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BRIEF COMMUNICATION

Environmental Influences Upon Morphine or d-Amphetamine Induced Suppression of Operant Behavior[1]

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SPARBER, S. B., H. A. TILSON AND D. W. PETERSON. *Environmental influences upon morphine or d-amphetamine induced suppression of operant behavior.* PHARMAC. BIOCHEM. BEHAV. 1(1) 133–136, 1973.–Rats trained in an operant chamber to lever press for food on a FR-30 schedule of reinforcement were given 1.0 mg d-amphetamine sulfate (IP)/kg. Injection of the drug followed by immediate placement into the operant chamber resulted in a disruption of behavior, after a 10 min latency, which lasted for approximately 25 min. Removal of the animals from the chamber soon after the onset of behavioral disruption and replacement into the chamber as soon as 2.5 min later resulted in resumption of lever pressing almost immediately. Animals left in their home cages for 10 min after being injected with d-amphetamine and then placed in the operant chamber started to bar-press almost immediately, taking an average of 4 min to receive 5 reinforcers (150 responses). A second group of rats, maintained on the same schedule of reinforcement, showed similar striking effects of delayed placement after injection with 10 mg morphine sulfate/kg. The 10 min delay resulted in a significant reversal of the behavioral suppression observed at a comparable time after injection of morphine and immediate placement into the operant chamber.

Morphine d-Amphetamine Operant behavior Environmental manipulation

IN A recent report [5], it was shown that the behavioral disruptive effects of mescaline and lysergic acid diethylamide-25 (LSD) on food reinforced fixed ratio (FR) responding may be attenuated by a procedure as simple as removing the rat from the operant chamber for a few minutes or manipulating the time between the injection and initial placement of the animal into the operant chamber. The purpose of this investigation was to extend the previous observation to two other classes of compounds. d-Amphetamine was chosen for study because it produces autonomic [3] and behavioral effects (disruption of FR behavior; [1,6]) which are similar to LSD and mescaline. Morphine was chosen to determine if a similar effect is observed with a drug of another class which has a different behavioral effect, as indicated by the shape of the cumulative record being generated by the same schedule of reinforcement.

METHOD

Animals and Apparatus

For amphetamine experiments, four male albino rats (Sprague-Dawley) used in a previous experiment [5], and for the morphine study six naive male albino rats (Sprague-Dawley) were experimental animals. All were housed individually in air-conditioned quarters. The animals (400–425 g) were food deprived to 70–80% of their free feeding weight and maintained on a 23 hr schedule of food deprivation. The animals had been trained to press a lever for food pellets (Noyes, 45 mg) on a fixed ratio (FR) 30 schedule of reinforcement and were responding at stable rates upon initiation of these studies. Experiments were performed in an operant chamber (Foringer, Rockville, Md.) contained within a sound and light attenuated outer cubicle [5]. Delivery of food pellets and recording of lever

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presses were performed automatically by electromechanical programming equipment.

Procedure

Following three days of baseline FR 30 responding during 42 min sessions, the four animals used in the amphetamine study were injected IP with 0.5 ml of 0.9% saline (NaCl) immediately before placement into the operant chamber (NaCl control). The next day, 1 mg of d-amphetamine sulfate/kg (d-amph, K and K Labs, Plainview, N.Y.) dissolved in NaCl, was injected in a similar manner for use as a drug control. This dose of d-amph has been shown to produce a disruption of FR behavior for approximately 15–30 min [1,6]. Another NaCl control session was run 24 hr later, followed by an experimental session the next day. In these experimental sessions, each of the four rats was injected with 1.0 mg d-amph/kg and immediately placed into the operant chamber. Following the onset of behavioral disruption (i.e. 2 min without reinforcement), the animals were removed from the operant chamber and placed in their home cages for 5.0 min. They were subsequently replaced in the operant chamber and allowed to resume lever pressing for food. Additional NaCl and drug control sessions (i.e. the rats were allowed to remain in the operant chamber) followed on successive days. Seventy-two hr later, the entire sequence was repeated, except that the duration in the home cage during the experimental session was shortened to 2.5 min.

We have previously shown that the time between injection and placement into the operant chamber is an important determinant in the effect of LSD or mescaline on FR responding [5]. Another experiment was performed to test for the effects of delayed placement on the behavioral effects of 1.0 mg d-amph/kg. The duration of this delay was equal to the mean of the latencies to observe 2 min without reinforcement (i.e. latency to onset of behavioral disruption) established in five previous drug control sessions (about 10 min). Again, each animal was given one drug control session (immediate placement into operant chamber after injection) before and after one experimental session (delayed placement). All drug sessions were separated by at least 72 hr.

A similar procedure was used to test for effects of delayed placement upon the behavioral response to 10 mg of morphine sulfate (MS)/kg. On the day following a NaCl control session all the rats in the second group were injected with MS. Half were placed immediately into the operant chamber while the other half were given a 10 min delay (in their home cages) after the injection. One week later the procedure was repeated, reversing the two subgroups. All animals were removed from the chamber 60 min after the injection.

