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Effects of Dopamine Supersensitivity on Lateral Hypothalamic Self-Stimulation in Rats

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ETTENBERG, A. AND P. M. MILNER. *Effects of dopamine supersensitivity on lateral hypothalamic self-stimulation in rats.* PHARM. BIOCHEM. BEHAV. 7(6) 507–514, 1977. – Dopamine (DA) receptor supersensitivity was demonstrated by potentiated d-amphetamine stereotypy after a three-day treatment regimen in which the DA receptor blocker pimozide (4.0 mg/kg) was administered twice daily. Similarly-induced DA supersensitivity produced a significant increase in the rate of lever-pressing for lateral hypothalamic (LH) intracranial self-stimulation (ICSS) and a significant decrease in ICSS thresholds. No change from pretreatment baselines was observed in vehicle-treated control animals. Following three-day treatment with the noradrenaline–(NA) and DA-receptor blocker, haloperidol (4.0 mg/kg twice daily), a single injection of the alpha-adrenergic agonist clonidine (0.15 mg/kg) caused increased running behavior. In contrast clonidine decreased running in rats pretreated with chronic pimozide or vehicle. These results indicate an increase in the sensitivity of central NA receptors following chronic haloperidol but not chronic pimozide. Taken together, these findings were interpreted as a potentiation in the reinforcing properties of LH–ICSS after chronic pimozide treatments due to increases in the sensitivity of DA and not NA receptors.

Dopamine supersensitivity Self-stimulation Pimozide Haloperidol Amphetamine stereotypy Clonidine Locomotor activity

ALTHOUGH an involvement of central catecholamine (CA) neurons in the mediation of intracranial self-stimulation (ICSS) has been fairly well established [6,15], specific attempts at distinguishing between the relative roles of noradrenaline (NA) and dopamine (DA) have not yet met with any clear success. A major contributing factor to this problem has been that most of the data presented as evidence of mediation by one system, the other, or both, are derived from techniques that act to decrease ICSS behaviors. Demonstrations of decreased ICSS behaviors are, however, most difficult to interpret since surgical or pharmacological procedures that produce such results may do so by causing general malaise, sedation, motor, arousal or sensory deficits all of which may be independent of reward.

Fibiger *et al.* [13], for example, reported that cumulative records of ICSS responding after DA receptor blockade or 6-hydroxydopamine revealed a uniform suppression of responding throughout the experimental session. These results were interpreted in terms of a motor deficit in treated animals since, if the rewarding properties of the ICSS had been reduced, the cumulative record should have shown an extinction curve. There is no way of knowing, therefore, whether the reductions in ICSS rates were the result of a decrease in the reinforcing properties of the stimulation or an alteration in the animals' ability to perform the required responses.

The possibility that various CA manipulations produce reductions in ICSS behaviors by performance deficits of some kind is further supported by demonstrations that supposedly specific DA receptor blocking or NA receptor blocking agents reduce ICSS from electrodes located in both DA and NA brain sites [14, 24, 25]. Fibiger *et al.* [13] have therefore concluded: "It remains possible of course that dopaminergic systems serve as important substrates of reward . . . Our results indicate, however, that demonstration of such a role will require techniques other than those which have been most commonly used. (p. 26)" In view of this, Ettenberg and Wise [11] proposed that recent pharmacological techniques developed to produce central DA receptor supersensitivity might be of use in clarifying the presently uncertain role of DA in ICSS.

Receptor supersensitivity refers here to an increased sensitivity of postsynaptic receptors deprived of their normal neurotransmitter for a period of time ranging from several days to several weeks. The phenomenon can be demonstrated centrally following either the destruction of presynaptic fibers [12, 36, 38] or long-term postsynaptic receptor blockade with various drugs [16, 33, 35].

If a dopaminergic substrate is in some way involved in the mediation of ICSS reinforcement, then treatments that produce DA receptor supersensitivity might be expected to increase the reinforcing properties of any given level of brain stimulation. Ettenberg and Wise [11] have demonstrated similar increases in ICSS rates with either locus coeruleus or substantia nigra electrode placements following chronic pimozide treatments. Although these results might be interpreted as implicating a dopaminergic substrate in ICSS reinforcement, other explanations can certainly be proposed that adequately account for the data.

