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Preexposure to Foot-Shock Sensitizes the Locomotor Response to Subsequent Systemic Morphine and Intra-Nucleus Accumbens Amphetamine

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LEYTON, M. AND J. STEWART. *Preexposure to foot-shock sensitizes the locomotor response to subsequent systemic morphine and intra-nucleus accumbens amphetamine.* PHARMACOL BIOCHEM BEHAV **37**(2) 303–310, 1990.—The effect of repeated exposure to foot-shock on locomotor activity was examined by testing rats in the shock boxes for one hour following shock exposure. Early in testing activity was elevated relative to the nonshocked control group, between 40–60 min following shock. Over days this period of elevated activity occurred sooner in time and lengthened in duration. When these animals were tested in the absence of shock, those preexposed to shock were more active following either saline or morphine (0.5 and 5.0 mg/kg IP) injections. In a second experiment, elevated spontaneous and morphine-induced activity was also found when rats had been preexposed to shock in boxes distinct from the activity test boxes. In a final experiment, animals preexposed to shock were tested after bilateral infusions of either amphetamine (5 and 10 μg/μl/side) or morphine (5 μg/μl/side) into the nucleus accumbens (NAS). On the amphetamine tests, previously shocked animals were significantly more active than control animals. In contrast, intra-NAS infusions of morphine failed to differentiate between the two groups. These results suggest that repeated mild foot-shock sensitizes the mesolimbic dopamine system by mechanisms similar to those mediating the sensitized behavioral and dopaminergic responses seen following repeated opioid or stimulant administration.

Sensitization Foot-shock Locomotor activity Morphine Amphetamine Nucleus accumbens Mesolimbic dopamine system

THE mesolimbic dopamine (DA) system has been implicated in the positive incentive motivational properties of abused drugs, electrical brain stimulation, and naturally occurring rewards such as food and sex: each of these events has been demonstrated to engage this system. The simultaneous activation of this pathway by other means enhances an animal's approach toward, and by implication interest in, each of these events. Conversely, DA receptor antagonists or lesions of this system severely disrupt an animal's approach toward these normally rewarding events.

Systemic administration of either the opioid, morphine, or the stimulant, amphetamine, causes an increase in extracellular DA in both cell body and terminal regions (7, 15, 20, 22). In the case of opioids, this is brought about by increased cell firing (11,31); amphetamine causes direct release and reuptake blockade (25). Behaviorally, these increases in DA activity elicit increased locomotor activity, although at high doses of morphine and amphetamine the hyperactivity may be preceded by hypoactivity or stereotypy respectively (4, 7, 18, 24). Repeated systemic injections or infusions into the cell body region of these agents leads to increases in both terminal DA release and turnover, and potentiated locomotor activity responses (42) referred to as sensitization. Over days the behavioral sensitization to repeated systemic opiate injections is characterized by a successively greater hyperactivity that appears sooner and sooner in time. Similarly, the behavioral response to an amphetamine challenge following repeated administrations is a potentiated locomotor activity or stereotypy response depending on the dose and number of previous administrations.

Acute injections into the nucleus accumbens (NAS) of either amphetamine or opioids also elevate locomotor activity. In contrast to systemic or intra-VTA pretreatments, however, repeated intra-NAS infusions do not precipitate later augmented responses to systemic or further intra-NAS applications (47,48). These findings support suggestions that sensitization results from alterations in the cell body region. Kalivas and his colleagues (8, 20, 21) have provided evidence indicating that in behaviorally sensitized animals there is reduced somatodendritic release of DA to drug challenge. Such an alteration would lead to decreased activation of somatodendritic autoreceptors and, in turn, reduced autoinhibition (42).

There are, as well, changes in the terminal regions that accompany the development of sensitization. The striatal tissue from animals previously sensitized to amphetamine releases elevated levels of DA when later challenged with amphetamine in

vitro (37,49). Further, a recent study using microdialysis in the NAS indicates that in animals previously sensitized to amphetamine DA release is elevated following amphetamine challenge, compared to that in animals injected with amphetamine for the first time (38). These reports suggest that following sensitization greater amounts of DA are released from the terminals.

Cross-sensitization between different agents known to sensitize the mesolimbic DA system has also been demonstrated. Both the behavioral and biochemical responses to either opioids or stimulants are observed to be potentiated in animals previously treated with another of these compounds (9, 30, 41, 46).

