ABSTRACT

There is accumulating evidence that kappa opioid agonists attenuate cocaine’s behavioral effects, and we recently reported that the kappa opioid agonists ethylketocyclazocine (EKC) and U50–488 decreased cocaine self-administration by rhesus monkeys. In the present study, we first examined the effects of acute intramuscular administration of six kappa opioid agonists on responding maintained by food under an FR30 schedule. Each kappa agonist produced dose-dependent decreases in schedule controlled behavior, and the relative potencies were enadoline ≥ bremazocine > Mr2033 > spiradoline = spiradoline > PD117302. We then studied the effects of chronic administration of these kappa agonists in monkeys responding under a second order schedule of food delivery and cocaine self-administration. The effects of 10 days of intravenous treatment with three ary lacetamides [enadoline (0.00032–0.0032 mg/kg/hr), (−) spiradoline (0.0032–0.018 mg/kg/hr), PD117302 (0.032–0.32 mg/kg/hr)] and three benzonorphans [bremazocine (0.00032–0.0032 mg/kg/hr), Mr2033 (0.0032–0.032 mg/kg/hr), cyclazocine (0.001–0.10 mg/kg/hr)] were compared with saline treatment. Enadoline (0.001 and 0.0032 mg/kg/hr) and bremazocine (0.0032 mg/kg/hr) significantly decreased cocaine self-administration (0.01 mg/kg/injection) (P < .05–.01). Cyclazocine (0.001–0.10 mg/kg/hr), (−) spiradoline (0.0032–0.018 mg/kg/hr) and PD117302 (0.032–0.32 mg/kg/hr) had no significant effects on cocaine self-administration across the dose-range studied. When gradually increasing doses of enadoline (0.0032–0.01 mg/kg/hr) or Mr2033 (0.0032–0.032 mg/kg/hr) were administered over 28 consecutive days, cocaine self-administration was dose-dependently decreased in all monkeys. Food-maintained responding was usually decreased at doses that decreased cocaine self-administration. Adverse side effects (emesis and sedation) were transient, and laboratory indices of hematology and blood chemistry were normal throughout chronic enadoline and Mr2033 treatment. These data extend our earlier findings with EKC and U50,488 and suggest that kappa opioid agonists may be a useful approach to the development of new pharmacological treatments for cocaine dependence. The extent to which undesirable side effects may limit their clinical usefulness remains to be determined.

Cocaine abuse remains a serious public health problem (NIDA, 1997) and as yet no consistently effective pharmacotherapy has been identified (Mendelson and Mello, 1996). Kappa opioid agonists may provide a new approach to the continuing search for effective treatment medications (Archer et al., 1996). There is increasing evidence that kappa opioid agonists modulate the behavioral and neurobiologic effects of cocaine. Cocaine-induced place preferences and hyperactivity, as well as sensitization to cocaine-induced hyperactivity and stereotypies, were blocked or decreased by kappa opioid agonists in rodents (Crawford et al., 1995; Heidbreder et al., 1995; Shippenberg et al., 1996; Suzuki et al., 1992; Ukai et al., 1994). Behavioral studies in squirrel monkeys have shown that kappa opioid agonists antagonized the discriminative stimulus and rate-stimulating effects of cocaine (Bergman and Spealman, 1997; Spealman and Bergman, 1992, 1994).

It has been postulated that these behavioral interactions with cocaine reflect kappa opioid agonist-related modulation of dopaminergic activity. Kappa agonists attenuate dopamine release from the nucleus accumbens (DiChiara, 1995; DiChiara and Imperato, 1988; Maisonneuve et al., 1994) and decrease striatal dopamine levels in rats (Devine et al., 1993; Donzanti et al., 1992, Spanagel et al., 1992). Moreover, administration of the kappa agonist spiradoline decreased cocaine-induced expression of c-fos and zif-268 immediate-early genes in rat dorsal striatum and cortex (Steiner and Gerfen, 1995). In addition to kappa opioid agonist modulation of the effects of cocaine, there is emerging evidence that cocaine also influences endogenous kappa opioid systems. The nucleus accumbens contains kappa opioid receptors (Mansour et al., 1987, 1988, 1994) as well as high levels of an opioid peptide dynorphin, which binds to kappa receptors (Chavkin et al., 1982; Hokfelt et al., 1984). Cocaine administration...
up-regulates kappa opioid receptors (Hurd and Herkenham, 1993; Unterwald et al., 1994) and increases levels of both dynorphin and dynorphin mRNA (Daunais and McGinty, 1994, 1995; Daunais et al., 1995; Hanson et al., 1995; Hurd and Herkenham, 1993). Thus, it is possible that cocaine directly stimulates kappa opioid systems, and this in turn modulates the effects of cocaine (see Hyman and Nestler, 1996, for discussion).

Kappa opioid agonist inhibition of the behavioral effects of cocaine suggests that these drugs might also reduce cocaine self-administration, but relatively little is known about the effects of kappa opioid agonists on cocaine-maintained responding. Glick and co-workers. (1995) were the first to report that acute administration of the kappa agonists U50,488 and spiradoline decreased self-administration of an intermediate dose of cocaine (0.4 mg/kg/injection) (Glick et al., 1995). U50,488 and spiradoline also decreased water-maintained responding, but these kappa agonists were slightly more potent in decreasing responding maintained by cocaine than by water. These effects of U50,488 were completely antagonized by the kappa receptor selective antagonist nor-BNI (Portoghese et al., 1987), which had no effects on cocaine-maintained responding when administered alone (Glick et al., 1995). U50,488 (2.5 mg/kg) also decreased initiation of cocaine self-administration (30 μg/injection) in rats (Kuzmin et al., 1997). However, at lower cocaine doses (7 or 15 μg/injection) that were not reinforcing under placebo conditions, U50,488 significantly increased initiation of cocaine-maintained responding (Kuzmin et al., 1997). These effects of the aryloacetamide kappa agonists U50,488 and spiradoline also have been observed with benzomorphans showing kappa opioid activity. For example, cyclazocine (0.5 mg/kg i.p.) also reduced cocaine self-administration with minimal effects on water-maintained responding in rats, and these effects persisted for 48 hr (Archer et al., 1996).

We recently examined the effects of chronic treatment with two kappa opioid agonists EKC and U50,488, on cocaine and food self-administration in rhesus monkeys (Negus et al., 1997). Ten days of treatment with EKC (0.002–0.032 mg/kg/hr) or U50,488 (0.032–0.1 mg/kg/hr) dose-dependently decreased cocaine self-administration at unit doses of 0.01 and 0.032 mg/kg/injection that were at the peak of the cocaine-dose-effect curve. These decreases in cocaine self-administration were often sustained throughout the 10 days of treatment. Food-maintained responding was usually decreased at doses that reduced cocaine self-administration but often less than cocaine-maintained responding. Both kappa agonists often produced emesis and sedation during the first 2 days of treatment, but tolerance developed rapidly to these effects. The kappa antagonist nor-BNI (3.2 mg/kg) blocked the effects of the highest doses of EKC (0.032 mg/kg/hr) and U50,488 (0.1 mg/kg/hr) on cocaine- and food-maintained responding, but nor-BNI had no behavioral effects alone. We concluded that chronic administration of kappa opioid agonists produced a dose-dependent, kappa receptor-mediated and often sustained decrease in cocaine self-administration (Negus et al., 1997).