It may be, for example, that increases in ICSS rates were a result of increases in arousal or general activity following DA receptor blockade. It is also possible that chronic pimozide treatments produced an alteration in noradrenergic as well as in dopaminergic functioning. In the first case an increase in ICSS rates could be explained independently of any change in the reinforcing properties of the stimulation and, in the second case, even if an increase in the reinforcement-value of the ICSS occurred, it remains to be determined whether the increase was caused by DA or NA.

EXPERIMENT 1

In producing DA receptor supersensitivity Ettenberg and Wise [11] used a treatment regimen which involved twice daily injections of the DA receptor blocker pimozide over an eight-day period. Most other researchers have also used treatments which produce receptor blockade over periods of one to three weeks. A recent report by Christensen *et al.* [4], however, has demonstrated potentiated apomorphine-induced and methylphenidate-induced gnawing following single injections of various catecholamine receptor blocking drugs. These results suggest that much shorter periods of receptor blockade may be sufficient to demonstrate receptor supersensitivity.

An intermediate position was taken for the present study where a three-day period of DA receptor blockade was employed. Experiment 1 was done to determine whether such a treatment does in fact produce DA receptor supersensitivity. Since amphetamine stereotypy is generally believed to be mediated by central DA neurons [26,34] increase in the intensity of stereotypic responses to d-amphetamine following the three-day DA receptor blockade was taken as a measure of DA receptor supersensitivity.

METHOD

Animals

Sixteen male Sprague-Dawley rats weighing between 275 and 300 g were used. Each animal was individually housed and provided with ad lib access to food and water.

Procedure

Each animal was injected in its home cage with 4.0 mg/kg d-amphetamine sulfate – a dose which had previously been determined to produce moderate levels of stereotypic behavior (unpublished data). The d-amphetamine was dissolved immediately before use in normal saline solution and injected intraperitoneally (IP) in a volume of 1.0 mg/kg body weight.

Stereotypy of each animal was rated during 15-sec observation periods according to a five-point scale (where 0 = no stereotypic behavior and 4 = highly stereotypic or repetitive body movements combined with convulsive gnawing and biting at wires of the cage) modified from Ernst [10]. Stereotypy ratings were made for each animal every five min for 90 min and all ratings were made by the same observer without knowledge of the group to which each animal belonged.

Twenty-four hr after the completion of the initial 90-min stereotypy test just described, each animal was randomly assigned to either of two equal groups. The experimental animals were injected with 4.0 mg/kg pimozide twice daily (11:00 a.m. and 11:00 p.m.) on three successive days. Pimozide was dissolved in a hot aqueous solution of six parts tartaric acid to one part pimozide and was injected IP in a volume of 4.0 ml/kg body weight. The control animals were injected with the same volume of solution containing only the tartaric acid.

Forty-eight hr after the final injections of pimozide or vehicle, each animal was again injected with 4.0 mg/kg d-amphetamine sulfate. This posttreatment stereotypy test was conducted in an identical manner to the pretreatment test previously described.

RESULTS

Posttreatment stereotypy scores for animals withdrawn from pimozide were significantly higher than pretreatment stereotypy scores for the same animals. This was not true of vehicle-control animals whose stereotypy scores were essentially the same before and after tartaric acid. Figure 1 shows the mean scores for the experimental and control groups before and after treatments with pimozide or vehicle.

The peak behavioral effects of a single injection of 4.0 mg/kg d-amphetamine sulfate occurred at 80 min following injection. The mean pretreatment stereotypy scores at 80 min were 1.50 for the control group and 1.75 for the experimental group (out of a maximum of 4.00). Forty-eight hr after vehicle treatments the 80 min score for the control group was slightly elevated to 1.75 (a nonsignificant increase). The mean score for the experimental group 48 hr after termination of pimozide, however, was greatly increased to 3.13 (one-tailed t-test for correlated means; $t(7) = 2.99, p<0.05$).

DISCUSSION

Stereotypic responses to a single injection of d-amphetamine were of a significantly greater intensity after three-day treatment with pimozide than before such treatment. No change in the intensity of the stereotypy was observed after vehicle treatments. Since pimozide is believed to act by specifically antagonizing central dopamine receptors [1] and since amphetamine stereotypy is believed to be mediated by a central DA substrate [26,34]. It seems reasonable to suggest that these data provide a behavioral demonstration of DA receptor supersensitivity.

