyttel, 1982; Hyttel and Larsen, 1985). In addition to producing cocaine-like discriminative stimulus effects when administered alone, indatraline pretreatment also enhanced the discriminative stimulus effects of low doses of cocaine. These findings agree with a previous report that indatraline pretreatment enhanced the discriminative stimulus effects of a low cocaine dose in rats (Kleven and Koek, 1998).

The relative contribution of specific monoaminergic systems to the cocaine-like effects of indatraline is unknown.
Like indatraline, selective dopamine reuptake inhibitors were found to substitute for cocaine and enhance the discriminative stimulus effects of low cocaine doses (Cunningham and Callahan, 1991; Mansbach and Balster, 1993; Spealman, 1993; Kleven and Koek, 1998). Selective inhibitors of norepinephrine and serotonin are generally less effective in substituting for cocaine (Kleven et al., 1990; Cunningham and Callahan, 1991; Spealman, 1993, 1995; Kleven and Koek, 1997), although norepinephrine reuptake inhibitors may produce high levels of cocaine-appropriate responding when the training dose of cocaine is low (Terry et al., 1994; Spealman, 1995), and both norepinephrine and serotonin reuptake inhibitors enhanced the discriminative stimulus effects of cocaine in rats (Cunningham and Callahan, 1991; Kleven and Koek, 1997). In contrast, the serotonin reuptake inhibitor citalopram attenuated cocaine discrimination in monkeys (Spealman, 1993). These findings suggest that the cocaine-like effects of indatraline were mediated primarily by dopaminergic systems, although effects on norepinephrine, and possibly serotonin, may also have contributed.

**Reinforcing Effects of Indatraline.** Despite its ability to produce cocaine-like discriminative stimulus effects, indatraline maintained low rates of self-administration similar to those maintained by saline. It was shown previously that decreases in the rate at which a cocaine dose was infused produced corresponding decreases in the rate of cocaine self-administration (Balster and Schuster, 1973). These findings have been interpreted to suggest that rate of onset may be an important determinant of a drug’s reinforcing effects. Thus, indatraline’s slow onset may limit its ability to function as positive reinforcer.

A more likely possibility, however, is that rates of indatraline self-administration were limited by its long duration of action (necessitating fewer injections per unit of time than cocaine to produce a given behavioral effect) and by nonspecific effects on operant responding. This interpretation is consistent with our finding that indatraline substitution produced a dose-dependent decrease in food-maintained responding. In addition, two of three monkeys developed behavioral stereotypies during availability of the highest indatraline dose. Finally, monkeys self-administered an average of 0.43 and 0.78 mg/kg/day indatraline during availability of 0.01 and 0.032 mg/kg/injection indatraline, respectively, and as described above, these doses produced long-lasting cocaine-like discriminative stimulus effects. These findings suggest that monkeys self-administered behaviorally active doses of indatraline despite the low rates of self-administration. Similar low rates of self-administration have also been observed for the long-acting dopamine-selective reuptake inhibitor 2-[3-(4-tolyl)-tropane (Nader et al., 1997; Birmingham et al., 1998).

Although we cannot rule out the possibility that indatraline may produce more robust reinforcing effects under different experimental conditions, the results of the present study indicate that indatraline maintains lower rates of self-administration than cocaine when the rate of drug intake is controlled primarily by the subject. These findings suggest that indatraline and other long-acting monoamine reuptake inhibitors may be less likely than cocaine to maintain binge patterns of drug use. Moreover, the ability of indatraline to produce some cocaine-like effects may facilitate compliance in a treatment setting.

**Effects of Indatraline on Cocaine Self-Administration.** We have argued previously that treatments that produce a sustained decrease in cocaine self-administration across a broad range of cocaine doses are more likely to be effective medications for the treatment of cocaine dependence than treatments that do not affect or that increase cocaine self-administration (Mello and Negus, 1996). Consistent with this profile of effects, indatraline produced a dose-dependent decrease in the self-administration of a dose of cocaine (0.01 mg/kg/injection) near the peak of the cocaine dose-effect curve. These decreases in cocaine self-administration were sustained throughout the 7-day treatment period, and treatment with 0.56 mg/kg/day indatraline eliminated cocaine self-administration by the end of the treatment period. Indatraline also produced sustained decreases in the self-administration of other unit doses of cocaine (0.0032–0.1 mg/kg/injection), which resulted in an overall downward shift in the cocaine self-administration dose-effect curve.

