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Effects of Repeated Methamphetamine Administration on Methamphetamine Self-Administration in Rhesus Monkeys[1]

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WOOLVERTON, W. L., L. CERVO AND C. E. JOHANSON. *Effects of repeated methamphetamine administration on methamphetamine self-administration in rhesus monkeys*. PHARMACOL BIOCHEM BEHAV 21(5) 737–741, 1984.—The effects of prolonged exposure to high doses of stimulants on stimulant self-administration in rhesus monkeys have not been established. In the present experiment, rates of methamphetamine self-administration as well as the effects of methamphetamine on food-maintained responding were determined before and after a regimen of repeated methamphetamine injections. Increases in self-administration of some doses of methamphetamine as well as tolerance to the rate-decreasing effects of the drug on food-maintained responding were observed following the repeated injection regimen. The results suggest that while tolerance may develop to the rate-decreasing effects of the drug, there may be an increased sensitivity to its reinforcing properties. In addition, since this injection regimen has been shown in previous studies to deplete central monoamines, especially dopamine, the results suggest a role for these monoamines in these behavioral effects of methamphetamine.

Methamphetamine Self-administration Food-maintained responding Tolerance Supersensitivity
Rhesus monkeys Monoamines

THE repeated administration of a psychoactive drug often results in a modification of its behavioral effects. A frequent observation is a change in the sensitivity of the organism to the effects of the drug which may be long-lasting. Among the drugs which have been studied extensively in this regard are the psychomotor stimulants. Both increased sensitivity [1,20] and tolerance [3,20] to the behavioral effects of this class of compounds have been reported to result from their repeated administration. Behavioral as well as pharmacological variables appear to play a role in these seemingly contradictory reports. For instance, tolerance has been reported to develop to the effects of cocaine on milk intake in rats when the drug was given repeatedly before an experimental session while sensitivity to the drug increased when it was administered after the session [20]. However, the effects of exposure to high doses of stimulants on the reinforcing properties of these drugs in rhesus monkeys have not been established.

The present experiment is part of a series of studies designed to evaluate the behavioral, pharmacological and neurochemical effects of prolonged exposure to high doses of methamphetamine (MA). Tolerance has been demonstrated to develop to several of the behavioral effects of MA and has been shown to be correlated with deficits in central monoamine, particularly dopamine, systems [3, 9, 16]. The purpose of the present experiment was to examine the effects of repeated MA injections on subsequent MA self-administration in rhesus monkeys. Since drug self-administration results using rhesus monkeys have proven to be a valid animal model of human drug self-administration [4,6], the results are relevant to the issue of changes in the pattern of drug misuse as a function of prolonged drug exposure.

METHOD

Animals and Apparatus

Two male (7.1–8.0 kg) and three female (4.4–5.8 kg) rhesus monkeys *(Macaca mulatta)* served as experimental subjects. All monkeys were drug-naive at the beginning of the experiment. Each was fitted with a stainless steel harness and spring arm for restraint and catheter protection. The spring arm attached to the rear of the experimental cubicle (68×84×91 cm high) in which the monkey lived for the duration of the experiment, except during the repeated drug regimen (see below). Each cubicle had a Plexiglas window on the front wall that allowed the monkey visual access to the laboratory. The window was covered during experimental

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sessions. Water was continuously available and each monkey received a multiple vitamin supplement in the form of a chewable tablet 3 days/week.

Two response levers (BRS/LVE, PRL-001, Beltsville, MD) were mounted on the inside front of each experimental cubicle 10 cm above the floor and a food dish was mounted between them. Four stimulus lights, two red and two white, were mounted directly above each lever behind a panel of translucent Plexiglas. The ceiling had two 15 watt houselights, one white and one red, covered with translucent Plexiglas. Drug injections were delivered by a peristaltic infusion pump (Cole-Parmer Co., Chicago, IL) at a rate of approximately 1.0 ml/10 sec. Banana flavored food pellets (P. J. Noyes Co., Lancaster, NH; 1 g each) could be delivered to the food dish by an automatic pellet dispenser (Model A, Ralph Gerbrands Co., Arlington, MA). All programming and recording was accomplished by electromechanical equipment located in an adjacent room.