Another event known to activate the mesolimbic DA system is the exposure to a stressor. In contrast to mild, acute stressor application, an event generally associated with the selective activation of mesocortical pathways (6, 26, 35, 40, 43), more severe or repeated exposure to a stressor increases DA release and metabolism in mesolimbic terminals (10, 13, 14). Recent microdialysis studies indicate, however, that exposure to even mild acute stress is sufficient to elevate mesolimbic terminal DA release and metabolism (1,16). Repeated exposure to stressors has also been demonstrated to potentiate the subsequent behavioral response to an acute amphetamine application (2, 12, 14, 27, 36). A further indication that the effects of repeated stress and repeated amphetamine are similar is the finding that striatal tissue from repeatedly stressed animals also show increased DA release following amphetamine challenge (49).

The endogenous opioids may be involved in the response to stress in the mesolimbic DA system. Many stressors elicit the release of endogenous opioids (5, 19, 28, 32) and the systemic administration of naloxone blocks stress-induced increases in DA release and metabolism (33). It has been reported that a mild stressor enhances the motor response to a low dose of morphine, whereas a more severe stressor elicits behavioral hypoactivity to the same dose of morphine (39), suggesting that a stress-induced release of endogenous opioids can summate with exogenous morphine to mimic the effects of a higher dose. Furthermore, previously foot-shocked animals display an enhanced behavioral response to intra-VTA injections of the long-lasting, synthetic, delta agonist 2-D-Ala-5-Leu (DALA) (23). This effect of foot-shock can be blocked if intra-VTA injections of naltrexone-methyl-bromide (NMB) are given immediately prior to the shock sessions (23). It would seem that the stress-induced release of endogenous opioids can sensitize the later behavioral response to intra-VTA injections of DALA as can the previous administration of morphine.

In the present set of experiments, locomotor activity was measured in groups of animals following repeated daily exposures to mild, intermittent, inescapable foot-shock and were subsequently tested for locomotor activity following either systemic morphine injections or intra-NAS infusions of either morphine or amphetamine.

EXPERIMENT 1(A,B)

METHOD

Subjects

Forty-six male Wistar rats obtained from Charles River Canada Inc. (St. Constant, Quebec) were individually housed in a controlled environment (temperature of $21 \pm 1°C$; lights on from 8 a.m. to 8 p.m.) with food and water available ad lib. The animals weighed 275–300 g upon arrival to the laboratory; testing began two weeks later. The animals were habituated to the handling and injection procedure during this period. Testing was performed during the light cycle between 12:00 p.m. and 5:00 p.m.

Apparatus

Eight activity boxes equipped with photocell beams were used to record locomotor activity. The boxes ($40 \times 30 \times 25$ cm) were constructed of a transparent Plexiglas front and rear, pressed wood sides, a removable wire screen ceiling, and 22 stainless steel rods for the floor. Two evenly spaced photocell beams cut across the width of the box while a third pair monitored from the sides. The photocells were 4.0 cm off the rod floor. Breaking a beam was recorded as an activity count by a computer. Four of the boxes were wired for inescapable foot-shock produced by a Lafayette shock generator and grid scrambler.

Morphine sulphate was mixed with physiologically isotonic saline (0.9%) and injected in a volume of 1 ml/kg.

Procedure

Preexposure to shock.

Experiment 1A. Two groups of fifteen animals each were tested in Experiment 1A, one shock (SH) and one no-shock (N-SH) control group. Group SH was shocked for 20 min (0.8 mA, 1 sec/10 sec), every other day for five sessions. Immediately following the shock session the activity level of the animals was monitored for a further 60 min. Group N-SH was placed in the activity box for 80 min.

Experiment 1B. In order to replicate the findings of Experiment 1A, and to make additional tests after shock, two additional groups of eight animals were exposed to the same procedure.

Morphine and saline test days. Two weeks following the last shock session animals in Experiment 1B were tested every two or three days in the activity box after IP injections of saline, 0.5, and 5.0 mg/kg IP morphine sulphate, in that order.