Our encouraging findings with the benzomorphan EKC and the aryloacetamide U50,488 prompted us to study the effects of a number of structurally related kappa agonists on cocaine and food self-administration to determine the generality of our initial findings (Negus et al., 1997). We were also interested in learning if there were consistent differences in the effects of benzomorphans and aryloacetamides on cocaine- and food-maintained responding. In our initial study, the benzomorphan EKC produced fewer undesirable side effects than the aryloacetamide U50,488 at doses that decreased cocaine self-administration (Negus et al., 1997). Moreover, we found that nor-BNI was less effective in antagonizing the effects of EKC on cocaine self-administration than in blocking the effects of U50,488 (Negus et al., 1997), and similar results have been reported in studies of kappa agonist-mediated diuresis (Takeemori et al., 1988) and antinociception (Broadbear et al., 1994; Butelman et al., 1993a). It is not clear if these differences between EKC and U50,488 reflect activity at different kappa receptor subtypes and/or the mu opioid agonist effects of EKC (compare Broadbear et al., 1994; Butelman et al., 1993b; Clark et al., 1989; Gmerek et al., 1987). However, several lines of evidence suggest the existence of kappa receptor subtypes (Nock et al., 1990; Rothman et al., 1990; Su, 1985; Zuki et al., 1988), and if different kappa agonists act preferentially at different kappa receptor subtypes in primates, this also could influence cocaine’s interactions with kappa opioids. For example, behavioral studies indicated that both nor-BNI and the low efficacy kappa agonist dynorphin 1–13 selectively antagonized the effects of the aryloacetamides U50,488 and U69,593 but did not antagonize the effects of the aryloacetamide enadoline or the benzomorphans bremazocine and Mr2033 (Butelman et al., 1993a, 1995). These findings were interpreted to suggest that many aryloacetamides may act at one kappa receptor subtype (designated the kappa-1 site), whereas enadoline and the benzomorphans may act preferentially at a different kappa receptor subtype.

In the present study, we compared the behavioral effects of three other aryloacetamide kappa agonists (enadoline, (-) spiradoline and PD117302) and three other benzomorphans (bremazocine, Mr2033 and cycloclazocine). We selected these kappa opioid agonists in part because four [enadoline, (-) spiradoline, Mr2033 and cycloclazocine] had been administered to humans for other applications (Archer et al., 1996; Jaffe and Brill, 1966; Martin et al., 1965, 1966; Pfeiffer et al., 1986; Reeve et al., 1994; Rimoy et al., 1991). Moreover, all of these compounds have been shown to produce similar kappa opioid-mediated effects under some conditions (e.g., Brandt and France, 1996; Dykstra et al., 1987; France et al., 1994, 1998; Smith and Picker, 1995). However, there may be important differences in their pharmacological profiles that could influence their interactions with cocaine. For example, although all of these compounds have high affinity for kappa opioid receptors as determined by in vitro receptor binding assays, they differ in relative selectivity for kappa receptors in comparison to mu and delta opioid receptors (Emmerson et al., 1994; France et al., 1994; Magnan et al., 1982; Wood, 1982). Specifically, the aryloacetamides enadoline, spiradoline and PD117302 have >50-fold selectivity for kappa vs. mu receptors and >1000-fold selectivity for kappa vs. delta receptors (Emmerson et al., 1994; France et al., 1994). In contrast, the benzomorphans bremazocine, Mr2033 and cycloclazocine, have much lower selectivity for kappa vs. mu and delta opioid receptors (Emmerson et al., 1994; Magnan et al., 1982; Wood, 1982).

Three sets of experiments were conducted to evaluate the effects of three aryloacetamide and three benzomorphan
kappa agonists on cocaine- and food-maintained behaviors. In Study 1, schedule controlled responding maintained by food was used to determine the behaviorally active dose range of these six kappa agonists after acute intramuscular administration. The effects of EKC and U50, 488 were also examined in Study 1 because these compounds reduced cocaine self-administration in our previous study (Negus et al., 1997). Several doses of each kappa agonist were administered to monkeys trained to respond for banana pellets, and the acute effects of each drug on response rates were examined. This procedure is useful for determining the potency of novel compounds (Gatch et al., 1996; Negus et al., 1993, 1994). Results from Study 1 provided an empirical basis for selecting doses to use for chronic treatment in subsequent studies of drug self-administration. In Study 2, the chronic effects of six structurally diverse kappa agonists on cocaine- and food-maintained responding were evaluated. The operant behavioral procedures used in Study 2 were identical to those in our previous report of the effects of EKC and U50,488 on cocaine and food self-administration by rhesus monkeys (Negus et al., 1997). In Study 3, the effects of 28 days of chronic enadoline and Mr2033 administration on cocaine- and food-maintained responding, as well as laboratory indices of medical status, were examined. Enadoline and Mr2033 were selected for these additional studies because they were most effective in decreasing cocaine self-administration in Study 2. Study 3 was designed to determine if gradually ascending doses of kappa agonists decreased the severity of adverse side effects (emesis, sedation) sometimes seen immediately after abrupt administration of high doses of kappa agonists. A second goal was to evaluate the extent to which tolerance developed to the effects of these kappa agonists on cocaine- and food-maintained responding over 4 weeks of treatment.

Methods

Subjects

Four female adult rhesus monkeys (75C, 343T, 371A and CH96) were subjects in Study 1, the acute effects of kappa opioid agonists on operant performance maintained by food. Ten adult rhesus monkeys (four females: 075F, R801, 89B157 and CH701; six males: 152F, 89B058, 89B084, 90E, 90B134, 90B154) were subjects in Studies 2 and 3, the effects of chronic kappa agonist treatment on cocaine and food self-administration. All monkeys had a history of cocaine and food self-administration on the second-order schedule of reinforcement described below. Monkeys 075F, CH701, 152F, 89B058, 89B084, 90E and 90B134 were subjects in our previous studies of the effects of EKC and U50,488 on cocaine- and food-maintained responding (Negus et al., 1997). R801 had a history of both alcohol and cocaine self-administration, and monkeys 89B157 and 90B154 had histories of cocaine self-administration. This report is one of a series of studies designed to evaluate the effects of potential treatment medications on cocaine self-administration in rhesus monkeys, and details of the training procedures have been described previously (Mello et al., 1990; Negus et al., 1995, 1997).