Procedure

Following adaptation to the cubicle and restraint system, each animal was removed from the cubicle and injected with a combination of phencyclidine hydrochloride (1.0 mg/kg, IM) and atropine sulfate (0.04 mg/kg, IM) followed in 20–30 min by sodium pentobarbital (10–20 mg/kg, IV). When anesthesia was adequate, a silicone catheter (0.08 cm i.d., Ronsil Rubber Products, Belle Mead, NJ) was surgically implanted into a major vein. Internal and external jugular and femoral veins could be catheterized. Following surgery the monkey was returned to the experimental cubicle and the catheter was threaded through the spring arm, out the back of the cubicle and connected to the infusion pump. If a catheter became non-functional during the experiment, a new catheter was implanted as before following a 1–2 week period to allow any infection to clear.

The experiment was divided into three phases. The details of each phase are outlined below. As a convenient shorthand the word "chronic" is used to mean the repeated MA injection regimen.

Pre-chronic phase. In the pre-chronic phase, experimental sessions were conducted daily and were 30 min in duration. Initially, each animal was trained in the presence of the white house light and white right lever lights to press the right lever for a 10-sec injection of 0.025 mg/kg/inj MA hydrochloride. During an injection the white lights were extinguished and the red house light and lever lights were illuminated. Responses occurring on the right lever during an injection were counted but had no other programmed consequences. Responding on the left lever had no consequence. Following acquisition of the lever press response, the number of responses required for drug delivery was increased to 30 over a period of several experimental sessions (fixed-ratio 30:FR 30). Occasionally animals failed to acquire the lever press response with MA available. In these cases, training was undertaken with 0.1 mg/kg/inj cocaine and the animals were switched to MA after responding had been established.

Following the establishment of stable rates of responding for 0.025 mg/kg/inj MA (less than 10% variation in total number of inj/session for 3 consecutive sessions), saline and 3 other doses of MA were made available during experimental sessions. Each dose was available until responding stabilized at a new level. Order of dose availability was counterbalanced across animals.

After this dose-response relationship had been determined, the monkeys were food-deprived for 2–3 days and responding during the 30-min experimental sessions was maintained by the presentation of one gram food pellets under a FR 30 schedule. When responding was stable, dose-effect functions were determined for MA (0.125–0.5 mg/kg) given 5 min before the session. The drug was given intravenously via the catheter if it was still intact or, alternatively, via the sapphenous vein. Pre-session MA injections were usually administered on Tuesdays and Fridays. The order of dosing was counterbalanced across animals and each animal was tested twice with each dose.

Chronic phase. The chronic regimen of MA administration was similar to that described by Finnegan *et al.* [2] and Preston *et al.* [11]. The animals were removed from their experimental cubicles and placed in stainless steel squeeze cages (61 × 67 × 71 cm high). If the catheter was still intact, the end was tied off and placed subcutaneously between the scapulae. Prior to beginning the chronic regimen, each animal was allowed a period of at least one month (range 1–3 months) of ad lib food and water to re-establish normal body weight.

When body weight had stabilized, the chronic injection regimen was begun. Four times daily (6:00, 12:00, 18:00 and 24:00) the monkey was injected SC with a dose of MA and behavioral observations were recorded. The initial dose of MA was 4 mg/kg/day (1 mg/kg/inj) and was usually increased by 4–8 mg/kg/day every other day unless the physical condition of the animal dictated a more gradual dose increase. A typical injection regimen for one monkey over 14 days was: days 1 and 2, 4 mg/kg; days 3 and 4, 8 mg/kg; days 5 and 6, 16 mg/kg; days 7 and 8, 24 mg/kg; day 9, 28 mg/kg; days 10 and 11, 32 mg/kg; day 12, 36 mg/kg and days 13 and 14, 40 mg/kg. If intake of regular monkey chow failed to recover during the regimen, animals were fed as much fresh fruit as they would eat. In addition, fluid intake occasionally had to be supplemented with forced fluid from a syringe. At the end of this period, animals were again allowed a minimum recovery period of 1 month (range 1–1.5), during which body weight recovered to pre-drug levels, before continuing with the experiment.

To control for the possibility of changes in sensitivity to the effects of MA due to food deprivation and weight loss, 2 monkeys (1001 and 1002) were subjected to a control regimen. Over a period of 2 weeks they were injected SC with saline and food-deprived to the extent that the decrease in body weight was comparable to that seen in animals during the chronic MA regimen, i.e., to 85–90% of original body weight. Subsequently, the behavioral effects of MA were redetermined in these control monkeys. These animals were then exposed to chronic MA as described above.