RESULTS

Changes in Activity During Shock Preexposure

Experiment 1A. The activity scores for each 20-min interval per day were subjected to a three-way, repeated ANOVA for group $\times$ interval $\times$ day. The analysis yielded a main effect only for interval, $F(3,84) = 44.92$, $p < 0.001$, reflecting the overall tendency for animals to be more active at the beginning of the session than they were at the end. A significant group $\times$ interval $\times$ day interaction, $F(12,336) = 10.05$, $p < 0.001$, was also obtained reflecting a different and changing pattern of activity by the two groups by time interval over days. The mean activity scores ($\pm$SEM) for groups SH and N-SH during the four 20-min intervals of the test sessions on the five days of the experiment are shown in the four panels of Fig. 1a–d. Figure 1a shows that during the first 20-min interval, when shock was being administered, group SH was more active than group N-SH on the first day, but by the third day had become less active. Inspection of panels b–d shows that initially, on Day 1, immediately following shock, group SH was less active than group N-SH, but that over days group SH became more and more active, at earlier intervals following the shock. By the final test day, group SH was significantly more active throughout the entire 60 min following shock. This changing pattern of activity over days is reminiscent of the developing hyperactivity seen in animals repeatedly injected with morphine: over successive days, previously shocked animals displayed a hyperactivity that lengthened in duration and appeared progressively sooner and sooner in time.

A finer analysis done for the first five-min interval on the data from the 20 min immediately following shock, indicated that on

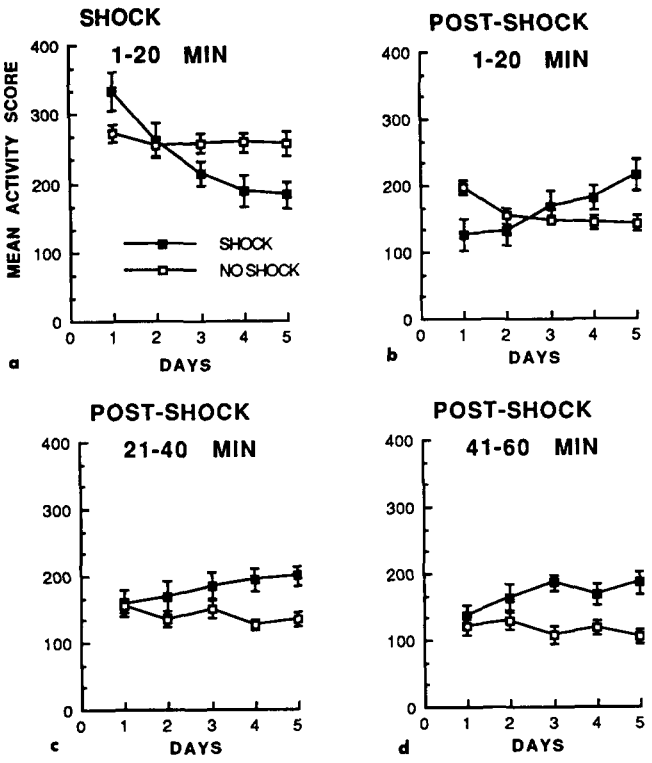

FIG. 1. Successive 20-min group mean activity scores (±S.E.M.) over foot-shock days for animals tested in the shock boxes in Experiment 1A.

Day 1 group SH displayed motor suppression for about five min, compared to group N-SH, $F(1,28)=19.81$, $p<0.001$. This difference between the two groups diminished by Day 5 (see Fig. 2a). Between five and 20 min, however, group SH became more active than group N-SH over successive days as reflected by the group × day interaction, $F(4,112)=7.23$, $p<0.001$ (see Fig. 2b).

Experiment 1B. The second group of animals subjected to the same procedure replicated these findings (data not shown). The three-way repeated ANOVA for group × interval × day done on the activity scores yielded a significant group × interval × day interaction, $F(12,168)=3.81$, $p<0.001$, reflecting group SH's different pattern of activity from group N-SH: as in Experiment 1A

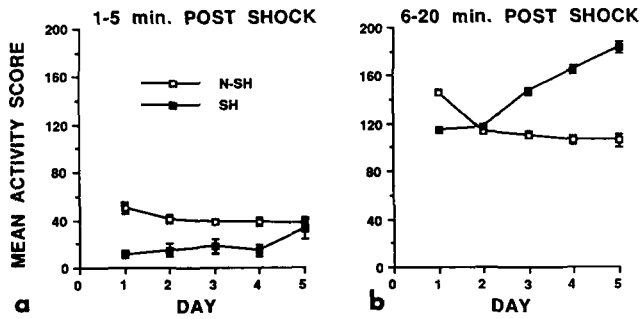

FIG. 2. Group mean activity scores (±S.E.M.) over foot-shock days 1–5 for animals tested in the shock boxes in Experiment 1A. (a) The change over days during the five min immediately following each day's shock session. (b) The period of 6–20 min after each day's shock session.

group SH displayed an early motor suppression followed by a period of hyperactivity that over successive days began sooner in time and lengthened in duration.