Monkeys weighed 4.4 to 10.9 kg and were fed a diet of multiple vitamins, fresh fruit and vegetables (2 pieces per day) and Lab Diet Jumbo Monkey biscuits (3–5 per day; PMI Feeds, St. Louis, MO). This diet was sufficient to maintain constant body weights (±10% of mean body weight) in these adult monkeys. In addition, monkeys could earn up to 100 banana pellets (1 g) (P.J. Noyes, Lancaster, NH) during daily operant sessions as described below. Water was continuously available. A 12-hr light/dark cycle was in effect (lights on from 7 a.m. to 7 p.m.), and room lights were off during sessions conducted when the light cycle was in effect (see below).

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 licensed 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 provided an opportunity for environmental manipulation and enrichment (Line et al., 1989).

Apparatus and Operant Response Requirements

Schedule controlled behavior studies. Monkeys lived in well ventilated stainless steel cages (60 × 65 × 75 cm), and all experiments were conducted in the home cage. Each cage had a detachable operant panel (28 × 28 cm) mounted on the front. The operant panel contained three plastic keys (6.4 × 6.4 cm) spaced 2.54 cm apart, and only the center key was used in this study. Each key could be transilluminated by red, green or white 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, green or white stimulus lights (Superbright LEDs). A pellet dispenser (Gerbrands, model GS210) was mounted above the panel to deliver 1 g of fruit-flavored food pellets (Precision Primate Pellets Formula L/I Banana Flavor, P. J. Noyes) to a food receptacle mounted on the cage beneath the operant response panel. Monkeys were trained to press the transilluminated center key to acquire food pellets. On completion of an FR of 30 responses, one banana-flavored food pellet (1 g) was delivered to a food receptacle mounted below the operant panel on the front of the cage. Schedules of reinforcement were controlled by a MED-PC interface and an IBM compatible computer programmed in MEDSTATE Notation (MED Associates, East Fairfield, VT).

Training sessions consisted of 5 cycles, each lasting for 15 min. Each cycle consisted of a 10-min pretreatment interval followed by a 5-min response period. During the pretreatment intervals, no stimulus lights were illuminated and responding produced no scheduled consequences. During the response period, the center key was transilluminated and, and monkeys could respond for up to 10 food pellets on an FR 30 schedule of reinforcement. If all 10 food pellets were earned before 5 min had elapsed, the stimulus lights were turned off, and responding produced no scheduled consequences for the remainder of that response period. All monkeys were trained until they responded for food at rates of >1.0 responses/sec during all five cycles for 10 consecutive days. Sessions were conducted at the same time of day, 5 days a week in all monkeys.

During test sessions, the effects of kappa agonists on rates of food-maintained responding were evaluated. Intramuscular drug injections were administered at the beginning of each cycle on test days, and sham control injections were administered periodically on training days. Test sessions were conducted no more than twice a week and only after a training session during which the monkeys responded at rates of >1.0 responses/sec for all five cycles. Test sessions were conducted using a cumulative dosing procedure in which a dose of each kappa agonist, which increased the total cumulative dose by 1/2 log units, was administered (intramuscular) at the beginning of each 15-min cycle. Otherwise, test sessions were identical to training sessions.

Drug self-administration studies. 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 mounted on the front wall. The operant panel was identical to that used in studies of food-maintained behavior described earlier. Two syringe pumps (model B5P1E, Braintree Scientific, Braintree, MA; or model 980210, Harvard Apparatus, South Braintree, MA) were used to administer drugs into the intramuscular vein. Test sessions were conducted in the home cages of the monkeys. The home cages were identical to that used in studies of food-maintained behavior described earlier. Two syringe pumps (model B5P1E, Braintree Scientific, Braintree, MA; or model 980210, Harvard Apparatus, South Braintree, MA) were used to administer drugs into the intramuscular vein. Test sessions were conducted in the home cages of the monkeys. The home cages were identical to that used in studies of food-maintained behavior described earlier.
Natick, MA) were mounted above each cage for delivery of saline or drug solutions through the two lumens of the intravenous catheters.

The second-order schedule response requirement was identical for food and drug acquisition. The final schedule was an FR4 (VR 16:S) for all monkeys except for monkey 90B154, for which the schedule was an FR2 (VR16:S). Thus, an average of 32 or 64 responses was required for each injection or food pellet. 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 requirement of the second-order schedule was followed by a 10-sec time-out period, during which the stimulus light illuminating the center response key was turned off for 10 sec and responding had no scheduled consequences. In addition, the appropriate colored stimulus light (red for food, green for injections) was illuminated for 1 sec below the center response key. The experimental room was dark during all food and drug sessions.

There were four periods of food availability and four cocaine self-administration sessions during each 24-hr period. Food sessions began at 11 a.m., 3 p.m., 7 p.m. and again at 6 a.m. the next morning. Cocaine sessions followed food sessions at 12 noon, 4 p.m., 8 p.m. and 7 a.m. the next morning. Each cocaine or food session lasted for one hr or until a maximum of 20 injections or 25 banana pellets had been delivered, whichever occurred first. Schedules of reinforcement and data collection were controlled by Apple IIGS computers located in a separate room.

**Chronic Kappa Opioid Agonist Treatment**

Two studies were conducted to evaluate the effects of kappa opioid agonists on cocaine self-administration. Once cocaine dose-effect curves were determined, the effects of saline and kappa opioid agonists on the self-administration of cocaine (0.01 mg/kg/injection) were evaluated (Study 2). Kappa agonist doses that significantly decreased 0.01 mg/kg/injection cocaine self-administration were also evaluated at a higher cocaine unit dose (0.032 mg/kg/injection) to determine if these effects were surmountable. The behaviorally-active dose ranges of the six kappa opioid agonists evaluated in Study 2 were based on results from Study 1. In Study 2, the behavioral effects of three benzomorphans, bremazocine (0.00032–0.0032 mg/kg/hr), Mr2033 (0.0032–0.032 mg/kg/hr) and cyclazocine (0.001–0.10 mg/kg/hr) were compared with the effects of three aryloacetamides, enadoline (0.00032–0.0032 mg/kg/hr), and spiradoline (0.0032–0.018 mg/kg/hr) and PD117302 (0.032–0.32 mg/kg/hr). All kappa agonists were administered intravenously through one lumen of a chronically implanted double-lumen catheter (see description of surgical procedures below). Doses of each drug were studied in an irregular order in groups of three or four monkeys. Each kappa agonist dose was evaluated for 10 consecutive days to be comparable to our previous studies of the effects of chronic treatment on cocaine- and food-maintained responding (Negus et al., 1995, 1996, 1997).

Experimental procedures were identical to those used in our previous evaluations of other kappa agonists (EK2 and U50,488) (Negus et al., 1997). Most kappa opioid agonists are relatively short-acting, so it was necessary to give multiple injections throughout the experimental day. Kappa opioid agonist injections were administered intravenously every 20 min, for a total of 3 injections per hour and 69 injections per day. No injections were delivered between 9:30 a.m. and 10:30 a.m., and during this period, monkeys received their morning ration of food, and their health status was evaluated by the technical staff. During saline base-line treatment, 0.1 ml of saline was delivered every 20 min for a total of 6.9 ml per 23 hr. At the conclusion of each 10-day treatment period, base-line conditions (the maintenance dose of 0.032 mg/kg/injection cocaine and saline treatment) were reinstated for ≥4 days and until responding for cocaine and food returned to base-line levels.

