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Time Course of Dopaminergic Hypersensitivity Following Chronic Narcotic Treatment

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CARLSON, K. R. AND J. ALMASI. *Time course of dopaminergic hypersensitivity following chronic narcotic treatment.* PHARMAC. BIOCHEM. BEHAV. 11(3) 283–287, 1979.—Guinea pigs were injected SC for 3 weeks with 3 different dosage schedules of morphine or methadone, or with saline. For 8 weeks thereafter they were challenged weekly with the dopamine agonist apomorphine. Hypersensitivity was manifested in more intense stereotypies, as compared to the saline group, by all morphine and methadone groups. Hypersensitivity persisted longer after the termination of methadone treatment (maximum of 8 weeks) than after morphine administration (maximum of 3 weeks). The degree of hypersensitivity, and its duration after treatment, was positively related to methadone dosage. In some groups a period of hyposensitivity was seen following hypersensitivity. These data are interpreted with reference to the hypothesized mechanism underlying the development of hypersensitivity, the different durations of action of morphine and methadone, and the retention of methadone in brain following treatment.

Dopamine Hypersensitivity Morphine Methadone Apomorphine Narcotics

WHEN animals are treated chronically with narcotic analgesics, they become hypersensitive to dopaminergic agonists. In tests conducted immediately following morphine treatment, hypersensitivity is manifested in more intense aggression or stereotyped behaviors in response to l-dopa, amphetamine, or apomorphine in mice and rats [11, 16, 20].

Dopaminergic hypersensitivity can persist longer than the phase of acute abstinence. For example, enhanced aggression in response to apomorphine was seen for 30 days following morphine treatment in rats [12], and enhanced stereotypies were elicited by methamphetamine in guinea pigs for 3 weeks following methadone treatment [9]. Further, rhesus monkeys which had been treated chronically with oral methadone were hypersensitive to methamphetamine for as long as 17 months [4, 8, 10], and a monkey challenged with apomorphine 26 months following methadone was still hypersensitive [5].

In a recent study [6] we treated guinea pigs with parenteral methadone (MD) or morphine (MS) for 3 weeks (20 mg/kg/day the first week, 30 mg/kg/day the second week, and 40 mg/kg/day the third week), and challenged with apomorphine (APO) 1 week and 5 weeks following treatment. The MS group showed enhanced stereotypies, in comparison to a control group which had been treated chronically with saline, during the first test, but this hypersensitivity had dissipated by the second test. In contrast, the MD group was not only hypersensitive during both tests, but the intensity of their stereotypies actually increased from the first to second tests. These data suggest that although MD and MS are equianalgesic in this species [26], they may not be equipotent in producing dopaminergic hypersensitivity, and that there may be different time courses for the retention or expression of hypersensitivity following treatment with these two narcotics. Thus, in the present experiment we systematically investigated this question by treating for 3 weeks with different dosage schedules of MD and MS, and at weekly intervals thereafter measuring the intensity of APO-elicited stereotypies in a manner identical to that used earlier [6].

METHOD

Subjects

Male albino guinea pigs of the English smooth hair strain were used. Since intrinsic sensitivity to APO increases with age [7], to control for this variable Ss were ordered from the supplier (Camm Research Institute) with a specified birthdate ± 1 day. Ss were 3 weeks old on arrival and were given a 1-week adaptation period before the experiment was begun. They were housed 5 or 6 to a cage with continuous access to Purina guinea pig chow and tap water. Ss were weighed before each increase in chronic drug dosage and on the first day of each APO test series.

