Discriminative Stimulus Effects of the Nonpeptidic δ-Opioid Agonist SNC80 in Rhesus Monkeys

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ABSTRACT

Five rhesus monkeys were trained to discriminate the nonpeptidic, δ-opioid agonist SNC80 (0.32 mg/kg i.m.) from saline by using a food-reinforced drug-discrimination procedure. Cumulative doses of SNC80 produced a dose-dependent increase in SNC80-appropriate responding and a dose-dependent decrease in response rate. In time-course studies, peak effects of the training dose of SNC80 were observed after 15 min, and these effects diminished over 240 min. In substitution studies, other piperazinyl benzamide δ agonists (SNC86, SNC162, and SNC243A) substituted for SNC80 with relative potencies similar to those of SNC80. However, SNC67, the (-)-enantiomer of SNC80, did not occasion SNC80-appropriate responding up to a dose (32.0 mg/kg) that produced convulsions in one monkey. The μ agonists morphine and fentanyl and the κ agonists U-50,488 and enadoline failed to substitute for SNC80 up to doses that eliminated responding. Two nonopioids (the N-methyl-D-aspartate antagonist ketamine and the monoamine reuptake inhibitor cocaine) also produced primarily saline-appropriate responding. Both the discriminative stimulus and rate-decreasing effects of SNC80 were antagonized by the δ-selective antagonist naltrindole (0.01–1.0 mg/kg) but not by doses of the opioid antagonist quadazocine (0.1–1.0 mg/kg) that block the effects of μ and κ agonists. These data suggest that the discriminative stimulus effects of SNC80 are mediated by δ-opioid receptors and that the discriminative stimulus effects of δ opioids in primates can be differentiated from the effects of other opioid and nonopioid compounds.

Opioids act at three main types of opioid receptors, the μ, κ, and δ receptors (Martin et al., 1976; Lord et al., 1977; Wood et al., 1981). Drug-discrimination procedures have been used extensively to characterize the stimulus properties of compounds that interact with these types of opioid receptors (Schaefer and Holtzman, 1977; Herling and Woods, 1981a; France and Woods, 1989; Negus et al., 1994). Often in drug discrimination studies, drugs that share pharmacologic mechanisms of action with the training drug substitute for the training drug (i.e., produce training drug-associated responding), whereas drugs that do not share pharmacologic effects with the training drug do not substitute (i.e., produce vehicle-associated responding). For example, in monkeys trained to discriminate the μ agonist etorphine from saline, other μ agonists produced etorphine-associated responding, whereas κ agonists did not (Herling and Woods, 1981b).

Conversely, in monkeys trained to discriminate the κ agonist ethylketocyclazocine (EKC) from saline, other κ agonists substituted for EKC, whereas μ agonists did not (Hein et al., 1981). The discriminative stimulus effects of μ and κ opioids have been well characterized, in part because of the availability of selective agonists and antagonists that readily cross the blood-brain barrier after systemic administration.

In contrast, the discriminative stimulus effects of δ opioids have not been fully characterized. Until recently, the only δ agonists available have been peptidic compounds such as [δ-Pen²-δ-Pen⁵]enkephalin (DPDPE). However, DPDPE and other peptides do not readily cross the blood-brain barrier and must be administered i.c.v. This poses some methodological problems for training and maintaining discrimination performance. However, one study did train pigeons to discriminate between i.c.v. injections of DPDPE and saline (Jewett et al., 1996). In substitution studies, other peptidic δ agonists substituted for DPDPE, whereas the κ agonist U-50,488 and the μ agonists DAMGO and morphine did not (Jewett et al., 1996). This study demonstrated a clear, δ-selective discrimination with DPDPE. Recently, (-)-BW373U86 was described as the first nonpeptidic, systemi-

ABBREVIATIONS: EKC, ethylketocyclazocine; % DR, percent drug responding; DPDPE, [δ-Pen²-δ-Pen⁵]enkephalin.