In Study 3, the effects of gradually ascending doses of enadoline and Mr2033 on cocaine- and food-maintained responding were examined. One goal of this study was to determine if administration of gradually ascending doses of these kappa agonists reduced the incidence and severity of adverse side effects often noted after abrupt introduction of relatively high doses. A second goal was to determine if tolerance developed to the effects of these kappa agonists on cocaine- and food-maintained responding over 28 days of continuous kappa agonist treatment. A third objective was to evaluate the relative safety of several weeks of kappa opioid agonist exposure by monitoring hematology and blood chemistry indices.

Each experiment in Study 3 lasted for 42 days. The unit dose of cocaine (0.01 mg/kg/injection) was at the peak of the cocaine dose-effect curve and remained available throughout Study 3. On days 1 to 7, monkeys were treated with repeated saline injections. On days 8 to 14, a low dose of kappa agonist was administered as described earlier (0.00032 mg/kg/hr enadoline or 0.0032 mg/kg/hr Mr2033). The kappa agonist dose was increased from days 15 to 21 to 0.001 mg/kg/hr enadoline or 0.01 mg/kg/hr Mr2033 and from days 22 to 28 to 0.0032 mg/kg/hr Mr2033. In the enadoline experiment, the dose of enadoline was increased to 0.01 mg/kg/hr from days 29 to 35 in 2 monkeys, whereas in the other 2 monkeys, the dose remained at 0.0032 mg/kg/hr during this time. In the Mr2033 experiment, all monkeys continued to receive 0.032 mg/kg/hr Mr2033 from days 29 to 35. Kappa agonist treatment was terminated after 28 days (on study day 35), and saline treatment was reinstated from days 36 to 42. There was no drug-free interval between successive treatment doses as in Study 2. Monkeys were weighed under mild ketamine sedation (3 mg/kg), and blood samples for analysis of hematology and blood chemistry indices were collected from the saphenous vein at the end of each dose condition (on study days 7, 14, 21, 28 and 35).

Formal behavioral observations were conducted each morning between 9 and 9:30 a.m. During this time, each monkey was observed for 5 consecutive min for the presence or absence of the following signs: sedation, salivation, muscle twitches, convulsions, unusual tongue movements, yawning, excessive scratching/grooming, coughing/retching/vomiting, excessive vocalization and wet dog shakes. Each of these signs has been associated previously either with acute administration of kappa opioid agonists or with withdrawal from chronic administration of kappa agonists (Dykstra et al., 1987; Gmerek et al., 1987). The observer was not blind to the treatment condition. In addition to these formal observation periods, monkeys were also observed at least twice every day for evidence of behavioral toxicity. Blood sampling and weighing were conducted after the formal observation period between 9:30 and 10:00 a.m. (i.e., −1 hr after the last injection of saline or the kappa agonist). On discontinuation of treatment with enadoline or Mr2033, monkeys were observed for withdrawal signs that may occur after long-term kappa agonist treatment (Gmerek et al., 1987).

**Surgical Procedures**

Double-lumen silicone rubber catheters (inside diameter, 0.7 mm; outside diameter, 2.0 mm) were implanted in the jugular or femoral vein and exited in the midscapular region. All surgical procedures were performed under aseptic conditions. Monkeys were initially sedated with ketamine (5 mg/kg s.c.), 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. After insertion of a tracheal tube, anesthesia was maintained with halothane (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 units/day i.m.) was administered every day for 5 days. The intravenous 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 intravenous administration of either ketamine (5 mg/kg) or the short-acting barbiturate methohexitol (3 mg/kg). The catheter was considered to be patent if
intrapavaneous administration of ketamine or methohexital produced a loss of muscle tone within 10 sec.

Drug Preparation

Cocaine hydrochloride was obtained in crystalline form from the National Institute on Drug Abuse, National Institutes of Health (Bethesda, MD), and purity was certified to be $>$98%. All drugs were dissolved in sterile saline or sterile water and were filter-sterilized using a 0.22-μm Millipore filter. Drugs were stored in pyrogen-free vials. In Studies 2 and 3, cocaine, and each kappa agonist were delivered intravenously in a volume of 0.1 ml/injection. Enadoline and PD117302 were kindly donated by Warner-Lambert Pharmaceuticals, Parke-Davis Research Division (Ann Arbor, MI), and Mr2033 was provided by Boehringer Ingelheim (Ridgefield, CT). (−)-Spiradoline was generously provided by Upjohn Laboratories (Kalamazoo, MI). Cyclazocine was provided by the late Dr. Sydney Archer, Rensselaer Polytechnic Institute (Rensselaer, NY). Bremazocine was purchased from Research Biochemicals International (Natick, MA).

Data Analysis

Study 1. Rates of food-maintained responding in each cycle were converted to percent of control using the average rate from the previous training day as the control value. Dose-effect curves for each kappa agonist were constructed by plotting the percent control rate of responding as a function of dose. For cumulative kappa agonist dose-effect curves, the dose that produced a 50% decrease in the percent control rate of responding (ED$_{50}$) was determined by linear regression for each subject. ED$_{50}$ values were averaged for individual subjects to yield mean ED$_{50}$ values ± S.E.M. (95% confidence). The criterion for a significant ED$_{50}$ was set at $P$ < .05.

Studies 2 and 3. The dependent variables were the number of food pellets and the number of cocaine injections delivered each day. The effects of kappa opioids and saline treatment on the numbers of injections/day and food pellets/day were evaluated using a two-factor ANOVA, with in-group means (Morrison, 1990). A one-factor ANOVA for repeated measures was used to evaluate the significance of changes in body weight, food, food-maintained responding, and saline are shown in figure 3. In comparison to the saline treatment base-line, Mr2033 (0.01 and 0.032 mg/kg/hr), bremazocine (0.0032 mg/kg/hr) and enadoline (0.001 and 0.0032 mg/kg/hr) each produced statistically significant decreases ($P$ < .05–.01) in self-administration of cocaine at a

Results

Acute Effects of Kappa Opioid Agonists on Food-Maintained Responding (FR30)

Control performance. Average rates of responding on the FR30 schedule ranged from 1.89 ($±$0.08) to 2.58 ($±$0.13) responses/sec for individual monkeys. The overall average response rate during training sessions conducted immediately before test sessions was 2.22 ($±$0.15) responses/sec.

