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Reward System Depression Following Chronic Amphetamine: Antagonism by Haloperidol[1]

ROBERT J. BARRETT[2] AND DAVID K. WHITE

Tennessee Neuropsychiatric Institute, Veterans Administration Medical Center
Department of Psychology, Vanderbilt University, Nashville, TN

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BARRETT, R. J. AND D. K. WHITE. *Reward system depression following chronic amphetamine: Antagonism by haloperidol.* PHARMAC. BIOCHEM. BEHAV. 13(4) 555–559, 1980.—The effect of pre-treatment with haloperidol, a dopamine antagonist, on chronic amphetamine's suppression of intra-cranial self-stimulation of the lateral hypothalamus was investigated. Rats treated with 15 mg/kg d-amphetamine per day for seven days displayed a marked increase in reward threshold for electrical brain stimulation responding with an accompanying suppression of response rate. This disruption of intracranial self-stimulation responding was not observed when 30 min prior to each amphetamine injection, animals were injected with 1.0 mg/kg of haloperidol. This study demonstrates that post-synaptic mechanisms play an integral role in the development of chronic amphetamine depression, and suggests that changes at the dopamine receptor are involved in this reward system alteration.

Amphetamine Animal models Drug-induced depression Haloperidol Self-stimulation

IT HAS been well documented that in humans while acute amphetamine elevates mood [19], tolerance to this effect develops rapidly with continued use [10]. Furthermore, following prolonged use of high doses of amphetamine, depression invariably occurs which is not easily differentiated from other types of clinical depressions [7, 24, 31]. Barrett and Leith [2] and Leith and Barrett [20,21] have used these findings as the basis for developing an animal model of human depression. In these experiments, chronic amphetamine's effects on responding for rewarding stimulation of the brain were investigated.

In rats, acute doses of d-amphetamine (AMPH) potentiate reward as evidenced by enhanced responding for intracranial self-stimulation (ICSS) [20, 27, 28]. However, tolerance to this effect has recently been demonstrated subsequent to chronic AMPH injections [20]. Furthermore, following chronic AMPH there occurred a suppression of ICSS response rate and a shift in reward threshold such that higher current intensities were required to maintain ICSS responding equal to that seen prior to treatment [20,21]. Thus, the effects of acute and chronic AMPH on human affect and on ICSS responding in rats may be mediated by similar physiological processes.

There is considerable evidence that the catecholamine's (CA) are involved in mediating ICSS behavior [14] and that AMPH facilitation of self-stimulation responding is due to the drug's enhancing catecholamine function [9,32]. Acutely, AMPH has been shown to release norepinephrine (NE) and dopamine (DA) from pre-synaptic stores, block re-uptake, and inhibit monoamine oxidase [5, 15, 27]. All of these actions would be expected to increase the availability of NE and DA at receptor sites. Likewise, the disruption of ICSS responding observed following chronic AMPH suggests a reduction in neural transmission at these same sites. Similar results have been reported following other treatments known to interfere with catecholamine function [3, 23, 32]. Biochemical studies following chronic AMPH have presented evidence for reduced NE and DA stores [25] but the effects on post-synaptic processes are less clear [1,16].

The purpose of the present study was to determine whether blocking the DA receptor with haloperidol [8] prior to AMPH injections would alter the development of post-amphetamine depression of ICSS responding in the lateral hypothalamus.

METHOD

Subjects

The subjects of this experiment were 12 naive male F344 rats (Harlan Industries, Indianapolis, IN). Their weight at the time of surgery was 275–300 g. They were individually housed under standard laboratory conditions with free access to food and water. All animals were maintained on a 12 hour light-dark cycle (7:00 a.m.–7:00 p.m. light). Surgery and testing were performed during the light phase.

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[2]Requests for reprints should be mailed to R. J. Barrett, Psychology Research Laboratory, Veterans Administration Hospital, 1310 24th Avenue South, Nashville, TN 37203.

Apparatus

Testing occurred in three grid-floored Lehigh Valley operant chambers (26.67 cm high×30.48 cm wide×24.13 cm deep) each equipped with a single lever (2.22×2.86×0.095 cm) located 4.92 cm above the floor and requiring a 24 G force through an excursion of 0.16 cm to activate. The chambers were housed in sound attenuated cubicles and white noise was used to mask extraneous auditory stimuli. Behavioral contingencies were programmed with electromechanical equipment housed in an adjacent room.