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cally active δ-opioid agonist (Chang et al., 1993). In pigeons, the discriminative stimulus effects of (+)-BW373U86 (0.56 mg/kg) were antagonized by the δ-selective antagonist naltrindole (Comer et al., 1993b; Picker and Cook, 1998). These antagonism studies suggested that the discriminative stimulus effects of (+)-BW373U86 were mediated by δ receptors. However, in substitution studies, μ agonists (e.g., morphine) substituted for the (+)-BW373U86 discriminative stimulus in more than half of the pigeons studied (Comer et al., 1993b; Picker and Cook, 1998). Moreover, in pigeons trained to discriminate μ agonists (e.g., fentanyl), (+)-BW373U86 produced high levels of substitution in the majority of pigeons (Negus et al., 1996; Picker, 1997). These findings suggest that the discriminative stimulus effects of (+)-BW373U86 are not selective for δ agonists in pigeons.

Although (+)-BW373U86 appears to share discriminative stimulus effects with μ agonists in pigeons, it may produce more selective δ agonist effects in primates. For example, (+)-BW373U86 did not produce μ- or κ-like discriminative stimulus effects in rhesus monkeys trained to discriminate the μ agonist alfentanil or the κ agonist EKC from saline (Negus et al., 1994). In addition, piperazinyl benzamide derivatives of (+)-BW373U86 recently have been developed that are far more selective for δ receptors than the parent compound. For example, SNC80 is the methyl ether derivative of (+)-BW373U86, and SNC80 was reported to have more than 800-fold selectivity for δ versus μ receptors, whereas (+)-BW373U86 was between 7- and 50-fold selective for δ receptors (Chang et al., 1993; Calderon et al., 1994, 1997; Knapp et al., 1996). Moreover, we reported recently that SNC80 functions as a systemically active and highly selective δ agonist in rhesus monkeys (Negus et al., 1998). Taken together, these findings suggest that a drug discrimination based on SNC80 in rhesus monkeys could provide a sensitive and selective assay for the evaluation of δ opioids.

Accordingly, the purpose of the present study was to determine whether SNC80 would serve as a selective δ opioid discriminative stimulus in monkeys. After adequate stimulus control was established, the potency and time course of SNC80 were determined. In substitution studies, the other piperazinyl benzamide δ agonists SNC86 [the (+)-enantiomer of (+)-BW373U86], SNC162, and SNC243A were evaluated to determine whether these compounds shared discriminative stimulus effects with SNC80. The stereoselectivity of the discriminative stimulus effects of SNC80 also was assessed by using SNC67, which is the (-)-enantiomer of SNC80. Substitution studies with compounds selective for μ (i.e., fentanyl and morphine) and κ (i.e., enadoline and U-50,488) opioid receptors, as well as nonopioid receptors (i.e., the N-methyl-D-aspartate receptor antagonist ketamine and the monoamine reuptake blocker cocaine), were conducted to determine the selectivity of the SNC80 discriminative stimulus. In antagonism studies, the δ-selective antagonist naltrindole and μ- and κ-selective doses of the opioid antagonist quazocine were administered as pretreatments to SNC80 to assess the role of δ, μ, and κ receptors in mediating the discriminative stimulus effects of SNC80.

Materials and Methods

Subjects. Two male and three female rhesus monkeys (Macaca mulatta) were studied. All monkeys had previous exposure to cocaine self-administration procedures; however, none of the monkeys had previous drug-discrimination experience. Monkeys had free access to water and were maintained on a diet of multiple vitamins, fresh fruit, and five to eight Lab Diet Jumbo Monkey biscuits (PMI Feeds, Inc., St. Louis, MO). In addition, monkeys could earn up to 70 g food pellets (Precision Primate Pellets Formula L/I Banana Flavor; P.J. Noyes Co., Lancaster, NH) during daily operant sessions (see below). 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 laboratory facility was licensed by the United States Department of Agriculture, and research protocols were approved by the McLean Hospital Institutional Animal Care and Use Committee. Consulting veterinarians periodically monitored the health of the monkeys. Monkeys had visual, auditory, and olfactory contact with other monkeys throughout the study. Operant procedures for food-maintained responding provided an opportunity for environmental enrichment (Line et al., 1989).