Post-chronic phase. Dose-response relationships for MA were redetermined in the post-chronic phase. All aspects of the procedure were identical to those in the pre-chronic phase.

Data Analysis

The number of injections per session over the last three sessions of self-administration of each dose of MA was used in the data analysis. For effects on food-maintained responding, data were calculated as responses/second and averaged for the two test sessions with each pre-session dose of MA.

RESULTS

Dose-Response Relationships

Because of differences in sensitivity to the effects of MA

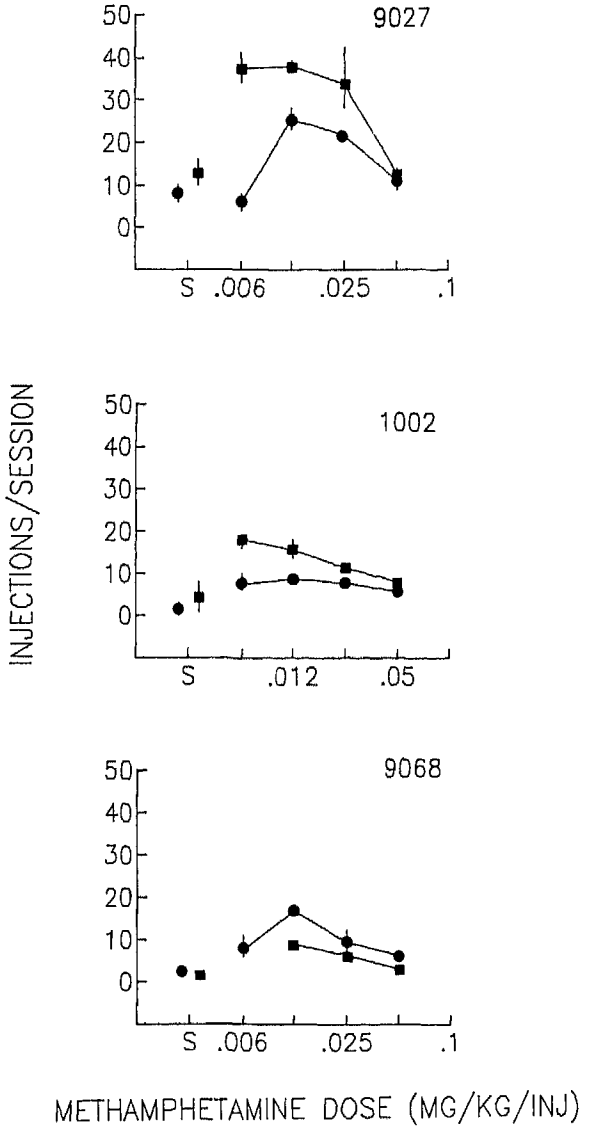

FIG. 1. Self-administration of methamphetamine before (●) and after (■) a regimen of repeated methamphetamine injections. Each point represents the mean number of injections self-administered during the last three sessions of access to the doses noted on the abscissa or saline (S). Vertical lines are the range of these values. For monkey 1002, before (●) values are those collected after the regimen of repeated saline injections. For monkey 9068, after (■) values are those collected after a second injection regimen.