Acute effects of kappa opioid agonists. As shown in figure 1, each of the eight kappa agonists produced dose-related decreases in rates of food-maintained responding. There was a significant difference between mean ED$_{50}$ values (mg/kg) for these kappa agonists ($P$ = .0001). Individual means comparisons indicated the following order of relative potencies ($±$S.E.M.): enadoline [0.0010 ($±$0.0002)], bremazocine [0.0014 ($±$0.0003)], EKC [0.0047 ($±$0.0008)], Mr2033 [0.0079 ($±$0.0018)], cyclazocine [0.016 ($±$0.005)], spiradoline [0.016 ($±$0.003)], PD117302 [0.054 ($±$0.005)], U50,488 [0.075 ($±$0.003)]. The acute doses of EKC and U50,488 in units of mg/kg intramuscularly that decreased food-maintained responding in this assay of schedule-controlled behavior were similar to the doses of EKC and U50,488 in units of mg/kg/hr intravenously that decreased cocaine self-administration in our previous study (Negus et al., 1997). Thus, this approach to identifying the behaviorally active dose-range of novel compounds to be used in subsequent studies of cocaine self-administration (Study 2) was justified by comparison with our previous study (Negus et al., 1997).

Effects of Chronic Kappa Opioid Agonist Administration on Cocaine Self-Administration (Study 2)

Control performance. Baseline cocaine- and food-maintained responding before each kappa agonist treatment is shown for each study group in figures 2 and 3. During saline control treatment, cocaine (0.01 and 0.032 mg/kg/injection) maintained high levels of responding, and monkeys self-administered an average of 70 to 80 injections per day. Food-maintained responding was somewhat more variable, and averaged between 80 and 100 pellets per day.

Chronic effects of kappa opioid agonists. Group data for the effects of 10 days of treatment with benzomorphon kappa agonists and saline on operant responding are shown in figure 2 and the effects of arylacetamide kappa agonists and saline are shown in figure 3. In comparison to the saline treatment base-line, Mr2033 (0.01 and 0.032 mg/kg/hr), bremazocine (0.0032 mg/kg/hr) and enadoline (0.001 and 0.0032 mg/kg/hr) each produced statistically significant decreases ($P$ < .05–.01) in self-administration of cocaine at a
unit dose of 0.01 mg/kg/injection, and these effects were sustained over 10 days of treatment (figs. 2 and 3). These decreases in cocaine self-administration ranged between 48% and 64% of saline treatment base-line levels. Mr2033 (0.032 mg/kg/hr) and bremazocine (0.0032 mg/kg/hr) also significantly decreased self-administration of a higher unit dose of cocaine (0.032 mg/kg/injection) in comparison to the saline treatment base-line (fig. 2) (P < .05–.01). Although there was a trend toward a dose-dependent decrease in cocaine (0.032 mg/kg/injection) self-administration during enadoline treat-
ment (0.001 and 0.0032 mg/kg/hr), the observed decreases of 19% and 39%, respectively, were not significantly different from baseline (fig. 3). In contrast, cyclazocine (0.001–0.10 mg/kg/hr), (-) spiradoline (0.0032–0.018 mg/kg/hr) and PD117302 (0.032–0.32 mg/kg/hr) had no statistically significant effects on cocaine self-administration (0.01 mg/kg/injection) across the dose range studied (figs. 2 and 3). The effects of the highest dose of spiradoline (0.018 mg/kg/hr) on 0.032 mg/kg/injection cocaine self-administration were examined, and spiradoline did not significantly decrease cocaine self-administration (fig. 3). Because cyclazocine and PD117302 did not decrease self-administration of 0.01 mg/kg/injection cocaine (figs. 2 and 3), and quantities of these drugs were limited, their effects on self-administration of a higher unit dose of 0.032 mg/kg/injection cocaine were not examined.

**Effects of Chronic Kappa Opioid Agonist Administration on Food-Maintained Responding (Study 2)**

Each kappa agonist at doses that significantly decreased cocaine self-administration also decreased food self-administration with two exceptions. Mr2033 (0.01 mg/kg/hr) selectively decreased 0.01 mg/kg/injection cocaine self-administration and Mr2033 (0.032 mg/kg/hr) significantly reduced 0.032 mg/kg/injection cocaine self-administration, without significant reductions in food self-administration (fig. 2). In contrast, endadoline (0.001 and 0.0032 mg/kg/hr) and spiradoline (0.01 and 0.018 mg/kg/hr) significantly reduced food-maintained responding (P < .05–.10) at doses that did not affect cocaine-maintained responding (fig. 3). Cyclazocine and PD117302 had no significant effects on food-maintained responding across the dose-range studied (figs. 2 and 3).

**Daily Patterns of Cocaine- and Food-Maintained Responding (Study 2)**

Daily patterns of responding during treatment with the three kappa agonists (enadoline, Mr2033 and bremazocine) that were most effective in reducing cocaine self-administration in comparison to the saline treatment baseline are shown in figure 4. Cocaine- and food-maintained responding during 10 days of treatment with the lowest dose of endadoline, Mr2033, and bremazocine that significantly decreased cocaine self-administration in figs. 2 and 3 are shown for each group of monkeys in figure 4 (left panel). Enadoline (0.001 mg/kg/hr) produced a sustained decrease in cocaine self-administration, whereas food-maintained responding started to increase after 3 days of treatment and this trend was statistically significant (P < .01). Mr2033 (0.01 mg/kg/hr) also decreased cocaine self-administration by 55%, and this decrease was sustained across the 10 days of treatment. Mr2033 also reduced food-maintained responding by 32%, and this decrease was less than the decrease in cocaine self-administration. Bremazocine (0.0032 mg/kg/hr) treatment also resulted in a significant and sustained decrease in cocaine-maintained responding over 10 days of treatment, but this decrease was not selective. Food-maintained responding was initially decreased, but tended to increase after 4 days of bremazocine treatment (P < .01).

There were differences between individual monkeys in response to kappa agonist treatment, and some monkeys were very sensitive to the effects of kappa opioids on cocaine-maintained responding. Figure 4 (right) shows daily patterns of responding for kappa-sensitive individual monkeys during 10 days of treatment with the same doses of the kappa agonists shown for each group in the left. These kappa-sensitive monkeys each had relatively selective decreases in cocaine self-administration but there were differences in the duration of the kappa agonist effects. For example, during enadoline treatment (0.001 mg/kg/hr), cocaine-maintained responding was initially decreased but returned toward control levels by days 9 and 10 of treatment in monkey 152F (fig. 4, row 1). In contrast, Mr2033 treatment (0.01 mg/kg/hr) resulted in a sustained and selective decrease in cocaine self-administration over the 10 days of observation in monkey 90B154. Bremazocine treatment (0.0032 mg/kg/hr) resulted in a sustained and selective decrease in cocaine self-administration in monkey 075F.

Daily patterns of responding during 10 days of treatment with the three kappa agonists (cyclazocine, spiradoline and PD117302) that were least effective in reducing cocaine self-administration in comparison to the saline-treatment baseline are shown for each group in figure 5 (left). Cyclazocine (0.0032 mg/kg/hr) initially decreased food- more than cocaine-maintained responding, but levels of food acquisition returned toward baseline after 5 days of treatment. Spiradoline (0.018 mg/kg/hr) also decreased food-maintained responding more than cocaine-maintained responding, and these decreases were sustained over 10 days of treatment. PD117302 (0.32 mg/kg/hr) decreased both cocaine- and food-maintained responding during the first 2 days of treatment; then, a gradual recovery began on day 3.