Surgery

Subjects were anesthetized with 50 mg/kg of sodium pentobarbitol and were administered 0.6 mg/kg of atropine to inhibit respiratory duress. They were then mounted in a stereotaxic instrument and implanted with a single bi-polar electrode aimed at the lateral hypothalamus. Electrode placement was derived from the sterotaxic atlas of König and Klippel [18]. The coordinates used were 3.8 mm anterior to the interaural line, 1.2 mm lateral to the sagittal suture, and 8.5 mm down from the top of the skull. The electrode consisted of two stainless steel wires, each 0.254 mm in diameter twisted together and insulated except for the tip (purchased from Plastic Products Co., Roanoke, VA). Following surgery, the animals were injected with 150,000 units of bicillin, half in each hind leg muscle for protection against infection.

Procedure

One week following surgery, the animals were trained during daily 30 min sessions to bar-press for a 0.2 sec pulse of 60 Hz AC. Following bar-press acquisition, the animals were given additional training on a current step-down procedure [20] which involved decreasing the current intensity every five seconds by one-twentieth of the original starting intensity. After 15 steps the current was automatically reset to the original intensity at which time a cue light came on signalling this to the animal. Following reset, current step-down did not begin again until the animal made at least one response on the lever. This cycle of 15 current steps was repeated throughout the session and the cumulative number of responses made at each of the 15 different current intensities was recorded.

For graphing purposes, the number of responses made at each intensity was converted to a rate measure (bar-presses/minute) so that a response rate by current intensity profile could be obtained. Current intensities were adjusted for each animal to produce a similar rate×intensity profile which took the form of a descending ogive. This type of rate×intensity function allowed for a sensitive baseline from which treatment related shifts in ICSS responding reflecting either enhanced or disrupted reward properties, could be detected.

In addition to response rate, a measure of reward threshold, independent of rate, was computed for each subject by recording on each trial (cycle of 15 descending current steps) the value of the first current intensity at which the animal failed to respond. For a given session, the average of these values was determined for each animal and used as an estimate of the threshold rewarding current intensity.

When responding stabilized so that there was little day to day variation on both the response rate×current intensity function and the reward threshold measure, the animals were assigned to one of four groups (Saline-Saline, Saline-AMPH, Haloperidol-Saline, Haloperidol-AMPH) matched on the aforementioned measures. All animals were then started on an injection regimen consisting of two morning injections spaced 30 min apart (8:00 a.m.; 8:30 .m.) and two afternoon injections spaced 30 min apart (3:30 p.m.; 4:00 p.m.) for a total of four injections per day for seven consecutive days. All injections were administered intraperitoneally.

The Saline-Saline animals received four injections per day of 0.9% saline solution. Animals from the Saline-AMPH group were injected in the morning with saline followed 30 min later by 5 mg/kg of AMPH. These animals received a second saline injection at 3:30 p.m. with a subsequent AMPH injection (10.0 mg/kg at 4:00 p.m.). The Haloperidol-Saline animals were injected with haloperidol (1.0 mg/kg) at 8:00 a.m. followed by a saline injection 30 min later. This same sequence of injections was repeated in the afternoon. Animals from the Haloperidol-AMPH group were injected at 8:00 a.m. with 1.0 mg/kg of haloperidol followed 30 min later by 5.0 mg/kg of AMPH. These animals received another 1.0 mg/kg of haloperidol at 3:30 p.m. followed by 10.0 mg/kg of AMPH at 4:00 p.m.

All of the saline injections were 1.0 ml/kg of 0.9% saline solution. Haloperidol was diluted in distilled water and AMPH in saline solution so that the desired dose per kg was contained in a volume of 1.0 ml. Following the seventh day of injections, the drug regimens were discontinued and daily testing on the step-down procedure resumed 40 hr subsequent to the final injection.

Histology

Following the completion of the drug regimens, all animals were given an overdose of sodium pentobarbitol and perfused intracardially with 60 ml of 0.9% saline followed by 100 ml of 10.0% Formalin. The brains were removed and stored in Formalin for several days prior to being blocked in the plane of the electrode tract. Brain sections were then frozen with CO_2 and 60 μ sections were made using a freezing microtome. Photomicrographs made directly from these slices were used to locate the electrode placements. All electrodes were located within the region of the lateral hypothalamus.