Apparatus. Monkeys were housed individually in stainless steel chambers (56 × 71 × 69 cm). An operant panel (28 × 28 cm) was mounted on the front of each home cage. Each panel contained a horizontal row of three response keys (6.4 × 6.4 cm) that were arranged 2.5 cm apart and 3.2 cm from the top of the operant panel. Keys could be transilluminated with red or green stimulus lights (Superbrights LEDs; Newark Electronics, Woburn, MA). An externally mounted pellet dispenser (model G5210; Gerbrands) delivered 1-g banana-flavored pellets to a food receptacle mounted on the cage beneath the operant panel. Control of experiments and data recording were accomplished with a microprocessor, interface, and software (MED Associates Inc., Georgia, VT) located in a separate room.

Discrimination Training. Monkeys were trained under a single-cycle procedure during experimental sessions conducted 5 days each week. Each training cycle consisted of a 15-min pretreatment period followed by a 10-min response period. During the pretreatment period, stimulus lights were not illuminated, and responding had no scheduled consequences. During the response period, stimulus lights were illuminated, and monkeys could earn a maximum of 20 food pellets under a fixed ratio 20 schedule of reinforcement. For three of the monkeys, the right key was illuminated red and the left key was illuminated green. The colors of the response keys were reversed for the other two monkeys. The center key was inactive, and responding on this key had no programmed consequences. If 20 food pellets were delivered before the end of the 10-min response period, the stimulus lights were extinguished, and further responses had no scheduled consequence for the remainder of the response period.

An injection of either saline or 0.32 mg/kg SNC80 was administered i.m. during the first minute of the pretreatment period. After the administration of saline, only responses on the green key (i.e., saline-appropriate key) were reinforced, and after the administration of SNC80, only responses on the red key (i.e., drug-appropriate key) were reinforced. Responding on the injection-inappropriate key reset the fixed ratio requirements on the injection-appropriate key.

Three dependent variables were determined during the response period of each cycle: 1) the number of responses on each key before the first food reinforcer; 2) the percent responses on the injection-appropriate key for the entire cycle ([injection-appropriate responses ÷ total responses] × 100); and 3) response rate for the cycle (total responses emitted ÷ total time stimulus lights were illuminated).

Monkeys were considered to have acquired the discrimination when the following three criteria were met for seven of eight training sessions: 1) fewer than 20 responses on the injection-inappropriate key before the first reinforcer, 2) at least 90% responding on the injection-appropriate key over the entire session, and 3) response...
rates of at least 0.5 responses/s after the administration of either saline or the training dose of SNC80.

Initially, training consisted of a double-alternation schedule of drug or saline administration on consecutive days (i.e., sal-sal-drug-drug). Analysis of performance during early training indicated that the criteria for adequate stimulus control more often were obtained on the first drug presentation than on the second drug presentation in the schedule. Because studies in our laboratory suggest that tolerance may develop to some of the behavioral effects of δ agonists (unpublished observations), training was modified so that drug was never administered on consecutive days (e.g., either a saline session or no session occurred between drug-training days).

**Discrimination Testing.** Once monkeys met the criteria for accurate discrimination, testing began using a multiple-cycle procedure. Each cycle during a test session was identical with training cycles except: 1) the 15-min pretreatment period was followed by a 5-min response period (i.e., 20-min cycles), 2) responding on either key produced food, and 3) a maximum of 10 food pellets was available during each cycle. Test sessions consisted of up to seven consecutive test cycles and were conducted only if the criteria for adequate stimulus control were met during the two training days immediately preceding the test day. If responding did not meet criterion levels, training was continued until criterion levels of performance were obtained for at least two consecutive sessions (one drug and one saline training session). A saline training day always preceded test days, and a minimum of 5 days separated tests with pipеразинyl benzamides.

Three series of experiments were conducted to examine the discriminative stimulus effects of SNC80. In experiment 1, the potency and time course of the discriminative stimulus and rate-decreasing effects of SNC80 were examined. The potency of SNC80 was assessed by using a cumulative dosing procedure. Under this procedure, saline was administered at the beginning of the first cycle, and cumulative doses of SNC80 were administered i.m. during the first minute of each subsequent cycle. Doses increased cumulatively in 0.25 or 0.5 log unit increments. SNC80 dose-effect curves were determined at the start and at the conclusion of these studies. In addition, a separate test session consisted of seven cycles during which saline was administered during the first minute of each cycle. To examine the time course of the training dose of SNC80, a single dose of 0.32 mg/kg SNC80 was administered, and 5-min response periods were scheduled to begin after 4, 15, 60, and 240 min.