Morphine and Saline Tests

The 20-min interval activity scores from the three tests when animals were returned to the activity boxes in the absence of shock were subjected to three separate two-way ANOVAs for group × interval. The results of the test with 5 mg/kg of morphine yielded a significant main effect of group, $F(1,14)=8.60$, $p<0.02$. As illustrated in Fig. 3c, group SH displayed a potentiated activity response to 5 mg/kg of morphine, and this continued throughout the entire 80-min session, relative to group N-SH.

The same analyses for the activity scores from days when the animals were injected with either 0.5 mg/kg of morphine or saline yielded neither main effects nor interactions. A finer analysis of the first 30 min in five-min intervals, however, did reveal significant differences between the two groups. Although the analyses still indicated no main effects, significant group × interval interactions were apparent on both test days [Saline: $F(5,70)=17.33$, $p<0.001$; morphine 0.5 mg: $F(5,70)=2.33$, $p=0.050$], reflecting different patterns of activity by the SH and N-SH groups. Figure 3a shows that during the first five min after the saline injection group SH displayed a profound motor suppression, as compared to group N-SH. By the second five-min interval group SH became more active than group N-SH. Following the injection of 0.5 mg/kg of morphine a pattern of activity similar to the saline day resulted. As can be seen in Fig. 3b during the first five min group SH displayed a slight motor suppression followed by a period of significantly elevated locomotor activity, relative to group N-SH.

DISCUSSION

The results from these two experiments demonstrate that repeated, intermittent, inescapable foot-shock can significantly alter later spontaneous locomotor activity. When activity was measured immediately following foot-shock, animals displayed a short-lived, but profound, motor suppression that was followed by a period of hyperactivity. Over successive shock sessions this hyperactivity appeared earlier in time and lengthened in duration. The change in activity seen over days was very similar to the sensitization of activity seen following repeated opioid administrations. This parallel may well be more than descriptive. Given the previous demonstration that foot-shock evokes release of enkephalins in the VTA (19), as well as DA release, it might be expected that both foot-shock and morphine injections would have similar behavioral activity effects.

The suppression of motor activity seen in group SH, on the saline and low dose morphine tests, was followed by a period of elevated locomotor activity, as compared to that seen in group N-SH. Furthermore, when animals were tested with the higher dose of morphine, previously shocked animals were more active than the N-SH group throughout the entire observation period. These higher levels of activity might have been brought about by a conditioned stress-induced release of endogenous opioids upon reexposure to the shock environment [e.g., (5)]. Alternatively, or in conjunction with the conditioned effect, the previous regimen of repeated shock might have sensitized the mesolimbic DA system to later activation by either morphine or the stress of handling and injection procedures. In sensitized animals, even the mild stress of handling might be sufficient to elicit greater DA release leading to elevated locomotor activity.

EXPERIMENT 2

Experiment 2 was carried out to determine whether the in-

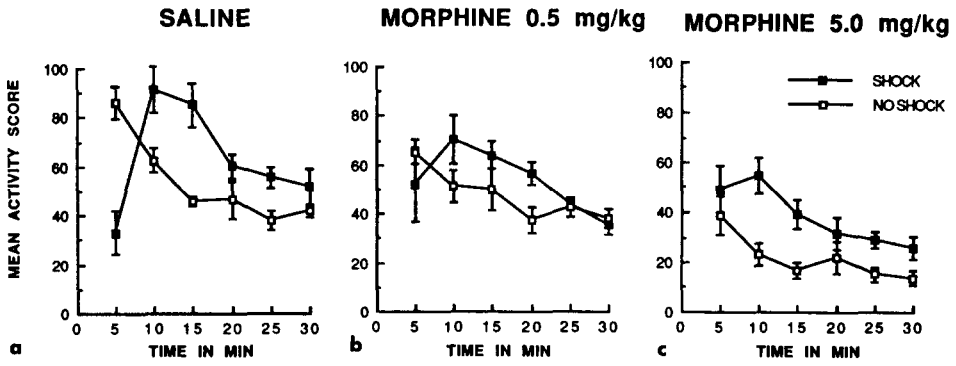

FIG. 3. Group mean five min activity scores (±S.E.M.) for saline- (a) and morphine- [0.5 mg/kg IP (b); 5.0 mg/kg IP (c)] injected animals tested in the shock boxes in the absence of foot-shock in Experiment 1B.