The initial studies of catecholamine receptor supersensitivity, on the cat nictitating membrane [22], demonstrated that the phenomenon developed only gradually over a period of several weeks [21]. However, recent experiments on central DA receptors have demonstrated supersensitivity shortly after single injections of various CA-acting drugs [4,39]. The present demonstration of potentiated d-amphetamine stereotypy after only three days of DA receptor blockade further supports the contention that central DA supersensitivity may develop soon after a relatively short period of receptor blockade.

EXPERIMENT 2

Although Ettenberg and Wise [11] demonstrated increased ICSS rates of responding during DA receptor supersensitivity, it has long been known that a change in lever-press rate alone is a weak index for measuring changes in the reinforcement value of the stimulation [18]. It is important therefore, to determine the effects of DA supersensitivity (as produced in Experiment 1) on ICSS

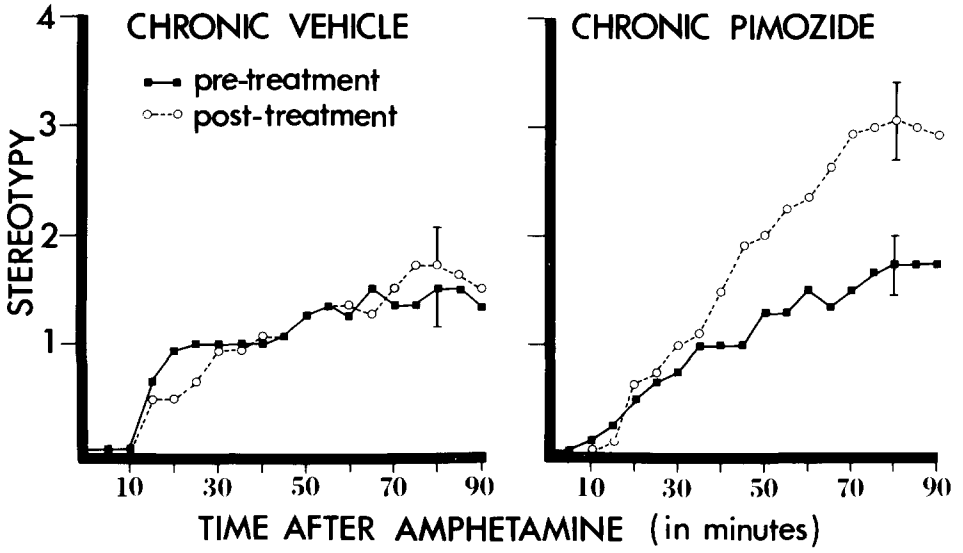

FIG. 1. Mean stereotypy-intensity scores from a single dose of *d*-amphetamine (4.0 mg/kg) before and after chronic vehicle or pimozide. Standard errors are shown at the time of peak behavioral effect.

thresholds. If DA supersensitivity should somehow produce an increase in the reinforcing properties of ICSS, then one would expect an increased rate of responding for ICSS for a given current intensity and a concomitant lowering of ICSS thresholds. Experiment 2 was devised to test this hypothesis.

METHOD

Animals

Thirteen rats similar to those used in Experiment 1 were used.

Surgery

Prior to surgery animals were allowed seven days to adapt to the laboratory environment. Each animal was then anesthetized with an IP injection of 50 mg/kg sodium pentobarbital (Nembutal) and stereotaxically implanted with a 250 μ bipolar electrode (Plastic Products) aimed at the lateral hypothalamus. The tooth-bar was set at 3.2 mm above the interaural line and the coordinates were: 0.8 mm posterior to Bregma; 1.5 mm lateral to midline; 8.6 mm ventral to skull surface.

Apparatus

The self-stimulation chamber was made of wood with dimensions of 30 × 30 × 30 cm. The lever was located 2.5 cm above the floor of the chamber, protruded 5 cm from the wall and was 5 cm wide. Each press of the lever produced a 0.5 sec train of intracranial stimulation originating from a 60 Hz sine-wave stimulator. Current intensity was controlled by a potentiometer and monitored with a microammeter.