RESULTS

The data graphed in Figs. 1 and 2 represent the means from the last two test sessions prior to chronic injections (pre-treatment) and the first two test sessions following the termination of injections (post-treatment). As can be observed in Fig. 1, chronic amphetamine (Saline-AMPH) produced a marked increase in the current intensity required to support responding on 50 percent of its presentations (threshold). However, pre-treatment with haloperidol (Haloperidol-AMPH) blocked the development of this increase in the reward threshold. A 4 (treatment)×2 (pre-post)×2 (days) ANOVA supports these conclusions in that a significant treatment×pre-post interaction occurred, $F(3,19)=5.93$, $p<0.005$, with the increase in reward threshold of Saline-AMPH animals being significantly greater than that observed in AMPH animals pre-treated with haloperidol (Haloperidol-AMPH) or with haloperidol (Haloperidol-Saline) alone (Simple Main Effect, $p<0.05$).

Figure 2 demonstrates response rate plotted as a function of current intensity. Prior to drug treatment, a characteristic and similar profile of responding was observed among all four groups. A 4 (treatment)×2 (pre-post)×2 (days)×15 (cur-

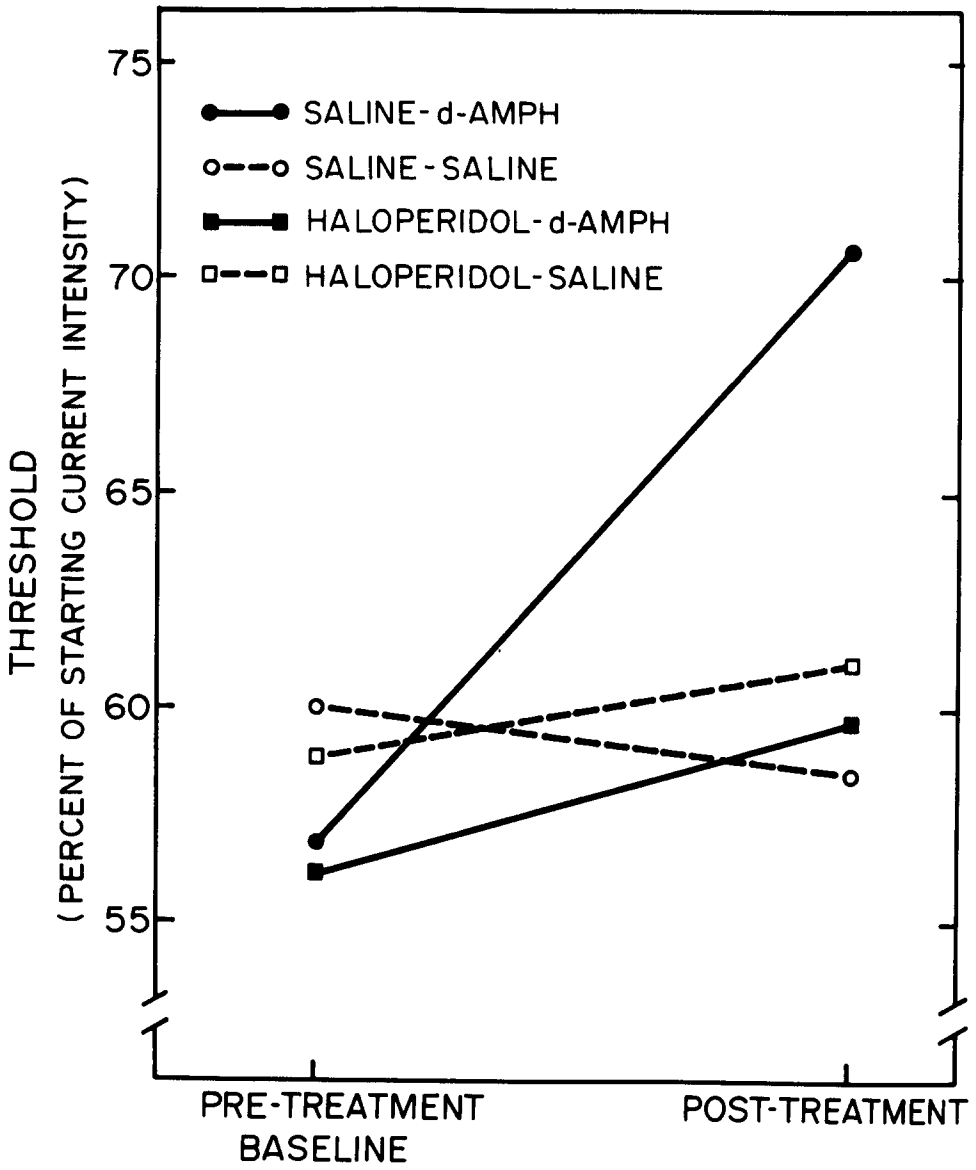

FIG. 1. Increase in reward threshold subsequent to chronic injections of d-amphetamine and antagonism of this effect by pre-treatment with haloperidol. Threshold current intensity (expressed as a percent of initial current intensity) represents the average of the first intensity on each cycle in the step-down procedure which failed to sustain ICSS responding.