creased locomotor activity observed in Experiment 1, in the environment previously associated with shock, could be observed in an environment other than the shock environment itself. To this end, animals were exposed daily to shock in a manner similar to that of Experiment 1. Tests for changes in locomotor activity were made in a different environment following either morphine or saline injections.

METHOD

Subjects

The subjects were 15 male Wistar rats obtained from Charles River Inc. (St. Constant, Quebec). The animals weighed 275–300 g upon arrival with testing beginning two weeks later.

Apparatus

In addition to the apparatus described for Experiment 1, eight distinctively different Grason-Stadler shock boxes (23.0 × 29.5 × 19.0 cm) were located in a second room. Inescapable foot-shock was provided through 18 stainless steel rods. Each box was sound attenuated and equipped with a miniature light bulb and a fan for ventilation.

Procedure

Activity monitoring and preexposure to shock. Each morning on five consecutive days, all animals were monitored for activity for 45 min in the activity boxes. In the afternoon one group of animals (SH; n = 7) was exposed to intermittent, inescapable foot-shock (0.4 mA, 1 sec/10 sec) for 30 min. No-shock (N-SH) control animals (n = 8) were placed in the shock box for 30 min, but no foot-shock was applied.

Drug tests. On the two days following the last shock session, animals were tested in the boxes for locomotor activity following counterbalanced injections of morphine (0.5 mg/kg, IP) and saline. Two days after these tests, all animals were administered 0.5 mg/kg/day IP morphine for eight days in the activity boxes. Five days later all animals were tested for activity following a saline injection.

RESULTS

The activity scores from the one test prior to, and the four tests following, shock administration were subjected to a two-way, repeated ANOVA for group × day. With the exception of Day 1, prior to the shock regimen (Day 1: SH: mean = 441.2, SEM = 28.1; N-SH: mean = 408.5, SEM = 37.8), group SH was more active than group N-SH, as indicated by the main effect of group, $F(1,14) = 4.75$, $p < 0.05$ (data not shown).

Acute Morphine and Saline Tests

The activity scores from the counterbalanced saline and morphine tests following the repeated shock regimen were subjected to a three-way, repeated ANOVA for group × drug × interval. Group SH was more active than group N-SH on both tests, $F(1,13) = 21.90$, $p < 0.0005$ (see Fig. 4). Although the main effect for drug was not significant, there was a significant effect of interval, $F(2,26) = 65.28$, $p < 0.0001$, and a significant drug × interval interaction, $F(2,26) = 8.90$, $p = 0.001$, indicating a different pattern of activity on the morphine and saline tests. Tukey post hoc tests made on the data at 20, 40 and 60 min (MOR 20: 109.8 ± 11.3; 40: 92.1 ± 18.4; 60: 92.6 ± 17.6; SAL 20:139.3 ± 12.2; 40: 60.1 ± 15.3; 60: 53.4 ± 11.2) yielded a significant effect at 60 min ($p < 0.05$) when morphine-treated animals were more active than saline-treated animals.

Repeated Morphine Tests

The total activity scores (60 min) from the eight subsequent

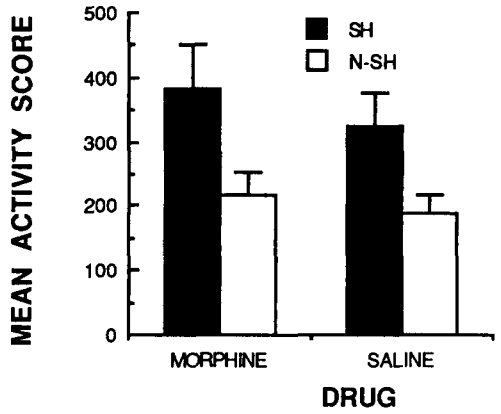

FIG. 4. Group mean 60-min activity scores (+S.E.M.) for the animal's first morphine (0.5 mg/kg IP) and saline injections in Experiment 2.