Procedure

The animals were allowed 10 days to recover from surgery, after which each was trained to lever press for intracranial stimulation. The training procedure also involved adjusting the current intensity for each animal to a value that produced a steady rate of responding (range: 20–45 μa).

Testing involved two successive sessions for each animal each day. During the first session, which lasted 15 min, ICSS rates were measured for each animal's predetermined current level. During the second session, which immediately followed the first, ICSS thresholds were determined. Beginning with a stimulation current level at which a rat was bar-pressing steadily, the current was then lowered in 5 μa steps. At each step the animal was given five trains of non-contingent stimulation and then allowed one min to adjust to the new stimulation intensity. The number of responses made during the next three min was then recorded. This procedure continued until the rat made fewer than five responses in a three min period. The current was then raised by 5 μa, the animal given 5 trains of non-contingent stimulation, and the procedure repeated. Three ascending and three descending runs were averaged such that threshold was defined as the average current intensity above which a rat made more than five responses and below which an animal made five or fewer responses in three min.

These tests were done every day for a three week period, by which time both ICSS rates and thresholds had stabilized. The mean ICSS rates and thresholds for each animal were then calculated for a period of three successive days which constituted a pretreatment baseline. Following the determination of baselines, pimozide or vehicle injections were begun.

Eight of the animals were chosen at random, half of which were injected intraperitoneally with 4.0 mg/kg pimozide twice daily (11:00 a.m. and 11:00 p.m.) for three days and half of which were injected with the vehicle solution of tartaric acid (as in Experiment 1). Forty-eight hr after the final injections daily testing resumed in the manner previously described and continued for three days by which time ICSS rates and thresholds had returned to pretreatment baselines.

Five days after return to baseline another three-day baseline was determined. Then twice daily pimozide and

vehicle injections were started again and the procedure previously described was repeated in its entirety. This time, however, the animals that had previously been given pimozide now received tartaric acid, while those that had previously been given the vehicle now received pimozide.

The remaining five animals constituted a control group used to test the effects of DA supersensitivity on operant levels of responding. These animals were each injected twice daily (11:00 a.m. and 11:00 p.m.) with 4.0 mg/kg pimozide for three consecutive days as previously described. Forty-eight hr after the final pimozide injection each animal was placed in the experimental chamber for 31 min with the stimulation off. The first min was provided as an adjustment period during which no data were recorded. The number of responses made by each animal during three min periods was recorded over the remaining 30 min.

All 13 animals were then sacrificed and perfused with physiological saline, followed by a 10% Formalin solution. The brains were removed and fixed in 10% Formalin after which electrode placements were determined from 40 μ thionin-stained frozen sections.

RESULTS

Figures 2 and 3 show individual performances and group means for ICSS response rates and thresholds, respectively, during the three-day test-period (expressed as percent of baseline). All eight animals demonstrated at least some increase over baseline in their rate of lever-pressing for ICSS during withdrawal from pimozide. In addition, six out of eight animals demonstrated lower than baseline thresholds during pimozide withdrawal. This was not the case following vehicle treatments where, compared to baseline, animals generally showed slightly decreased rates of ICSS and slightly elevated thresholds. Although there were several days between the measurement of the first and second baselines, baselines for ICSS rates and thresholds did not change significantly over that period of time.

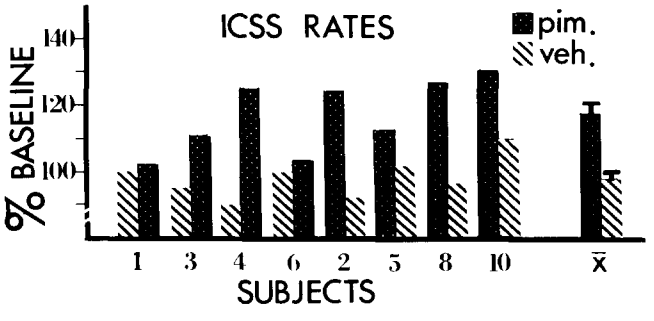

FIG. 2. Mean rates of ICSS responding over three test-days for each animal after chronic vehicle and pimozide, expressed as percent of pretreatment baseline. The overall means and standard errors of the means are indicated at the far right. For each subject the bar on the left indicates which treatment (chronic vehicle or chronic pimozide) was administered first.