rent intensity intervals) ANOVA indicated that response rates declined following the injection regimens, $F(1,9)=8.91$, $p<0.008$. Although the treatment×pre-post interaction failed to reach an acceptable level of significance, $F(3,19)=2.43$, $p<0.10$, it can never-the-less be seen that animals treated with amphetamine alone (Saline-AMPH) displayed the most pronounced suppression of response rate when compared with animals pretreated with haloperidol (Haloperidol-AMPH) or to animals treated with saline (Saline-Saline) or haloperidol (Haloperidol-Saline) alone.

DISCUSSION

The data presented replicate the findings of Barrett and Leith [2] and Leith and Barrett [20,21] in that chronic amphetamine altered responding for intra-cranial self-stimulation. In the present experiment in addition to depressed response rates, it was shown that chronic amphetamine elevated reward thresholds compared to pre-drug levels. Thus, a higher current intensity was required to sustain responding. This is an important finding since the threshold measure is not directly dependent on rate of responding and suggests that the changes observed in self-stimulation behavior following chronic amphetamine are related to changes in the physiological processes mediating reward rather than to non-specific disruption of performance. Further support for this comes from inspection of the individual response rate×current intensity profiles. Although not evident from the grouped data presented in Fig. 2, the individual profiles clearly chow that the primary effect of chronic amphetamine was to produce a shift in the