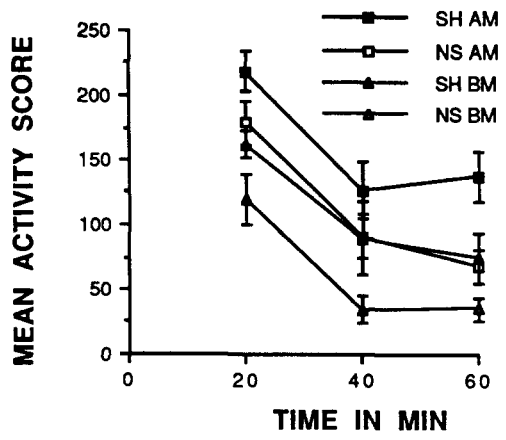

FIG. 5. Group mean activity scores (±S.E.M.) for saline test Day 1 (before the repeated morphine tests), and saline test Day 2 (after the repeated morphine tests) in Experiment 2 (SH: Shock; AM: After Morphine; NS: No Shock; BM: Before Morphine).

morphine tests were subjected to a two-way, repeated ANOVA for group × day. Both the group, $F(1,13)=6.83$, $p<0.02$, and day effects, $F(7,91)=5.04$, $p<0.001$, were significant. Group SH was more active than N-SH, throughout. Both groups also displayed an increase in activity over days (data not shown).

Final Saline Test

Figure 5 compares the results from the final saline test given following the eight days of morphine administered to all animals to those from the saline test given prior to repeated morphine. The activity scores from the two 60-min tests were analyzed in 20-min intervals and subjected to a three-way repeated ANOVA for group × test × interval. The main effects for group, $F(1,13)=20.69$, $p<0.001$, test, $F(1,13)=10.06$, $p<0.008$, and time intervals, $F(2,26)=65.28$, $p<0.001$, were all significant. As can be seen in Fig. 5, group SH was more active than N-SH on both tests, but both group SH and group N-SH were more active on the second saline test, given after the repeated injections of morphine, than they were on the first.

DISCUSSION

The results from Experiment 2 indicate that previous exposure to repeated foot-shock increases locomotor activity measured in an environment different from the shock environment. This finding suggests that the increased activity seen in Experiment 1A and B, in which tests were given in the environment where shock was given, cannot be explained by conditioning to the test environment, and, therefore, is probably not due to a conditioned release of endogenous opioids. The fact that the difference between the SH and N-SH groups was also seen following injections of morphine might suggest that both shock and morphine were sensitizing behavior through similar mechanisms, possibly by changes in the mesolimbic DA system. In the present experiment these tests for cross-sensitization to morphine are confounded by the behavioral depression usually seen following initial systemic injections of morphine [see also (41)]. As is seen in cross-sensitization between amphetamine and morphine (41), the behavioral suppressive actions of morphine appear to mask initially sensitization within the DA system. Interestingly, when tests are made with morphine injected directly into the VTA, evidence for sensitization is clear (23,41). Another way to test whether sensitized behavioral activity to SH and morphine were mediated via similar changes in the DA system would be to challenge with amphetamine, as has been done in the case of cross-sensitization between shock and amphetamine (36). In a final experiment, a direct test of this was made by challenging previously shocked and N-SH animals with microinjections of amphetamine into the terminal region of the mesolimbic DA neurons.

EXPERIMENT 3

In Experiment 3, subjects were exposed to a repeated shock regimen and tested for locomotor activity as in Experiment 2. In an attempt to specify the neurochemical changes underlying the shock-potentiated activity scores, tests were made following infusions of either amphetamine or morphine directly into the terminal region of the mesolimbic DA neurons. It was expected that if the increased locomotor activity seen in animals previously exposed to foot-shock was mediated by changes in the mesolimbic DA system, then intra-NAS amphetamine should cause greater DA release and facilitated activity in group SH animals, as compared to group N-SH. No such difference would be expected following intra-NAS morphine infusions inasmuch as NAS morphine-mediated activity is independent of the DA system.

METHOD

Subjects

The subjects were 24 male Wistar rats obtained from Charles River Inc. (Wilmington, MA). The animals weighed 275–300 g upon arrival with testing beginning two weeks later.