In experiment 2, the ability of opioid and nonopioid drugs to substitute for the SNC80 discriminative stimulus was examined. During these studies, saline was administered during the first cycle, and cumulative doses of the test drug were administered i.m. during the first minute of subsequent cycles. The four classes of drugs studied were as follows: 1) pipеразинyl benzamides structurally related to SNC80 (SNC67, SNC86, SNC162, and SNC243A), 2) μ agonists (morphine and fentanyl), 3) κ agonists (U-50,488 and enadoline), and 4) nonopioids (the N-methyl-D-aspartate receptor antagonist ketamine and the monoamine reuptake blocker cocaine). All test compounds were evaluated at least once in each of four monkeys. During the evaluation of the U50,488 dose-effect curve, which used 1/2 log unit increments between doses, U50,488 substituted for SNC80 in one of four monkeys. To assess the replicability of this finding, the U50,488 dose-effect curve was redetermined in these monkeys by using 1/4 log unit increments between doses. Experimental sessions were terminated when: 1) at least 90% of the responses occurred on the drug-appropriate key, 2) rates of responding decreased to less than 10% of the average rate of the five previous saline training sessions, or 3) signs of overt toxicity were observed.

In experiment 3, the δ-opioid selectivity of the behavioral effects of SNC80 was evaluated by pretreating monkeys with the opioid antagonists naltrindole or quazadocine. During antagonism studies, a single dose of naltrindole (0.01, 0.1, or 1.0 mg/kg) or quazadocine (0.1 or 1.0 mg/kg) was administered at the beginning of the first cycle, and cumulative doses of SNC80 were administered during the first minute of each subsequent cycle (i.e., 20-min antagonist pretreatment). We have reported previously that naltrindole, at doses of up to 1.0 mg/kg, acts as a selective δ antagonist in rhesus monkeys (Negus et al., 1998). Quazadocine selectively antagonizes μ agonists at a dose of 0.1 mg/kg and blocks both μ and κ agonists at a dose of 1.0 mg/kg in rhesus monkeys (Bertalmio and Woods, 1987; Negus et al., 1993).

**Data Analyses.** Results of drug-discrimination studies are presented as the average percentage of drug-appropriate responses (i.e., % DR) on the SNC80-appropriate key ± 1 S.E.M. and are plotted as a function of either dose or time. A drug was considered to substitute for SNC80 when at least 90% of the responses were on the drug-appropriate key. Rates of responding are presented as a percentage of the control response rate ± 1 S.E.M. The control response rate was defined as the average response rate of the five saline training sessions immediately preceding the test. The % DR was not calculated for a monkey for a given cycle if response rates were less than 10% of the control rate. In addition, a mean % DR was calculated and plotted in group graphs only if at least two monkeys contributed to the data point.

ED<sub>50</sub> values were defined as the dose of a drug that produced either 50% SNC80-appropriate responding or 50% decrease in control response rates. Individual ED<sub>50</sub> values were calculated by linear regression when at least three data points were available on the linear portion of the dose-effect curve or by interpolation when two data points (one above and one below 50%) were available. Individual ED<sub>50</sub> values were converted to their log values for calculation of means and 95% CL and then converted back to linear values for presentation. One monkey was removed midway through these studies because of health problems that were unrelated to the current experiment. The number of monkeys in each testing condition is indicated in the figure legends.

**Drugs.** SNC67, SNC80, SNC162, and SNC243A were synthesized as free bases by K.C.R. and colleagues (National Institutes of Health, Bethesda, MD). SNC86 HCl and naltrindole HCl also were synthesized by K.C.R. and colleagues. (-)-Cocaine HCl, fentanyl HCl, and morphine sulfate were supplied by the National Institute on Drug Abuse (Bethesda, MD). Quadazocine methanesulfonate was generously supplied by Sanofi Pharmaceuticals (Malvern, PA), and enadoline HCl was generously supplied by Warner-Lambert (Parke-Davis Research Division, Ann Arbor, MI). (+)-trans-U50,488 methanesulphonate was purchased from Research Biochemicals International (Natick, MA), and ketamine-HCl was purchased from Fort Dodge Laboratories (Fort Dodge, IA). SNC67, SNC80, SNC162, and SNC243A were dissolved in 3% lactic acid and water. The commercially available Ketaset solution was used for ketamine. All other compounds were dissolved in sterile water. Drugs were administered i.m., and the sites of injection were rotated so that the same site was never used on 2 consecutive days. Signs of tissue damage were not observed after injections with any of the test compounds. Doses were based on the free base or salt forms described above.