Two-tailed t-tests for correlated means were done to determine whether the performance of animals on the three test-days differed from their performance on the three baseline days. During pimozide withdrawal the rate of ICSS responding increased significantly from a mean baseline of 638.42 to 749.38 responses per session ($t(7) = 4.04$, $p<0.01$). In addition, following pimozide injections, there was a significant decrease in ICSS thresholds, from a baseline mean of 20.23 μa to 15.34 μa ($t(7) = 5.01$, $p<0.01$). There were no significant changes from baseline in ICSS thresholds or rates of ICSS responding following injections of tartaric acid (vehicle).

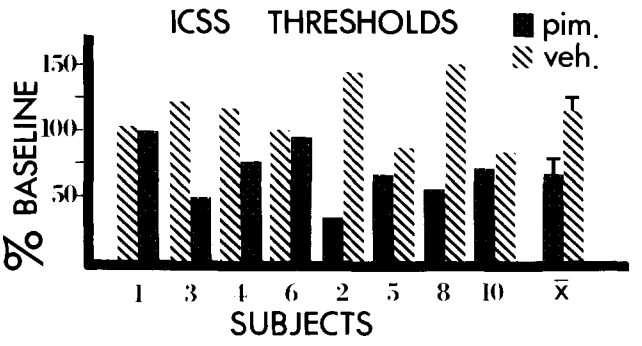

FIG. 3. Mean ICSS thresholds over three test-days for each animal after chronic vehicle and pimozide, expressed as percent of pretreatment baseline. The overall means and standard errors of the means are indicated at the far right. For each subject the bar on the left indicates which treatment (chronic vehicle or chronic pimozide) was administered first.

Pearson product-moment correlation coefficients were calculated in order to assess the degree of relationship between the performance of animals on the rate and threshold measures. Although the correlation between percent changes from baseline for rate and threshold measures was strong after vehicle treatments ($r = -.71$) it was rather poor following chronic pimozide ($r = -.053$). This result again suggests that, as has been previously stated [18], rate changes alone may not be a very good indicator of changes in reinforcement.

In the no-stimulation control group, although all five animals did respond during their first min in the experimental chamber, during no subsequent three-min period did the mean for the group exceed or equal five responses.

Histology revealed that all thirteen rats had electrode tips in the area of the lateral hypothalamus dorsolateral to the fornix.

DISCUSSION

During pimozide-induced supersensitivity, rates of responding for lateral hypothalamic ICSS increased significantly while vehicle treatments produced no significant changes from baseline. These data support the findings of Ettenberg and Wise [11] who reported 25% increases in ICSS responding following pimozide treatments, and more recently those of Simpson and Annau [31] and Eichler *et al.* [9] who demonstrated significant increases in ICSS responding during withdrawal from chronic exposure to chlorpromazine (a CA receptor blocker) and chronic exposure to spiroperidol (a DA receptor blocker), respectively.

Although increased ICSS rates during DA supersensitivity might be explained as an increase in the reinforcing properties of the stimulation, other hypotheses can account for these results. There is, for example, a great deal of evidence implicating DA pathways in the mediation of various motor responses [2, 5, 29]. Some investigators,

therefore, have argued that while DA pathways may be involved in central reinforcement mechanisms they are also critically involved in the performance of operant responses [13]. If this were the case, treatments which result in an increase in the postsynaptic sensitivity of DA receptors might produce various forms of hyperactive behavior. Increased ICSS rates might therefore be explained without reference to reinforcement.

This hypothesis seems unlikely, however, in view of the fact that following pimozide treatments animals demonstrated a significant decrease in ICSS thresholds. If increased rates of responding were a result of general hyperactive behavior, no drop in ICSS thresholds would be expected since increased activity in itself should produce no change in the reinforcing properties of the stimulation. This is further supported by the fact that operant response rates (for no stimulation) determined for the five control animals treated with pimozide, was never found to be above five responses per three minutes which was defined here as threshold. Threshold changes themselves cannot therefore be explained away as resulting from an increase in operant response rates.