Surgical Procedure

Twenty-four rats weighing 280–320 g were stereotaxically implanted with chronic bilateral guide cannulae (26-gauge stainless steel tubing) aimed at the NAS, positioned 1 mm above the final injection site (A/P +9.1, L ±1.7, D/V 0.0, relative to the interaural line) and angled at 10° with the incisor bar set at 5 mm superior to the interaural line. The animals were given 10 days to recover prior to beginning testing.

Histology

Following the experiment, all animals were perfused transcardially with saline and a 10% formalin solution under deep anesthesia. Brains were stored in a 10% formalin solution for at least 5 days. Histological verification of cannula tip placement was subsequently made on 30-μm thionin-stained coronal sections. On this basis one animal's activity scores from the intra-NAS infusion tests were removed from analysis.

Apparatus

The apparatus were as described in Experiment 2.

Procedure

The shock regimen was as described in Experiment 2. Twenty-four hours after the final shock administration the cannulated animals were returned to the activity boxes for 40 min. These subjects were then monitored for locomotor activity following an intra-NAS saline infusion (0.5 μl/side). Over the following three days animals were tested with counterbalanced injections of morphine (5 μg/0.5 μl/side) and amphetamine (5 μg/0.5 μl/side

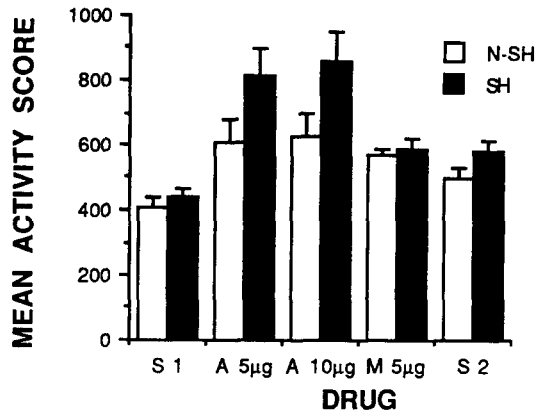

FIG. 6. Group mean 40-min activity scores (±S.E.M.) following intra-NAS drug infusions in Experiment 3 (S: Saline; A: Amphetamine; M: Morphine).

and 10 μg/0.5 μl/side). These tests were followed 24 hours later by a second intra-NAS saline test. Drugs were injected over 45 sec in freely moving animals. The injectors were then left in place for an additional 75 sec.

RESULTS

Activity During Foot-Shock Treatment Days

The activity scores for the two groups on each of the five daily activity sessions given in the mornings prior to the afternoon foot-shock sessions were subjected to a two-way, repeated ANOVA for group × day. There was a significant effect of day, $F(4,88) = 8.30$, $p<0.001$, and a significant group × day interaction, $F(4,88) = 3.59$, $p<0.01$. Analyses of simple main effects indicated that this interaction was attributable to a decrease in activity scores over days in group N-SH, $F(4,88) = 9.58$, $p<0.01$, but no change over days in group SH, $F(4,88) = 0.84$. As in Experiment 2, group SH tended to be more active than group N-SH; in these animals with NAS cannulae, however, the difference was not statistically significant.

Intra-NAS Saline Tests

The activity scores from the two saline test days were subjected to a two-way, repeated ANOVA for group × day. This analysis yielded a significant main effect for group, $F(1,21) = 4.97$, $p<0.04$, reflecting higher activity levels in group SH, and a significant effect for day, $F(1,21) = 19.67$, $p<0.0003$, reflecting the increase in activity from day 1 to day 2. There was no group × day, $F(1,21) = 1.46$, $p = 0.24$, interaction indicating that both groups became more active across days (see Fig. 6).

Intra-NAS Amphetamine Tests

The activity scores from the two amphetamine tests were subjected to a two-way, repeated ANOVA for group × day. The analysis indicated that group SH was more active than group N-SH, $F(1,21) = 5.20$, $p<0.04$, following NAS amphetamine infusions. Neither the main effect for dose, $F(1,21)<1$, nor the group × dose interaction, $F(1,21)<1$, were statistically significant (see Fig. 6).

Intra-NAS Morphine Test

The activity scores from the morphine test were analyzed by *t*-test. Following the intra-NAS infusion of morphine group SH did not significantly differ from group N-SH, $t(21) = 0.89$, $p = 0.38$ (see Fig. 6).