As in the drug discrimination studies, the relative contribution of specific monoaminergic systems to the effects of indatraline on cocaine self-administration remains to be determined. However, the effects of indatraline on cocaine self-administration are similar to the effects of more selective dopamine reuptake inhibitors such as mazindol (Mansbach...
and Balster, 1993), GBR12909 (Glowa et al., 1995a,b), 2β-carboxymethoxy-3β-(4-fluorophenyl)tropane (Glowa et al., 1995a), 2β-propanoyl-3β-(4-tolyloxy)tropane (Nader et al., 1997), and RTI-113 (Dworkin et al., 1998). Selective norepinephrine and serotonin reuptake inhibitors also decreased cocaine self-administration, although these compounds may not be as effective as dopamine reuptake inhibitors (Mello et al., 1990; Peltier and Schenk, 1993). Most of these compounds have not been evaluated during chronic treatment or in combination with doses from both the ascending and descending limbs of the cocaine dose-effect curve. However, like indatraline in the present study, GBR12909 was also found to produce a downward shift in the cocaine self-administration dose-effect curve and a sustained decrease in cocaine self-administration during repeated days of treatment (Glowa et al., 1995a,b). The dopamine releaser phentermine also produced similar decreases in cocaine self-administration (Wojnicki et al., 1999). In contrast to the effects of monoamine reuptake inhibitors, other candidate treatment medications produce different profiles of effects on cocaine self-administration. For example, dopamine antagonists produced rightward shifts in cocaine dose-effect curves and increases in self-administration of high cocaine doses, whereas D2/D3-selective dopamine agonists produced leftward shifts in cocaine dose-effect curves and increases in self-administration of low cocaine doses (Bergman et al., 1990; Caine and Koob, 1995). Furthermore, dopamine antagonists produced only transient changes in cocaine self-administration that were not sustained during chronic treatment (Kleven and Woolverton, 1990; Negus et al., 1996). Thus, relative to some other candidate medications, indatraline and some other monoamine reuptake inhibitors may produce a more optimal profile of effects on cocaine self-administration.

Other Effects of Indatraline. To provide one measure of behavioral selectivity in the present study, the effects of indatraline treatment on concurrent food-maintained responding were examined. In some cases, the effects of indatraline on cocaine self-administration were greater than the effects on food-maintained responding. For example, after 7 days of treatment with 0.56 mg/kg/day indatraline, responding for the 0.01 mg/kg/injection cocaine was eliminated, whereas responding for food was only partially suppressed and appeared to be recovering. Overall, however, doses of indatraline that decreased cocaine self-administration also usually decreased responding maintained by food. In addition, separate studies found that doses of indatraline that decreased cocaine self-administration also produced behavioral stereotypes, a trend toward weight loss, and changes in hematology suggestive of mild anemia. These effects are similar to those produced by chronic high-dose cocaine administration in animals or cocaine abuse in humans (Farrel et al., 1992; Burkett et al., 1994), and provide additional evidence to suggest that indatraline and cocaine produce a similar profile of behavioral effects. However, these findings also suggest that robust decreases in cocaine self-administration occur only at relatively high indatraline doses that may also produce other undesirable effects.

In contrast to these findings with indatraline, more selective dopamine reuptake inhibitors were reported to decrease cocaine self-administration without altering rates of food-maintained responding or producing other serious side effects (Glowa et al., 1995a,b; Dworkin et al., 1998). Those studies used different procedures to establish and measure cocaine- and food-maintained responding and to deliver pre-treatment drugs, so direct comparison with the results of this study is difficult. Moreover, selective decreases in cocaine self-administration were not always observed even with selective dopamine reuptake inhibitors (Mansbach and Balster, 1993; Glowa et al., 1995b). Consequently, the extent to which selective dopamine reuptake inhibitors may decrease cocaine self-administration more safely than nonselective monoamine reuptake inhibitors requires further study. In addition, the implications of the undesirable effects for the clinical treatment of cocaine abuse remain to be determined. For example, it may be possible to use lower and relatively safe doses of monoamine reuptake inhibitors for the treatment of cocaine abuse when pharmacotherapy is combined with other forms of treatment, such as psychotherapy or behavioral therapy (Mendelson and Mello, 1996).

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References


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