Figure 5 (right) shows daily patterns of responding for
Effects of Gradually Ascending Doses of Kappa Opioid Agonists (Study 3)

Safety and side effects. Because enadoline and Mr2033 were most effective in selectively reducing cocaine self-administration in Study 2, we evaluated these two compounds for safety and effectiveness over 28 days in Study 3. Concurrent measurement of body weight, hematological status and blood chemistry indices are summarized in tables 1 and 2. The pre-kappa opioid base-line values were all in the normal range for rhesus monkeys (Loeb and Quimby, 1989; McClure, 1975). Although these monkeys had a long history of cocaine self-administration, their blood chemistry values were comparable to values measured in drug-naive monkeys studied in this laboratory. There were no clinically significant changes in laboratory indices of hematocrit, white blood cell count, blood urea nitrogen, creatinine, serum alanine aminotransferase or blood glucose levels during 28 consecutive days of treatment with ascending doses of enadoline (table 1) or Mr2033 (table 2). There was a small but statistically significant increase in body weight as the doses of enadoline increased (table 1). These increases in body weight were unexpected in view of enadoline's diuretic effects (Reece et al., 1994) and may reflect sedation and associated decreases in caloric utilization during kappa agonist treatment.

Administration of gradually ascending doses of enadoline and Mr2033 decreased the incidence of vomiting occasionally seen when high doses of kappa agonists were administered after a period of saline control treatment (Study 2; Negus et al., 1997). Vomiting was seen occasionally at the transition between administration of intermediate and high doses of kappa agonists. Similarly, mild sedation was evident during the first 2 to 3 days of high dose treatment. Figures 6 and 7 show the effects of saline and gradually increasing doses of enadoline or Mr2033 on cocaine- and food-maintained re-

TABLE 1
Effects of chronic treatment with enadoline on body weight, hematology and blood chemistry

<table>
<thead>
<tr>
<th>Endpoint Measure</th>
<th>Saline</th>
<th>0.00032</th>
<th>0.0001</th>
<th>0.0032</th>
<th>0.0032 or 0.01</th>
</tr>
</thead>
<tbody>
<tr>
<td>Weight (kg)</td>
<td>8.5 (1.0)</td>
<td>8.9 (1.2)</td>
<td>9.2 (1.2)*</td>
<td>9.2 (1.1)*</td>
<td>9.3 (1.2)*</td>
</tr>
<tr>
<td>Hematology</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>39.0 (1.8)</td>
<td>40.8 (0.8)</td>
<td>39.0 (1.7)</td>
<td>37 (1.1)</td>
<td>36.5 (1.7)</td>
</tr>
<tr>
<td>WBC count (10^3/μl)</td>
<td>9.0 (1.0)</td>
<td>9.3 (0.8)</td>
<td>8.5 (0.4)</td>
<td>8.2 (0.8)</td>
<td>8.0 (0.7)</td>
</tr>
<tr>
<td>Blood chemistry</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BUN (mg/dl)</td>
<td>15.2 (0.5)</td>
<td>14.8 (0.6)</td>
<td>15.2 (0.5)</td>
<td>15.2 (0.5)</td>
<td>14.0 (0.4)</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>0.83 (0.09)</td>
<td>0.88 (0.09)</td>
<td>0.73 (0.11)</td>
<td>0.85 (0.12)</td>
<td>0.88 (0.09)</td>
</tr>
<tr>
<td>ALT (U/liter)</td>
<td>57.5 (17.5)</td>
<td>62.5 (14.1)</td>
<td>32.5 (4.0)</td>
<td>28.0 (2.7)</td>
<td>22.8 (7.9)</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>64.0 (3.1)</td>
<td>63.8 (3.6)</td>
<td>66.0 (6.3)</td>
<td>70.2 (2.2)</td>
<td>66.5 (2.3)</td>
</tr>
</tbody>
</table>

* P < .05 relative to saline base-line.
Enadoline decreased cocaine self-administration dose-dependently over a dose range of 0.00032 to 0.0032 mg/kg/hr, but there was considerable variability between monkeys in sensitivity to enadoline’s effects (fig. 6). A low dose of enadoline (0.00032 mg/kg/hr) had variable effects on cocaine self-administration in three monkeys and no effect in monkey R801. The dose of enadoline (0.001 mg/kg/hr) that significantly decreased cocaine self-administration in our initial evaluation in Study 2 (fig. 2) also selectively decreased cocaine self-administration in three of four monkeys. Enadoline (0.0032 mg/kg/hr) decreased cocaine self-administration in all monkeys, but these effects were selective only in monkey 89B084. 

Table 2

<table>
<thead>
<tr>
<th>Endpoint Measure</th>
<th>Saline</th>
<th>0.0032</th>
<th>0.01 (week 1)</th>
<th>0.032 (week 1)</th>
<th>0.032 (week 2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Weight (kg)</td>
<td>8.6 (0.9)</td>
<td>8.6 (0.9)</td>
<td>8.4 (0.7)</td>
<td>8.4 (1.0)</td>
<td>7.8 (0.8)</td>
</tr>
<tr>
<td>Hematology</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>40.2 (2.4)</td>
<td>40.8 (1.1)</td>
<td>41.8 (2.9)</td>
<td>39.2 (1.7)</td>
<td>41.2 (2.9)</td>
</tr>
<tr>
<td>WBC count (10^3/µl)</td>
<td>8.8 (0.5)</td>
<td>10.0 (0.7)</td>
<td>9.5 (1.4)</td>
<td>10.7 (1.0)</td>
<td>9.5 (0.5)</td>
</tr>
<tr>
<td>Blood chemistry</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BUN (mg/dl)</td>
<td>14.8 (1.5)</td>
<td>14.8 (1.0)</td>
<td>15.2 (1.6)</td>
<td>14.5 (2.1)</td>
<td>15.8 (2.9)</td>
</tr>
<tr>
<td>Creatinine (mg/dl)</td>
<td>0.85 (0.12)</td>
<td>0.90 (0.11)</td>
<td>0.83 (0.13)</td>
<td>0.80 (0.11)</td>
<td>0.85 (0.12)</td>
</tr>
<tr>
<td>ALT (U/liter)</td>
<td>51.2 (12.3)</td>
<td>51.5 (17.4)</td>
<td>34.8 (10.1)</td>
<td>28.5 (7.8)</td>
<td>28.8 (3.8)</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>66.0 (7.0)</td>
<td>62.2 (5.8)</td>
<td>73.0 (7.7)</td>
<td>69.5 (4.1)</td>
<td>65.5 (9.2)</td>
</tr>
</tbody>
</table>

Fig. 6. The effects of ascending doses of enadoline on cocaine- and food-maintained responding in individual monkeys. Successive doses of enadoline (mg/kg/hr) are shown at the top of the figure. Consecutive days of saline or enadoline treatment are shown on the abscissa. Cocaine injections per day (0.01 mg/kg/injection) are shown as black circles (left ordinates). Food pellets per day are shown as open squares (right ordinates). The dose of enadoline administered to individual monkeys during the last enadoline treatment period is indicated by asterisks after the monkey number.