DISCUSSION

This experiment further indicates that the previous application of repeated shock can increase locomotor activity. In Experiment 3, however, the shock-induced increases in spontaneous activity were smaller than those observed in Experiment 2, and were not always statistically significant.

In contrast to this weak effect seen during the monitoring of spontaneous activity, the previous exposure to repeated shock significantly potentiated the locomotor activating effects of intra-NAS amphetamine infusions. Given that NAS amphetamine elevates locomotor activity by increasing synaptic DA levels, this finding suggests that more DA is released in the mesolimbic terminal regions of previously shocked animals.

When these same animals were tested following intra-NAS morphine infusions, the neuronal and transmitter specificity of the effect was further indicated: although both groups were more active than they had been following saline infusions, this morphine test failed to differentiate between groups SH and N-SH.

Following the three counterbalanced drug tests all animals were tested with NAS saline for a second time. Both group SH and N-SH were more active on this day than on the saline day prior to the drug tests. This increase in activity may be an effect of conditioning: on the previous three days, the animals may have come to associate the boxes with the activity inducing effects of NAS morphine and amphetamine. The substrate mediating such conditioning is unclear, however. Conditioning has been reported to NAS infusions of amphetamine in the dose range used in the present study (3), however, conditioning was not apparent in a study carried out in this laboratory that used a lower dose (48). As has been suggested elsewhere, it may be necessary for the DA neuron to fire before the accompanying sensory stimuli can exert conditioned control over behavior (42).

These behavioral data indicate that previous repeated mild, intermittent, inescapable footshock sensitizes the response of the mesolimbic DA system to the effects of a DA releasing drug, but does not increase NAS opioid responsivity to later activation.

GENERAL DISCUSSION

These experiments demonstrate that the repeated exposure to mild, intermittent, inescapable foot-shock increases spontaneous locomotor activity, and potentiates the activity response to both systemic morphine and intra-NAS amphetamine. Previous research has indicated that all three of these treatments engage the mesolimbic DA system: morphine appears to increase DA cell body firing; repeated stress increases DA release and metabolism, possibly by increasing DA cell firing via endogenous opioid actions in the DA cell body region, and/or by reduced frontal cortical inhibitory feedback (17). Intra-NAS amphetamine functions as an indirect DA agonist by increasing DA release and decreasing its reuptake. This action of amphetamine in the NAS has also been proposed to mediate the increased locomotor activity following systemic administration. Behavioral cross-sensitization has been demonstrated between various combinations of these events: amphetamine and morphine, stress and amphetamine, and stress and intra-VTA opiates. We now report behavioral cross-sensitization between the effects of prior exposure to repeat foot-shock and both systemic morphine and intra-NAS amphetamine.

These findings support suggestions that repeated exposure to excessive extracellular DA, accomplished in this study by repeated

exposure to foot-shock, changes the system such that greater amounts of DA are released from the mesolimbic terminal regions upon later stimulation. That such an alteration might underlie a shock-induced elevation of locomotor activity finds support in Experiment 3 in which intra-NAS infusions of amphetamine led to significantly greater increases in locomotor activity in animals that had been repeatedly exposed to shock. Also consistent with this interpretation are Experiments 1 and 2 in which animals were tested following systemic injections of morphine. If repeated shock increases the mesolimbic DA release to subsequent challenges, then systemic morphine would also be expected to elicit greater behavioral activity in these animals through its effect on DA neuron firing. These results were obtained. Systemic morphine injections elicited significantly greater activity responses in previously shocked animals, and this effect was maintained over nine injections of a low dose.

In contrast to these findings, repeated exposure to shock did not potentiate the locomotor activity observed following subsequent intra-NAS applications of morphine suggesting that changes in opioid receptors in this region do not mediate sensitized responding. Morphine given into the NAS is thought to produce increased activity by acting on opioid receptors on cells intrinsic to that region (29, 44, 45), and to be effective independent of the DA neurons (24,34). It would seem, therefore, that the expression of sensitized locomotor activity following systemic morphine is dependent on DA activity.

In summary, repeated exposure to mild foot-shock potentiates the locomotor response to subsequent systemic injections of morphine and to intra-NAS injections of amphetamine, but not to intra-NAS morphine. These findings suggest that shock sensitizes the mesolimbic DA system in a manner similar to that seen following repeated exposure to opioid and stimulant drugs.

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