It may be argued that increased ICSS response rates and decreased ICSS thresholds after chronic pimozide, resulted from heightened arousal and not from any change in the reinforcing nature of the ICSS. While this remains a possibility it is nevertheless an unlikely one. The authors know of no data demonstrating a reduction in ICSS thresholds during increased arousal. Deutsch and Howarth [7] have shown that rats having undergone extinction of a lever-press response for ICSS will spontaneously commence responding during the presentation of fear-producing stimuli. However, the arousal produced in that situation was reported to greatly increase operant levels of responding. The control group in the present experiment shows no such increase in operant responding. Furthermore, Deutsch and Howarth [7] reported that the evocation of ICSS responding with fear-producing stimuli was very difficult to demonstrate with rats trained with electrodes in the hypothalamus. The present study did, of course, involve lateral hypothalamic self-stimulation.

A more likely explanation for the increased ICSS responding and decreased thresholds demonstrated after chronic pimozide might be that DA supersensitivity produces an increase in the reinforcing nature of lateral hypothalamic ICSS. This hypothesis, although adequately accounting for the data, is in no way conclusive and further research is needed to rule out alternative explanations. Furthermore, should this hypothesis be correct, the present data do not suggest how the increase in the reinforcement value of the stimulation might occur. Further work would be necessary to determine, for example, whether such a proposed increase in ICSS reinforcement results from a direct manipulation of a central DA reinforcement system or through the role of a modulatory non-reinforcement DA system.

EXPERIMENT 3

Although pimozide is reported to be a highly specific DA receptor blocker [1] a number of recent reports have been published suggesting that pimozide may also affect the functioning of central NA neurons [3,32]. These reports, combined with the fact that Ettenberg and Wise [11] demonstrated increases in ICSS with electrodes in the NA locus coeruleus after chronic pimozide, cast some doubt on the specificity of the pimozide treatment.

There is a large body of evidence clearly implicating central DA pathways in the mediation of locomotor activity [19,28]. Further evidence exists, however, suggesting at least some modulatory role of NA in locomotor activity. This evidence consists primarily of reports that demonstrate increases in locomotor behavior following NA stimulation. Segal and Mandell [30], for example, have shown that intraventricular infusion of NA can produce significant increases in locomotor activity and arousal. Related to this, Randrup and Scheel-Kruger [27] have demonstrated that diethyldithiocarbamate, which acts in part to inhibit dopamine-beta-hydroxylase (the enzyme responsible for converting DA into NA) thus producing a decrease in NA content, did not block amphetamine-induced stereotypy but did inhibit the usual increase in locomotor activity seen after amphetamine.

More recently, Dunstan and Jackson [8] have reported that clonidine, a selective alpha-adrenergic receptor agonist, produced a marked increase in the locomotor activity of animals withdrawn from long-term treatment with haloperidol. This stimulatory action of clonidine seen in haloperidol-treated animals was not evident in vehicle-treated animals thereby providing evidence for an increased sensitivity of central NA receptors after chronic haloperidol.

If the pimozide treatments used in the previous two experiments were producing an increase in the sensitivity of central NA receptors, then based on the work of Dunstan and Jackson [8], one should be able to demonstrate a locomotor stimulation with clonidine. A failure to produce increased locomotor activity with clonidine would suggest that no detectable change in NA receptor sensitivity had occurred. Experiment 3 was devised therefore to test the specificity of the pimozide treatment.

METHOD

Animals

Thirty-six male Sprague-Dawley rats similar to those used in Experiments 1 and 2 were used in the present study.

Apparatus

Two standard 14 in. (35 cm) diameter running-wheels (Lafayette Instrument Company, IN) each equipped with a mechanical counter to record the number of revolutions each animal ran during any given period of time, were used to measure locomotor activity.

Procedure

Each animal was placed in one of the two running-wheels for 15 min every day. Half of the animals were randomly assigned to one apparatus and half to the other. Each animal, once assigned, was tested on the same apparatus for the duration of the experiment.