**Results**

**Control Performance and Effects of SNC80.** Monkeys satisfied the criteria for adequate stimulus control after an average of 80 training sessions (range, 59–119). The mean control rate of responding (determined from saline training sessions preceding test sessions throughout the study) was 1.53 ± 0.20 responses per second. Figure 1 shows the potency and time course of SNC80. Cumulative administration of SNC80 produced a dose-dependent increase in SNC80-appropriate responding and a dose-dependent decrease in response rates (Fig. 1, left). Complete substitution was observed at doses of SNC80 higher than 0.32 mg/kg, and a dose of 3.2 mg/kg eliminated responding. The potency of SNC80 was determined by using cumulative dosing procedures at the
beginning and again at the end of these studies. Mean ED_{50} values (±95% CL) for the discriminative stimulus effects of SNC80 were similar for the first [0.13 (0.06–0.18)] and second [0.13 (0.09–0.17)] determination. Mean ED_{50} values (±95% CL) for the rate-decreasing effects of SNC80 were also similar for the first [0.69 (0.12–1.50)] and the second [1.36 (0.47–2.03)] determination. Because there was substantial overlap in 95% CL between the first and second determination, the two dose-effect curves were combined in all Figs. 1–4, and ED_{50} values were combined in Tables 1 and 2. In contrast to the effects of cumulative SNC80 administration, injections of saline during seven cycles produced primarily saline-appropriate responding and response rates between 83 and 108% of control throughout the test (data not shown).

The training dose of 0.32 mg/kg SNC80 had a rapid onset of action and a moderate duration of action (Fig. 1, right). Three of four monkeys responded more than 90% on the drug-appropriate key after 4 min, and all monkeys responded more than 90% on the drug-appropriate key after 15 min. After 4 h, monkeys responded primarily on the saline-appropriate key. The training dose of SNC80 did not markedly affect rates of responding.

**Effects of Other Piperazinyl Benzamide δ Agonists.** SNC86, SNC162, and SNC243A dose-dependently substituted for the SNC80 discriminative stimulus in nearly all monkeys and decreased rates of responding (Fig. 2). SNC86 substituted in three monkeys at a dose of 0.32 mg/kg and in the fourth monkey at a dose of 1.0 mg/kg. However, average drug-appropriate responding did not exceed 88%, because in two monkeys, a dose of 1.0 mg/kg SNC86 produced 80 and 74% drug-appropriate responding. SNC243A substituted in three of the four monkeys; in the fourth monkey, a dose of 0.32 mg/kg SNC243A produced 88% SNC80-appropriate responding and a higher dose of 1.0 mg/kg SNC243A eliminated responding. The potencies of SNC86, SNC162, and SNC243A in substituting for SNC80 and decreasing response rates were similar as indicated by overlapping 95% confidence limits (Table 1). SNC67, which is the (−)-isomer of SNC80, substituted for SNC80 in only one monkey at a cumulative dose of 3.2 mg/kg and produced primarily saline-appropriate responding in the other three monkeys. SNC67 had minimal effects on response rates up to 10.0 mg/kg. One monkey received a cumulative dose of 32.0 mg/kg SNC67, and this high dose produced tonic-clonic convulsions approximately 3 min after administration. Consequently, SNC67 was tested only up to 10.0 mg/kg in the other three monkeys.