Fig. 7. The effects of ascending doses of Mr2033 on cocaine- and food-maintained responding in individual monkeys. See legend for figure 6 for details. Catheter failure prevented collection of recovery data during the final saline treatment period for monkey 89B084.
tion of a higher dose of enadoline (0.01 mg/kg/hr) to monkeys 152F and R801 was less effective than a lower dose (0.0032 mg/kg/hr) in decreasing cocaine self-administration. Moreover, during the final week of the 28-day enadoline treatment, all four monkeys abruptly increased cocaine self-administration, but food-maintained responding remained low except in monkey 89B084.

Mr2033 also had variable effects on cocaine-maintained responding across monkeys (fig. 7). The lowest dose of Mr2033 decreased both cocaine- and food-maintained responding in three monkeys. These effects were even greater at 0.01 and 0.032 mg/kg/hr, the Mr2033 doses that significantly decreased cocaine self-administration in Study 2. Monkey 89B084 was relatively insensitive to Mr2033 except at the highest dose studied. During the last week of Mr2033 treatment, each monkey began to increase cocaine self-administration, and monkey 89B084 returned to base-line levels of cocaine self-administration.

On the day after termination of enadoline treatment, frequent yawning was observed in all four monkeys. In addition, occasional decreases in food and/or cocaine self-administration were observed after termination of both enadoline and Mr2033 treatment in some monkeys. No other discernible signs of kappa opioid withdrawal were observed. Cocaine-maintained responding returned to base-line levels in all monkeys during the final saline treatment and/or at the end of the kappa agonist treatment period. During the final saline treatment period, food-maintained responding returned to base-line levels in all monkeys treated with enadoline but remained suppressed in monkeys treated with Mr2033.

Discussion

The purpose of this study was to evaluate the safety and effectiveness of a series of kappa opioid agonists in reducing cocaine self-administration by rhesus monkeys. As discussed earlier in the introduction, there is now considerable evidence that kappa opioid agonists attenuate the behavioral and neurobiological effects of cocaine on several measures. These data from many laboratories support the hypothesis that activation of kappa opioid receptors may functionally antagonize some of the effects of cocaine and thus provide another pharmacological approach to the treatment of cocaine dependence (Hyman and Nestler, 1996). Findings in the present study extend our previous observations that the kappa opioid agonists EKC and U50,488 decreased cocaine self-administration in rhesus monkeys (Negus et al., 1997). In the present study, enadoline, bremazocine and Mr2033 were most effective in reducing cocaine self-administration, whereas cyclazocine, (–) spiradoline and PD117302 had no significant effects on cocaine-maintained responding across the dose range tested. Of the kappa opioid agonists that we have studied under identical conditions, three benzomorphans (EKC, Mr2033 and bremazocine) and two arylacetamides (U50,488 and enadoline) have dose-dependently reduced cocaine self-administration in some monkeys. The other arylacetamides (PD117302 and (–) spiradoline) and the benzomorphan cyclazocine had no appreciable effects on cocaine self-administration and sometimes significantly reduced food self-administration. The decreases in food-maintained responding, sedation and emesis observed during the first few days of treatment with (–) spiradoline, PD117302 and cyclazocine indicate that the lack of effects on cocaine self-administration was not due to inadequate dose levels (figs. 1, 4 and 5). The ineffectiveness of cyclazocine and spiradoline in reducing cocaine self-administration in rhesus monkeys observed in the present study is not consistent with previous reports in rats (Archer et al., 1996; Glick et al., 1995), and this probably reflects both species and procedural differences.

The factors that account for differences in the relative effectiveness of kappa opioids in reducing cocaine self-administration are unclear. The chemical class of these kappa agonists (benzomorphans, arylacetamides) does not necessarily predict their interactions with cocaine. This dissociation is consistent with a previous report that kappa agonists from the arylacetamide family may have different pharmacological profiles in primates. For example, the kappa-selective antagonist nor-BNI surmountably antagonized the antinoceptive effects of two arylacetamides U50,488 and U69,593 in rhesus monkeys, but did not alter the effects of another arylacetamide, enadoline (Butelman et al., 1993a). Differences in selectivity for kappa opioid receptors vs. mu and delta opioid receptors also do not appear to account for the observed differences in reducing cocaine self-administration. For example, enadoline, spiradoline and PD117302 have comparable selectivity for kappa receptors in comparison to mu receptors in rhesus monkey brain (France et al., 1994), but only enadoline (0.001 mg/kg/hr) consistently reduced cocaine self-administration (figs. 3, 4 and 6). The lack of correspondence between binding affinity and selectivity for kappa opioid receptors as assessed by displacement of [3H]U69,593 in membranes isolated from monkey brain cortex (France et al., 1994), and potency in reducing food-maintained behavior and cocaine self-administration observed in the present study is similar to findings in previous studies of the discriminative stimulus and antinoceptive effects of kappa agonists in rhesus monkeys (France et al., 1994).

It is interesting to note that the kappa agonists that were most effective in decreasing cocaine self-administration (EKC, Mr2033, bremazocine, enadoline) produce behavioral effects in primates that are relatively insensitive to antagonism by the kappa antagonist nor-BNI (Butelman et al., 1993a). Thus, it is possible that the relative effectiveness of these kappa agonists in reducing cocaine self-administration may relate to their non-kappa opioid properties. For example, bremazocine and EKC had high binding affinity for mu and delta, as well as kappa receptors, in isolated monkey brain membranes (Emmerson et al., 1994). In pharmacological studies, EKC, Mr2033 and enadoline each appeared to have activity at mu opioid receptors in some assays (Butelman et al., 1993b; Davis et al., 1992). The implications of mu activity for kappa agonist effects on cocaine are unclear. It is generally agreed that mu-selective opioid agonists increase dopamine release, whereas kappa selective opioid agonists decrease dopamine release (DiChiara and Imperato, 1988; Spanagel et al., 1992), and these findings would predict that mu and kappa agonists might have opposite effects on the reinforcing properties of cocaine. However, if EKC, Mr2033, enadoline and bremazocine act as partial mu agonists and have antagonist effects, this combination of mu and kappa activity could result in complementary effects on cocaine self-administration. Archer and co-workers (1996) suggested that the combination of a long-acting mu antagonist and a
short-acting kappa agonist could be especially useful for cocaine abuse treatment because both components reduce dopamine release from the nucleus accumbens.