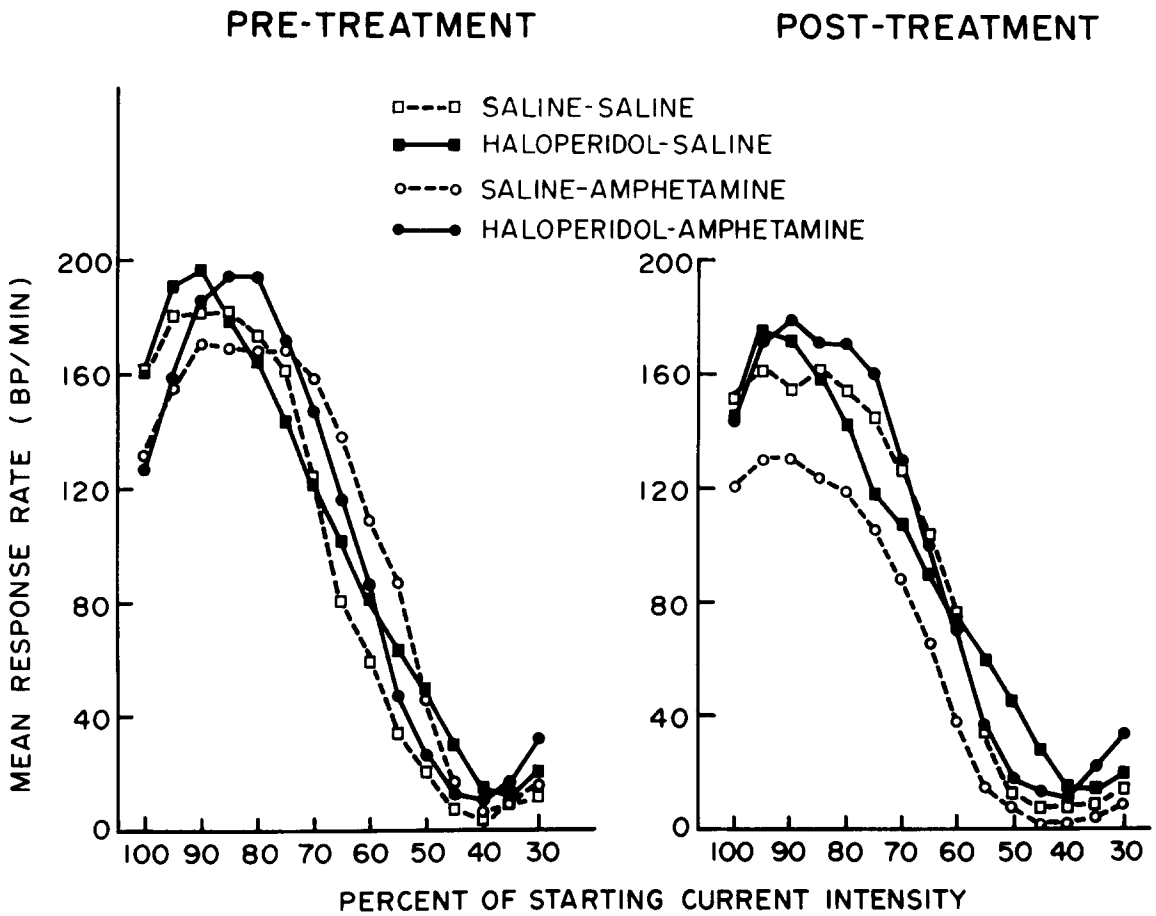

FIG. 2. Response rate plotted as a function of 15 descending current intensities demonstrating suppression of ICSS responding subsequent to treatment with chronic amphetamine and the antagonism of this effect by pre-treatment with haloperidol.

rate×intensity function similar to what is found when the starting current intensity is lowered.

Although previous experiments have reported alteration in brain serotonin [11, 29, 30], norepinephrine [22,26], and dopamine [12,17] function following repeated amphetamine administration, the finding in the present experiment that haloperidol effectively blocked chronic amphetamine's effects on self-stimulation responding suggests that post-amphetamine depression involves changes at the dopamine receptor which might, via feedback mechanisms, alter other aspects of dopamine neural activity as well. The present data do not distinguish between changes related to pre-synaptic (autoreceptor) versus post-synaptic receptor modification nor the extent to which changes in other neurotransmitter systems dependent on dopaminergic activity are involved in mediating reward system modifications following repeated amphetamine administration. However, what these data do seem to indicate is that dopamine receptor stimulation is a necessary condition for chronic amphetamine induced changes in self-stimulation.

The finding that haloperidol blocked post-amphetamine depression also argues against the interpretation that altered self-stimulation responding is due to the accumulation of the amphetamine metabolite, p-hydroxynorephedrine (PHN) in norepinephrine neurons. This metabolite has been reported to function as a false neurotransmitter which displaces endogenous norepinephrine and can reduce noradrenergic neural activity [6,13]. Previous reports have implicated PHN in explaining the development of tolerance to some of amphetamine's physiological effects [4]. However, since haloperidol does not seem to significantly alter the normal metabolism of amphetamine, it is unlikely that the behavioral change reported here is related to PHN. This conclusion is supported by the recent finding (Barrett, manuscript in preparation) that blocking the formation of PHN by preceding chronic amphetamine injections with iprindole, a drug which prevents the parahydroxylation of d-amphetamine [13], enhances chronic amphetamine depression of responding for intra-cranial self-stimulation.

In conclusion, the finding that chronic amphetamine disrupts responding for rewarding brain stimulation in rats and that pre-treatment with a dopamine receptor blocker effectively prevents the development of this effect, seem relevant to understanding the physiological basis for post-amphetamine depression in humans. Furthermore, the present data as well as previous reports [20,21] suggest that chronic amphetamine eventually results in an affective state opposite to that produced by amphetamine acutely. This "depressed" affect would predict the strong dependency which develops in amphetamine users since a primary motivation for continued drug use would be to avoid the depression which would accompany drug withdrawal.

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