**Effects of μ Agonists, κ Agonists, and Nonopioids.** The μ agonists fentanyl and morphine and the κ agonists enadoline and U50,488 failed to substitute for the SNC80 discriminative stimulus when administered in ½ log unit increments up to doses that eliminated responding (Fig. 3, left and center; Table 1). In two cases, mean group data for SNC80-appropriate responding were not shown in Fig. 3, because only one of four monkeys responded. First, a high dose of 0.001 mg/kg enadoline eliminated responding in three monkeys, and in the fourth monkey, this dose of enadoline produced 0% SNC80-appropriate responding and decreased response rates to 62% of control. Second, a high dose of 0.32 mg/kg U50,488 eliminated responding in three of four monkeys, and in the fourth monkey, this dose of U50,488 produced 100% SNC80-appropriate responding and decreased response rates to 23% of control. To evaluate further the effects of U50,488, the U50,488 dose-effect curve was reetermined by using ¼ log unit increments. Under these conditions, U50,488 produced primarily saline-appropriate responding in all four monkeys, including the monkey in which U50,488 substituted during the first test, and a dose of 0.32 mg/kg U-50,488 eliminated responding in all four monkeys.

Nonopioid drugs also failed to substitute for the SNC80 discriminative stimulus (Fig. 3, right; Table 1). The N-methyl-D-aspartate antagonist ketamine produced a maximum of 56% drug-appropriate responding at a dose of 3.2 mg/kg. This dose produced 83 and 29% SNC80-appropriate responding in two monkeys and decreased rates of responding to less than 10% of saline control in the other two monkeys. Cocaine occasioned less than 16% drug-appropriate responding across all doses studied.
agonists, did not significantly alter the dose-effect curve for
the discriminative stimulus effects of SNC80 (Fig. 4; Table 2).
A higher dose of 1.0 mg/kg quada-
zocine, which antagonizes both μ and κ agonists but not δ
agonists, also failed to alter significantly either the discrimina-
tive stimulus or rate effects of SNC80.

Discussion
The present study is the first demonstration that rhesus
monkeys can be trained to discriminate between SNC80 and
saline. Two lines of evidence suggest that the discriminative
stimulus effects of SNC80 were mediated by δ-opioid recep-
tors and not by μ- or κ-opioid receptors. First, the δ-selective
agonists SNC67, SNC162, and SNC243A each substituted for
SNC80, whereas μ-opioid agonists, κ-opioid agonists, and
nonopioids produced primarily vehicle-appropriate respond-
ing. Second, the δ-selective antagonist naltrindole dose-de-
pendently shifted the dose-effect curves for the discrimina-
tive stimulus and rate-decreasing effects of SNC80 to the
right. In contrast, the opioid antagonist quada-
zocine, up to doses that antagonize the effects of μ and κ agonists, did not
modify the discriminative stimulus or rate-decreasing effects
of SNC80. Taken together, these results provide the first
evidence that δ-opioid agonists serve as selective, discrimi-
native stimuli in primates.

Potency and Time Course of SNC80. SNC80 produced
dose-dependent increases in drug-appropriate responding and
decreases in response rates. The potency of SNC80 for
decreasing response rates in the present study was similar to
results obtained from a previous study of schedule-controlled
behavior in monkeys treated intermittently with δ-opioid
agonists (Negus et al., 1998). Moreover, determinations of
the potency of SNC80 at the beginning and at the end of the
study were similar. These findings suggest that tolerance did
not develop to the discriminative stimulus or rate-decreasing
effects of SNC80 under the current dosing conditions.

The time course of SNC80 in the present study also agrees
with previous results from this laboratory. Specifically, the
discriminative stimulus effects of 0.32 mg/kg SNC80 peaked
after 15 min and gradually declined over 240 min. This
duration of action is consistent with a previous study in

![Fig. 2. Discriminative stimulus and rate effects of the piperazinyl benz-
amides SNC67, SNC86, SNC162, and SNC243A. Abscissae, dose of the
test drug in milligrams per kilograms (log scale). Ordinate (top), percent-
age of responses on the SNC80-appropriate key (% Drug Responding).
Ordinate (bottom), rate of responding presented as a percentage of control
response rates. Points above “S” show data obtained after saline admin-
istration. All points show mean data (±1 S.E.M.) from four monkeys
except the following points in the top: SNC67 (1.0 mg/kg, n = 3), SNC243A (1.0 mg/kg, n = 3), and SNC162 (1.0 mg/kg, n = 3 and 3.2
mg/kg, n = 2).](image-url)

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a ND, not determined. See text for details.
b Determined twice in each of five monkeys.
c Determined twice in each of four monkeys.

either the discriminative stimulus or rate-decreasing effects
of SNC80 (Fig. 4; Table 2). A higher dose of 1.0 mg/kg quada-
zocine, which antagonizes both μ and κ agonists but not δ
agonists, also failed to alter significantly either the discrimina-
tive stimulus or rate effects of SNC80.
which a higher dose of 1.0 mg/kg SNC80 produced maximal decreases in response rates after 10 min and rates gradually returned to control values over 300 min (Negus et al., 1998).