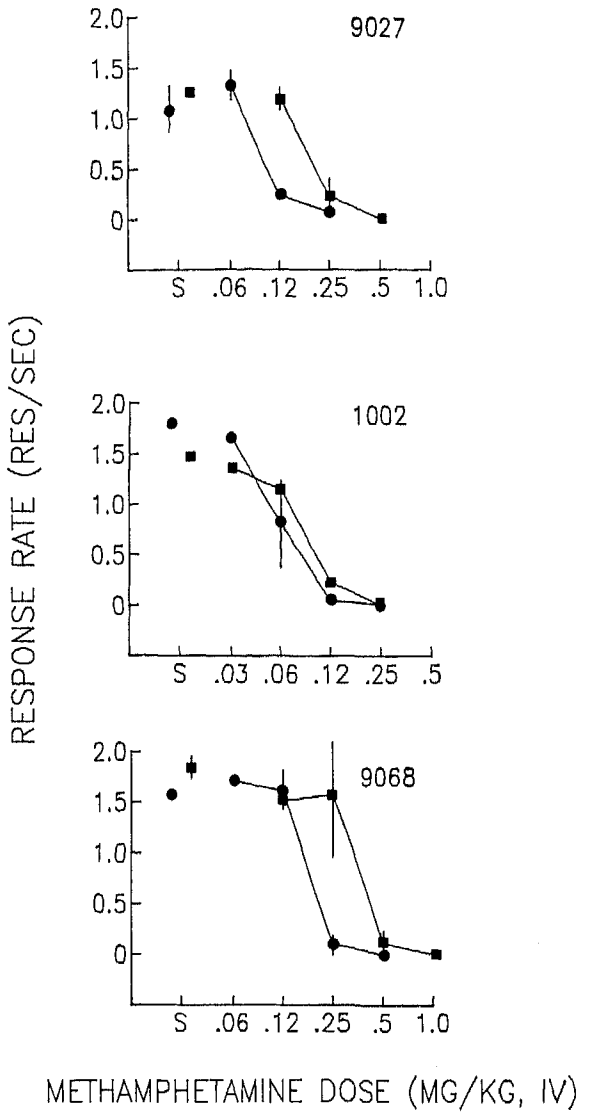

FIG. 2. Effects of methamphetamine on FR 30 food-maintained responding before (●) and after (■) a regimen of repeated methamphetamine injections. Each point represents the mean of 2 determinations of the effect of each dose and vertical lines are the range of these values. Other details are as in Fig. 1.

between animals, data are presented individually. The control regimen of saline injections with food deprivation for monkeys 1001 and 1002 did not systematically alter MA self-administration. Figure 1 shows rates of self-administration of MA before and after the chronic MA regimen. The dose-response relationship for MA was in general an inverted "U" shape in all animals. For monkeys 9027 and 1002, there was an increase in the rate of MA self-administration after the chronic regimen. This change was marked at lower doses and diminished at higher doses. In contrast, MA self-administration was not altered in monkey 9068 (data not shown). Consequently, this animal was exposed to the chronic regimen a second time, with doses in this exposure increasing to 60 mg/kg/day by day 14. Rates of MA self-administration in this animal were, if anything, reduced by this regimen (Fig. 1).

MA administered pre-session reduced the rate of responding for food in a dose-related manner (Fig. 2). Sensitivity to MA was increased by 2–4 fold after the control regimen in monkeys 1001 and 1002. For monkey 1001, the effects of 0.5 mg/kg initially were comparable to the effects of 0.25 mg/kg after the control regimen. Similarly for monkey 1002, the

effects of 0.25 mg/kg initially were comparable to the effects of 0.06 mg/kg after the control regimen (●, Fig. 2). In contrast, this dose-response relationship was shifted to the right (i.e., sensitivity decreased) by about two-fold in monkey 9027 after the chronic MA regimen and in monkey 9068 after its second exposure to chronic MA. There was only a slight decrease in sensitivity for monkey 1002 following chronic MA.

Chronic Methamphetamine

Typical psychomotor stimulant effects were observed in all animals during the chronic regimen. These effects included increased locomotor activity, decreased food and water intake and stereotyped behaviors. In one instance, convulsions were observed on day 7 and were terminated with diazepam (1 mg/kg, IM). It appeared that these effects neither intensified nor diminished during the course of the chronic regimen. Although no monkeys died during the chronic regimen, 2 monkeys died after the regimen was terminated. The direct cause of death was septicemia (8083, 2 weeks after the chronic regimen) in one monkey and apparent liver failure in the other (1001, 6 weeks after the chronic regimen).

DISCUSSION

The repeated administration of MA altered both the self-administration of MA and the effects of MA on food-maintained responding. Tolerance to the effects of the drug on food-maintained responding is consistent with previous reports. A high-dose MA regimen has been found to result in comparable tolerance to the effects of the drug on operant behavior of rats [18] and monkeys [2, 3, 7]. Results from the control animals make it apparent that the chronic MA regimen was responsible for this change in sensitivity, though it is unclear why sensitivity to MA increased in these animals. There was an increase in the self-administration of low and intermediate doses of MA in two monkeys (9027 and 1002). A similar increase in *d*-amphetamine self-administration has been reported in rats following repeated administration of *d*-amphetamine [10]. On the other hand, for the highest dose of MA (0.05 mg/kg/inj) there was little or no change in self-administration. This coincides with a report by Schuster [15] showing no difference in rate of self-administration of this dose when animals that had been treated with a high dose MA regimen were compared to controls.