The crossover design was incorporated in the morphine study to control for possible rapid (1–2 injection) tolerance [4], which if present, would show up as an erroneous attenuation attributable to the home cage manipulation (delay).

RESULTS

The average onset to behavioral disruption by d-amph (time between injection and no reinforcers for 2 min) in sessions involving injection and immediate placement was 10.3±0.5 min (M±SE). There was little intrasubject variability, indicating the absence of tolerance development. In addition, there was little variability between subjects, and the group mean (10 min) served as the time in the home cage in the delayed placement experiment. When the animals were placed immediately in the operant chamber (drug control sessions), the mean duration of drug action (time to obtain 5 reinforcers starting immediately after behavioral disruption) was 24.5 min. Removal of the animals from the operant chamber 2 min after the onset of behavioral disruption (i.e. no reinforcers for 2 min) and replacing them 5.0 or 2.5 min later had profound effects on the disruptive action of d-amph. In many cases the animals resumed responding almost immediately after replacement into the operant chamber, and within 10–12 min after disruption all animals had received 5 reinforcers, shortening the duration of action by at least half. Behavioral disruption was further reduced to a mean of 4 min (approximately 80% reduction) when initial placement into the chamber after injection was delayed by 10 min (Table 1).

When MS (10 mg/kg, IP) was injected and the animals placed immediately into the operant chamber there was an initial period (6–18 min) of little or no responding on the lever (Fig. 1). However, once the rats started to respond they did so at a depressed rate, as indicated by the decreased slopes of the cumulative records. Since there was no abrupt cessation (with no responding) followed by a return at about baseline rates, we analyzed the drug effect as a decrease in the number of reinforcers obtained during the last 50 min of a 60 min session. The result of placing the animals in their home cages for 10 min prior to placement into the operant chamber (a time approximately equal to the delay seen prior to response initiation after morphine and immediate placement) likewise attenuated the behavioral effect of the opiate. Where immediate placement into the chamber resulted in the rats receiving an average of 20% (21 ± 7, M± SE) of the reinforcers, when compared to the last 50 min of NaCl control sessions, the home cage manipulation significantly ($p<0.01$, paired t-test) reversed the attenuation. Animals left in their home cage for 10 min performed at a rate which brought the number of reinforcers received back up to 55% (55 ± 26, M ± SE) of control.

DISCUSSION

Previously, we have reported on the reversability of the operant behavioral effects of various doses of LSD and mescaline, using the methodology described herein [5]. The data of the present investigation confirm and expand upon the phenomenon in that the behavioral effects of a nonhallucinogenic stimulant, as well as an opiate depressant, can be greatly attenuated by this simple manipulation.

The observations of slight or complete reversal of the behavioral actions of psychoactive compounds by removal from the environment for as little as 2.5 min may have some practical significance for behavior-biochemical interaction studies. It is possible that biochemical modification could likewise ensue and the use of the same animal for both behavior and biochemcial determinations might lead to erroneous conclusions as to what is happening biochemically, at the time of disrupted behavior. The alternative of using paired animals, one for behavior and the other for biochemical experiments, has even greater obvious drawbacks. Perhaps the use of instantaneous, *in situ* sacrifice or perfusion of brain during the behavioral task or drug effect [8] can circumvent this problem.

Again, our interpretation of what may be responsible for the attenuating action of delayed placement relies upon a

TABLE 1

EFFECT OF HOME CAGE MANIPULATION UPON THE DURATION OF d-AMPHETAMINE INDUCED SUPPRESSION OF OPERANT BEHAVIOR

	Time to obtain 5 reinforcers (min)*	
Rat No.	Drug Control	Delay
A 17	23.0	4.0
	20.5	
A 18	25.5	4.0
	22.5	
A 19	27.0	5.0
	32.0	
A 30	22.0	3.0
	23.0	
Mean ± S.E.	24.5 ± 1.2	4.0 ± 0.4

*Drug controls consisted of injecting rats and placing them immediately into the operant chamber. Delayed sessions consisted of injection and placement in the home cage for 10 min prior to placing them in the operant chamber. Results are expressed as the time to obtain 5 reinforcers beginning 10 min after the injection, since the onset of behavioral disruption (10.3 min) was the same as the time in the home cage (10 min).