**Selectivity of kappa opioid agonist effects.** Doses of kappa agonists that decreased cocaine self-administration also usually produced a concomitant decrease in responding maintained by food. Food-maintained responding tended to decrease most during the first 1 to 3 days of treatment when the sedative effects of each kappa agonist were most pronounced, and then returned toward base-line levels and remained relatively stable during the last 5 to 6 days of treatment (figs. 4 and 5). These findings are consistent with our earlier study of EKC and U50,488 in rhesus monkeys (Negus et al., 1997). Also in agreement with our previous study, these findings indicate that kappa agonist-induced decreases in cocaine self-administration are usually not selective. As we have discussed elsewhere, ideally a potential treatment medication should selectively decrease drug self-administration with minimal effects on behavior maintained by nondrug reinforcers such as food (Mello and Negus, 1996). This profile of effects would indicate that decreases in drug self-administration could be attributed to the effects of the treatment on the reinforcing effects of cocaine and not to sedation or a general disruption of operant responding (Mello and Negus, 1996). Although the observed lack of selectivity is not ideal, kappa agonist treatment produced transient and relatively mild undesirable side effects (as described below), and the degree to which these undesirable side effects may limit their clinical usefulness is not known. The overall profile of kappa agonist effects may be acceptable for the treatment of cocaine dependence under some conditions. Because the kappa agonist doses evaluated chronically in Study 2 were estimated from those doses that decreased food-maintained responding after acute administration in Study 1, it is not surprising that food-maintained responding also decreased during chronic kappa agonist treatment (fig. 1). However, the degree and duration of kappa opioid related decreases in food-maintained responding during chronic treatment were often less than would have been anticipated from Study 1. This may reflect antagonism of the rate-decreasing effects of these kappa agonists by cocaine as well as the development of tolerance to kappa agonist effects. Alternatively, the lower potency of some kappa agonists during chronic treatment vs. acute treatment studies could reflect pharmacokinetic factors related to the different methods of kappa agonist administration (3 injections per hour intravenous in the chronic dosing studies vs. a single intramuscular injection in the acute dosing studies).

**Duration of kappa agonist effects on cocaine self-administration.** The effects of enadoline, Mr2033 and bremazocine on cocaine self-administration were usually sustained over 10 days of treatment (fig. 4). However, when ascending doses of enadoline and Mr2033 were administered over 28 consecutive days, cocaine self-administration tended to increase after three weeks of treatment (figs. 6 and 7). These data are consistent with the interpretation that tolerance developed to the effects of these kappa agonists on cocaine self-administration. Tolerance to kappa agonist-related sedative effects was observed in this study and in our previous study of EKC and U50,488 (Negus et al., 1997). Tolerance to kappa opioid agonist effects is also consistent with previous preclinical (Bergman et al., 1985; Gmerek et al., 1987) and clinical findings (Martin et al., 1965, 1966) discussed below. Because any clinical application of kappa opioid agonists for the treatment of cocaine abuse would require chronic administration, the development of tolerance to kappa agonist effects on cocaine self-administration could be a serious limitation.

**Safety and side effects of kappa opioid agonists.** Most of the kappa agonists studied thus far have produced emesis and sedation in some monkeys, but tolerance appears to develop rapidly to these effects, usually within 2 to 3 days. In Study 3, administration of gradually ascending doses of enadoline and Mr2033 decreased the frequency and severity of vomiting and sedation. These findings are consistent with previous reports that tolerance develops to the unconditioned behavioral effects of kappa agonists in rhesus monkeys (Gmerek et al., 1987). Although the undesirable side effects observed in rhesus monkeys disappeared rapidly, the impact of similar side effects on the clinical usefulness of kappa agonists for the treatment of cocaine dependence remains to be determined. Clinical studies indicate that the dysphoric subjective effects associated with cyclazocine administration diminished during chronic treatment, and tolerance developed to cyclazocine’s dysphoric effects when gradually increasing doses were administered for 2 to 4 weeks (Martin et al., 1965, 1966). In clinical trials of enadoline, dose-dependence increases in fatigue, dizziness, emotional lability and abnormal thinking were reported, but these side effects did not cause subjects to withdraw from the study (Reece et al., 1994). It will be important to examine the extent to which tolerance develops to kappa opioid agonist-related undesirable side effects in controlled clinical trials. If tolerance to the adverse subjective effects of kappa opioid agonists develops in humans as rapidly as tolerance developed to kappa-related sedative and emetic effects in the present study, the impact on treatment compliance could be negligible. It is also important to recognize that the relative risk for adverse side effects of a number of drugs under consideration for treatment of cocaine dependence has not been systematically evaluated. For example, dopamine agonists, dopamine antagonists and long-acting dopamine reuptake inhibitors also produce potentially severe effects, including extrapyramidal motor effects. The incidence and severity of undesirable side effects must be weighed against the potentially beneficial effects of any treatment medication.

Importantly, long-term exposure to high doses of enadoline and Mr2033 was not associated with any abnormalities in laboratory indices of hematology and blood chemistry (tables 1 and 2). All monkeys remained in good health throughout 4 weeks of treatment with these kappa agonists. Abrupt cessation of treatment was followed by yawning and transient decreases in operant responding. These behavioral changes might be indicative of kappa opioid withdrawal as previously reported by Gmerek et al. (1987), but the severity of withdrawal in the present study was less than that previously reported, especially after chronic treatment with Mr2033. In the earlier study, monkeys were maintained on Mr2033 (3.0 mg/kg given once every 6 hr) for 239 days (Gmerek et al., 1987), whereas monkeys in the present study received the highest dose of Mr2033 (0.032 mg/kg/hr) for only 14 days. These differences in the dose and duration of Mr2033 treatment probably account for the observed differences in withdrawal severity.
Conclusions. During chronic treatment, some kappa opioid agonists (enadoline, bremazocine and Mr2033) produced sustained and statistically significant decreases in cocaine self-administration in rhesus monkeys, whereas other benzomorphans (cyclazocine) and arylacetamides [PD117302, (- spiradoline)] had no effect on cocaine-maintained responding. At present, the basis for the observed differences in kappa opioid agonist effects is unknown. Kappa agonist-related decreases in cocaine self-administration were usually accompanied by decreases in food-maintained responding. However, endadoline, Mr2033 and bremazocine usually decreased food-maintained responding less than cocaine-maintained responding and decreased cocaine self-administration selectively in some monkeys at some doses. Tolerance developed rapidly to the unwanted kappa opioid side effects (sedation, emesis) usually within 1 to 3 days. These compounds appeared to be safe during chronic treatment, and laboratory indices of hematology and blood chemistry remained normal during 4 weeks of endadoline and Mr2033 treatment. Although selective decreases in cocaine self-administration did not occur during treatment with all kappa agonists or in all monkeys, these findings suggest that some kappa opioid agonists may have a role in the treatment of cocaine abuse and dependence. Theoretically it should be possible to design new kappa agonists that significantly reduce cocaine self-administration, yet have fewer sedative and emetic side effects.

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