**Effects of Other Piperazinyl Benzamides.** The greatest degree of substitution for SNC80 was obtained with the piperazinyl benzamides SNC86 [the (+)-enantiomer of (±)-BW373U86], SNC162, and SNC243A. These piperazinyl benzamides produced high levels (at least 88%) of SNC80-appropriate responding in all monkeys, and we have shown previously that these compounds produced other δ receptor-mediated effects in monkeys (Negus et al., 1998). These results also agree with the finding that (±)-BW373U86 and SNC80 shared discriminative stimulus effects in pigeons (Picker and Cook, 1998). The potencies of SNC80, SNC86, SNC162, and SNC243A for producing drug-appropriate responding and decreasing rates of responding were similar. These results were consistent with their potencies for decreasing rates of scheduled-controlled responding in other rhesus monkeys (Negus et al., 1998) and with their relative affinities at cloned human δ receptors but not cloned human μ receptors (Knapp et al., 1996).

SNC67, the (−)-enantiomer of SNC80, did not substitute for the SNC80 discriminative stimulus up to a dose of 10.0 mg/kg. These results provide evidence that the discriminative stimulus effects of SNC80 are stereoselective. A higher dose of 32.0 mg/kg SNC67 produced convulsions in one monkey and, therefore, was not tested in the other monkeys. In a previous study, a cumulative dose of 32.0 mg/kg SNC67 decreased rates of responding to less than 25% of control rates, and a larger dose of 56.0 mg/kg produced convulsions in one monkey (Negus et al., 1998). The mechanisms underlying the convulsant effects of SNC67 are not known. Both (±)-BW373U86 and SNC80 produce naltrindole-reversible convulsant effects in mice (Comer et al., 1993a; Bilsky et al., 1995), suggesting that the convulsant effects of these compounds are mediated by δ opioid receptors. However, SNC80 at doses of up to 56.0 mg/kg does not produce convulsions in rhesus monkeys (Negus et al., 1998; M.R.B., unpublished observations). To the extent that convulsions were obtained only with the (−)-enantiomer of SNC80, these results suggest that the convulsant effects of these compounds also may be stereoselective in primates.

**Effects of μ- and κ-Opioid Agonists and Nonopioids.** In rhesus monkeys, μ- and κ-opioid agonists failed to substitute for SNC80. Similarly, in pigeons, μ- and κ-opioid agonists did not substitute for DPDPE, and κ agonists did not substitute for (±)-BW373U86 (Comer et al., 1993b; Jewett et al., 1996). However, in contrast to the results of the present study, μ-opioid agonists produced high levels of substitution in pigeons trained to discriminate (±)-BW373U86 (Comer et al., 1993b; Jewett and Cook, 1998). Moreover, (±)-BW373U86 and SNC80 substituted in the majority of pigeons trained to discriminate μ agonists (Negus et al., 1996; Picker, 1997; Morgan and Picker, 1998). In contrast, (±)-BW373U86 did not substitute for the discriminative stimulus effects of the μ agonist alfentanil in monkeys (Negus et al., 1994), and μ agonists did not substitute for SNC80 in the present study. Together, these results suggest that the discriminative
stimuli associated with nonpeptidic δ-opioid agonists may be more selective in primates than in pigeons.