Following a 2-week period of familiarization with the running-wheels, the rats were randomly assigned to one of three equal groups. One group received 4.0 mg/kg pimozide injected intraperitoneally (IP) twice daily for three days as in Experiments 1 and 2. Another group was similarly treated for three days with IP injections of 4.0 mg/kg haloperidol. The haloperidol was prepared in a vehicle solution of distilled water containing six parts tartaric acid

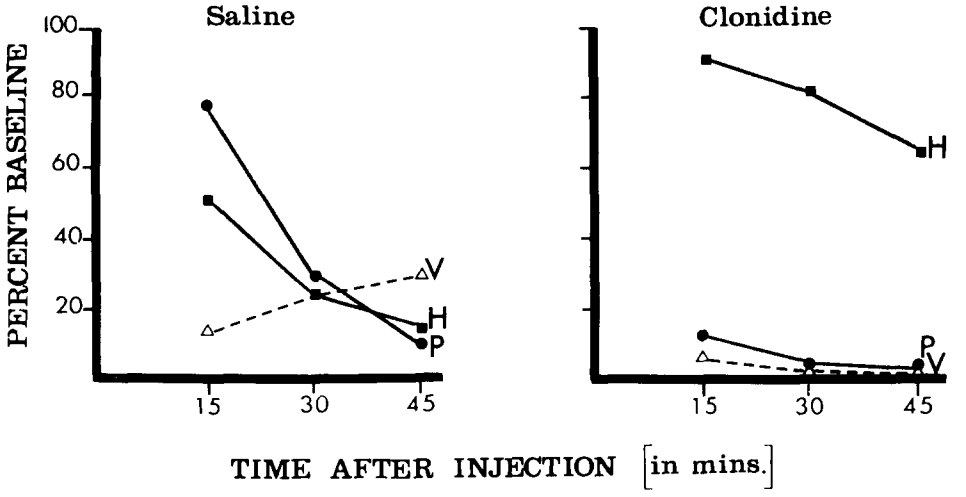

FIG. 4. Mean running-wheel performance after a single injection of clonidine (0.15 mg/kg) or saline in chronic pimozide (P), chronic haloperidol (H) or chronic vehicle (V) pretreated groups. Performance is expressed as percent of preclonidine (or presaline) baseline.

to one part drug. The injection volume was held constant at 4.0 ml/kg body weight. The third group was injected with the tartaric acid vehicle solution according to the schedule used for the pimozide and haloperidol groups.

Forty-eight hrs after the end of these treatments each animal was tested for a total of one hr in the running-wheel apparatus. The first 15 min constituted a no-drug baseline. Half of the animals in each group were then given a single IP injection of 0.15 mg/kg clonidine hydrochloride and the other half a control injection of 0.9% saline solution. Finally the animals were replaced in the apparatus and running-wheel performance was recorded 15 min, 30 min and 45 min after injection.

RESULTS

All groups but one tended to decrease their amount of running-wheel behavior over time; however, the degree to which they did so varied from one condition to another. These results are clearly illustrated in Fig. 4 where mean running-wheel performance for each group is expressed as percent of preinjection baseline.

An analysis of variance (three-factor mixed design with repeated measures on one factor) was computed on the percent-change data of each group from baseline. The analysis revealed a significant main effect of Pretreatment, $F(2,30) = 4.99$, $p<0.05$, indicating that animals' rate of running differed according to what pretreatment was administered. Figure 4 clearly illustrates the differences in performance between vehicle pretreated, pimozide pretreated, and haloperidol pretreated subjects. The main effect of Trials, $F(2,60) = 4.40$, $p<0.05$, indicates a significant change in performance as one tests at different times after saline or clonidine injections. More specifically all but one group of animals (vehicle pretreated, saline injected) showed decreases in mean running-wheel performance as one increased the time after saline or clonidine injection. This is illustrated by the negative slopes of the curves drawn in Fig. 4.

The final significant effect in the analysis was a highly significant Pretreatment × Drug interaction, $F(2,30) = 12.69$, $p<0.001$. The drug-factor in this case was the effect of saline or clonidine injections. The interaction, therefore, indicates that the effect of these injections on running-wheel performance differed for different types of pretreatment. In other words, animals' performance after saline differed for vehicle pretreated, pimozide pretreated and haloperidol pretreated animals. In addition, clonidine, which had a depressant effect on the running-wheel behavior of vehicle pretreated and pimozide pretreated animals, had a stimulatory effect on the performance of haloperidol pretreated animals.