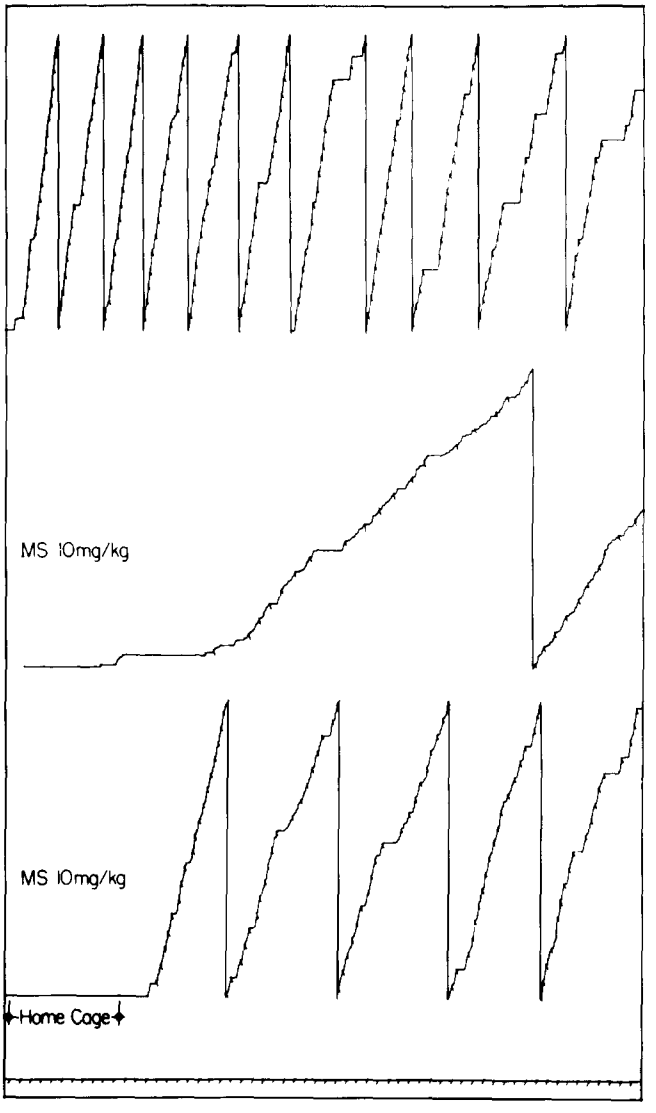

FIG. 1. Representative cumulative records of FR 30 responding for rat DA-1 showing the behavioral effects of morphine sulfate (10 mg/kg, IP). The upper record shows performance after a NaCl injection and immediate placement into the operant chamber. The middle record was generated after morphine and immediate placement into the operant chamber. The lower record shows the reversal (attenuation) of drug effects by prior placement into its home cage (10 min delay) before being placed into the operant chamber. The event marks beneath the cumulative records are a 1 min time base.

difference between stimuli controlling ongoing behavior as opposed to stimuli initiating the same behavior. For example, following the drug injection and placement into the operant chamber, the resulting drug elicited stimuli compete with those stimuli controlling the ongoing response. If the drug induced stimuli are intense enough, there is a loss of stimulus control and behavior is disrupted. The greater the stimulus change (e.g., greater the dose), the more rapid the onset of action and longer the duration of behavioral disruption [5]. On the other hand, when the animals are injected, placed in the home cage and then placed in the operant chamber, the same drug induced internal changes (stimuli) compete with discriminative stimuli associated with response initiation. The stimulus complex related to handling, placement into the chamber etc., appears to be of great enough magnitude to overcome the disruptive effects of the drug. In addition, stimulus control factors are presumably operating in the experiment involving removal of the animals for 5.0 or 2.5 min following the onset of behavioral disruption. Control experiments for novel stimulus changes (blinking the house lights accompanied by a tone at the proper time) did not have any effect on the duration of the disruptive effect by 1.0 mg d-amph/kg, further indicating the involvement of discriminative stimuli associated with handling, home cage and placement into the chamber.

In other studies [6,7] it was also shown that tolerance to the behavioral effects of mescaline (10 mg/kg) and d-amph (1.6 mg/kg) administered IP, was not conveyed to a behaviorally equieffective dose of the same drug infused into the lateral ventricles. Since the behavioral effects resulting from the IP administration of the same or similar doses of mescaline and d-amph are easily manipulated by these environmental contingencies, there is a suggestion that their initial disruptive effects on FR responding are at least partially due to peripheral effects common to both drugs.

What we are therefore proposing is that low doses or the initial behavioral action of higher doses of psychoactive compounds may be producing their effects through peripheral action, with its ensuing stimulus consequences.

The morphine data relating to the initial period of nonresponding immediately after IP injection may also be related. Morphine has been shown to release vasoactive substances and/or in other ways produce cardiovascular effects and resultant peripheral autonomic reflex activity [2]. Either or both of these effects might then act as novel or disruptive stimuli resulting in the initial behavioral action, to be followed later by its CNS depressant effects. The demonstration that a third class of compounds (opiates), which produces a differential effect (compared to psychotomimetics and psychomotor stimulants) upon behavior maintained by FR schedules, appears to be sensitive to the home cage manipulation is further evidence for the generality of the phenomenon. Further investigations are under way to determine the effects of these simple manipulations upon behavior maintained by other schedules of reinforcement.

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