The monoamine reuptake inhibitor cocaine also failed to substitute for the SNC80 discriminative stimulus. These findings agree with previous studies demonstrating that cocaine did not substitute for either DPDPDE or (±)-BW373U86 in pigeons (Jewett et al., 1996; Picker and Cook, 1998). However, both peptidic and nonpeptidic δ-opioid agonists produced high levels of cocaine-appropriate responding in monkeys and rats trained to discriminate cocaine (Ukai et al., 1993; Suzuki et al., 1997; Negus et al., 1998). The reasons for this asymmetrical cross-substitution are not clear. One possibility is that the rate-decreasing effects of SNC80 are not mediated by its actions on dopaminergic systems (Negus et al., 1998). Moreover, it is important to note that asymmetric cross-generalization also has been observed between cocaine and other drug classes (Speelman et al., 1991; Rosenzweig-Lipson and Bergman, 1993).

Thus, asymmetric cross-generalizations with cocaine may reflect the ability of cocaine to produce a relatively nonselective discriminative stimulus. Overall, μ agonists, κ agonists, cocaine, and the N-methyl-d-aspartate antagonist ketamine produced primarily saline-appropriate responding in SNC80 trained monkeys. These findings suggest that the discriminative stimulus effects of SNC80 in rhesus monkeys are highly selective and are shared only by other drugs that act as δ-opioid agonists.

### Table 2

<table>
<thead>
<tr>
<th>SNC80 Responding</th>
<th>Response Rate</th>
</tr>
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<tbody>
<tr>
<td>SNC80 alone</td>
<td>0.12 (0.08–0.16)</td>
</tr>
<tr>
<td>+ Naltrindole</td>
<td>0.13 (0.01–0.20)</td>
</tr>
<tr>
<td>0.01 mg/kg</td>
<td>0.22 (0.02–0.55)</td>
</tr>
<tr>
<td>0.1 mg/kg</td>
<td>0.49 (0.33–0.67)*</td>
</tr>
<tr>
<td>1.0 mg/kg</td>
<td>3.28 (1.41–5.83)*</td>
</tr>
<tr>
<td>+ Quadazocine</td>
<td>0.16 (0.08–0.25)</td>
</tr>
<tr>
<td>0.1 mg/kg</td>
<td>0.16 (0.04–0.71)</td>
</tr>
<tr>
<td>1.0 mg/kg</td>
<td>0.26 (0.04–0.71)</td>
</tr>
</tbody>
</table>

* 5% CL after antagonist treatment do not overlap with 95% CL for SNC80 alone.

The opioid agonist quadazocine at doses of up to 1.0 mg/kg failed to antagonize the discriminative stimulus and rate-decreasing effects of SNC80. These data are concordant with previous studies in rhesus monkeys that demonstrated that 1.0 mg/kg quadazocine antagonized the behavioral effects of μ and κ agonists but not δ agonists (Bertalmio and Woods, 1987; Negus et al., 1993). Consequently, the inability of quadazocine to antagonize the discriminative stimulus or mediated primarily by its effects on dopaminergic systems (Kleven et al., 1990; Speelman et al., 1991), and dopamine also has been implicated in some of the behavioral effects of δ agonists (Longoni et al., 1991; Spina et al., 1998). In monkeys, however, the cocaine-like discriminative stimulus effects of SNC80 were not antagonized by the dopamine antagonist flupenthixol, which suggests that the cocaine-like effects of SNC80 were not mediated by its actions on dopaminergic systems (Negus et al., 1998). Moreover, it is important to note that asymmetric cross-generalization also has been observed between cocaine and other drug classes (Speelman et al., 1991; Rosenzweig-Lipson and Bergman, 1993).
rate-decreasing effects of SNC80 suggests that these effects were not mediated by μ-κ-opioid receptors.

**Implications.** As the current study demonstrates, discriminations based on δ-opioid agonists in rhesus monkeys can provide an effective approach to the rapid identification and evaluation of novel δ-opioid ligands. These findings also have implications for the development of δ agonists as analgesics.

Under some conditions, δ agonists produce antinociceptive effects (Negus et al., 1998), and, therefore, they are being evaluated as possible alternatives to μ agonists in rhesus monkeys (Kumor et al., 1986; Kreek, 1992), and these subjective effects have been described as “euphoric” in humans (Kumor et al., 1986; Kreek, 1992), and these subjective effects might reduce patient acceptance. Although the clinical implications of δ agonist-induced, subjective effects remain to be determined, the discriminative stimulus effects of δ agonists in the present study suggest that δ agonists such as SNC80 might produce different subjective effects than either μ or κ agonists in humans.

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**References**