DISCUSSION

Clonidine at the dose used in the present study (0.15 mg/kg) has been shown to depress exploratory behavior [23] and self-stimulation responding [17]. The demonstration of depressed running-wheel behavior after clonidine in vehicle pretreated animals is therefore consistent with these findings. Since animals withdrawn from pimozide respond to clonidine in the same way as vehicle pretreated animals (i.e., they show a marked depression of running behavior), there is no evidence of any change in NA receptor sensitivity after chronic pimozide.

Haloperidol pretreated animals, however, were running at 92% of baseline 15 min after clonidine and still performing at 67% of preclonidine baseline 45 min after clonidine. Haloperidol while predominantly a DA receptor blocker, does have significant NA receptor blocking properties in high doses [1]. It is therefore conceivable that the relatively high dose of 4.0 mg/kg haloperidol administered twice daily might have produced an increase in the sensitivity of noradrenergic as well as dopaminergic receptors as suggested by Dunstan and Jackson [8]. If central NA neurons did play some role in locomotor activity as some have suggested [27, 30, 37], then the stimulatory effect of clonidine on running in haloperidol but not vehicle pretreated animals, could be explained by an increase in the postsynaptic sensitivity of central NA receptors after three-day blockade with haloperidol.

The effect of saline injections on running-wheel performance also differed with respect to pretreatment. Fifteen min after saline injections, pimozide and haloperidol

pretreated groups were still responding at 77% and 51% of pretreatment baselines, respectively. This increase in running demonstrated in pimozide and haloperidol pretreated groups supports the contention that supersensitivity of DA receptors may lead to some forms of hyperactive behavior.

GENERAL DISCUSSION

Administration of 4.0 mg/kg pimozide to rats twice daily for three days produces (48 hr after the final injection) a DA receptor supersensitivity as demonstrated by significant posttreatment increase in stereotypic responses to d-amphetamine. The nature of the mechanism underlying the phenomenon is at present unknown although it is unlikely that central receptor supersensitivity develops in the same manner as peripheral receptor supersensitivity since the time courses of development of the two phenomena differ greatly. Central supersensitivity can occur, as in the present study, within 48 hr of a relatively short period of receptor blockade while the peripheral phenomenon requires chronic receptor blockade and takes several weeks to develop.

It is proposed that the significant increases in LH–ICSS response rates and the significant decreases in LH–ICSS thresholds demonstrated during pimozide-induced DA supersensitivity, reflect an increase in the reinforcing properties of the brain stimulation. Although DA supersensitivity does produce increased locomotion (Experiment 3), the ICSS results cannot adequately be explained by a simple hyperactivity hypothesis since while such a hypothesis might predict increased responding for suprathreshold stimulation it would not predict any decrease in ICSS thresholds (i.e., the reinforcing properties of the stimulation should not change during hyperactive behavior alone). In addition, there was no significant increase in the operant response rates (for no stimulation) after pimozide treatments in a control group thereby further weakening the hyperactivity model.

Furthermore, in Experiment 3 the stimulatory effect of clonidine demonstrated in haloperidol pretreated animals was not evident in vehicle pretreated animals suggesting a change had occurred in the sensitivity of central NA neurons after three days of haloperidol. This finding further suggests that previous reports of dopamine receptor supersensitivity after chronic haloperidol must be viewed with caution, since NA supersensitivity may also be accounting for the reported findings.

The effect of clonidine in the pimozide pretreated group, however, was identical to its affect on the vehicle pretreated group (see Fig. 4). There is therefore no evidence to suggest any increase in the receptor sensitivity of NA neurons after three days of pimozide injections. The proposed increase in the reinforcing properties of LH–ICSS during pimozide-induced DA supersensitivity cannot therefore be attributed to a nonspecific increase in the sensitivity of NA fibers in the LH or elsewhere.

These findings, of course, do not suggest the mechanism by which the DA receptor supersensitivity produces its effect. It may be, for example, that DA receptor supersensitivity could potentiate ICSS reinforcement by direct facilitation of a central DA reinforcement system. However, it is equally possible that DA neurons might affect reinforcement only via their modulatory influence on some other non-DA system. Either way, the data from these experiments can be taken as evidence for a significant role of some kind for central DA neurons in LH–ICSS reinforcement. Furthermore, they suggest that pimozide-induced supersensitivity may represent a valuable tool for more clearly assessing the functional role of DA systems in other behaviors.

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