Although increases in self-administration following exposure to high doses of a drug have been attributed to tolerance to its reinforcing properties [10], interpretation of these results is made complicated by the multiplicity of drug effects that determine rate of responding in this situation. Reinforcing effects as well as direct drug effects on rate of responding simultaneously contribute to the rate of self-administration [5,17]. Changes in either or both of these drug effects as a function of chronic MA exposure could have contributed to the changes in self-administration rates observed. It is possible to make some tentative conclusions, however. At the lowest dose of MA tested (0.006 mg/kg/inj) self-administration was not observed initially. However, in the redetermination of the dose-response function, this dose was self-administered at relatively high rates. Since the rate-altering effects of this dose were minimal or non-existent initially, this finding suggests an increased sensitivity to the reinforcing properties of the drug. At the other extreme, the self-administration of the highest dose of MA (0.05 mg/kg/inj) was not altered by the chronic MA regimen in any of three monkeys. Assuming that the rate-disrupting effects are of maximal importance as a determinant of rate of self-administration at this dose, the data suggest little change in the sensitivity to the rate-decreasing effects of self-administered MA. This result is somewhat difficult to reconcile with the tolerance to the rate-decreasing effects of MA on food-maintained responding. It is possible that at this dose (0.05 mg/kg/inj) the total self-administered dose was so high that this chronic injection regimen was insufficient to produce tolerance. That is, the rate-decreasing effects may be equivalent to those of 0.25 or 0.5 mg/kg given before food-maintained responding. It should be noted that 10 injections of 0.05 mg/kg/inj equals 0.5 mg/kg and that most injections of this dose were self-administered in the first few minutes of the session. Alternatively, it is possible that behavioral variables play a role in this effect. That is, tolerance may develop to the rate-decreasing effects of the drug on responding maintained by one reinforcer (food) and not by the other (drug). In any case, the non-parallel shift in the dose-response relationship for self-administered MA suggests differential changes in sensitivity to the multiple effects of MA that determine rate of responding.

It is unlikely that the phenomenon of behavioral tolerance [15] plays a significant role in the observed changes in sensitivity since animals were removed from the experimental situation during the chronic regimen and drug administration did not interfere with the organism's ability to obtain reinforcement. Sensitivity increases following the control regimen argue that repeated dose-response determinations did not play a role in tolerance development.

It is possible, on the other hand, to suggest a neurochemical mechanism that may be involved in the observed changes in the effects of MA. Chronic MA has been shown to produce long-lasting depletions of central nervous system dopamine in rhesus monkeys [2,16] as well as in other species [19]. Although animals were not sacrificed for neurochemical assays in the present experiment, the same chronic regimen has recently been reported to deplete caudate dopamine in rhesus monkeys to 71% of control levels [11]. The evidence suggests that this depletion results from destruction of dopamine-containing nerve terminals in the CNS [8,12]. If, as has been suggested, the behavioral effects of MA are at least partially the result of its releasing dopamine from CNS dopamine-containing nerve terminals, it is relatively straightforward to suggest that tolerance to these behavioral effects is due to a reduction in the number of nerve terminals available to produce these effects. Increased sensitivity to the reinforcing effects of the drug is more difficult to account for in this context. Perhaps other neuronal systems are involved in self-administration. It is also possible that more substantial destruction of CNS dopamine-containing neurons is necessary to attenuate the reinforcing properties of the drug. Two observations are consistent with this hypothesis. First, in monkey 9068, there was a reduction in the rates of MA self-administration at all doses following chronic drug exposure, perhaps reflecting diminished reinforcing properties. This animal was exposed to the chronic regimen twice, the second time escalating to 60 mg/kg/day which might be expected to produce greater neuronal destruction. Secondly, it has been found by others that cocaine self-administration in rats, also thought to involve CNS dopamine-containing neurons, is attenuated by depletion of dopamine in the nucleus accumbens to less than 50% of control levels [13,14]. Thus, the possibility exists that the change

in drug self-administration is a biphasic effect of central neurotoxicity. That is, moderate neuronal destruction may increase self-administration while severe destruction may eliminate it.

In summary, exposure to a repeated MA regimen resulted in tolerance to the effects of the drug on food-maintained responding and increased self-administration of low and intermediate doses of the drug. Although a complex relationship exists between exposure to the drug and modification of its behavioral effects the data suggest that increased sensitivity to reinforcing properties may be a result of exposure to high drug doses. In any case, it is clear that such exposure can result in increases in drug self-administration. In addition, the results indicate the importance of research designed to investigate the relationship between destruction of CNS neuronal systems and psychomotor stimulant self-administration.

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