Chronic Drug Administration

For 3 weeks, Ss received equal doses in 2 daily SC injections separated by 12 ± 2 hr. Three dosage schedules of both MD (methadone hydrochloride; Lilly; 10 mg/ml) and MS (morphine sulfate; Lilly; 15 mg/ml) were employed. The 10–20 groups received a total daily dose of 10 mg/kg the first week, 15 mg/kg the second week, and 20 mg/kg the third week. Similarly, the 20–40 groups received during those respective weeks 20, 30, and 40 mg/kg (the same schedule as in ref. [6]), and the 40–80 groups received 40, 60, and 80 mg/kg. A saline control group (SAL) received sterile physiological saline in a volume equivalent to that given the MS 20–40

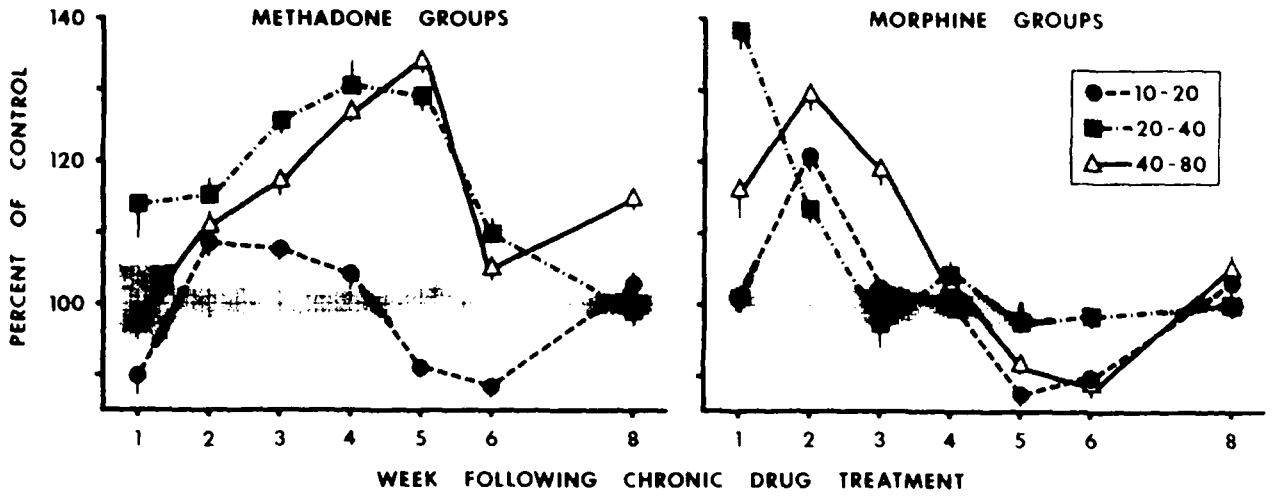

FIG. 1. Stereotypy scores as mean (± SEM) percent of control values on each week following chronic drug treatment, for the MD and MS groups. Chronic drug groups are identified in the outlined inset. The grey areas represent ± 1 SEM of the SAL group.

group. Ss were assigned randomly to groups; Ns began at 14 or 12, and eventual group Ns were between 14 and 10, owing to a few deaths which did not appear related to drug dosage since they occurred in the 10–20 groups.

APO Tests

Following the termination of chronic drug treatment, Ss were challenged with APO (apomorphine hydrochloride; Lilly). APO solutions were freshly prepared in sterile physiological saline each day (0.4 mg/ml as the salt) and kept on ice. Doses of 0.1, 0.2, and 0.4 mg/kg were injected SC at the back of the neck, the order of administration determined for each group by a Latin Square. Tests were conducted on the last 3 days of each week, i.e., Days 5–7, 12–14, and so forth, on Weeks 1, 2, 3, 4, 5, 6 and 8.

On each test day Ss were placed in individual 28×18×12 cm high Plexiglas cages with metal wire tops, located in a separate testing room. Each cage contained sawdust bedding and half a plastic specimen cup suitable for gnawing. After a 10-min adaptation period, during which no stereotypy was observed, each S was injected and rated for the intensity and continuity of stereotyped behavior (chewing and gnawing) according to an 8-point rating scale [6]. Ratings were made at 5, 10, 15, 20, 25, 30, and 40 min postinjection by an E who was blind to chronic drug group and APO dose.

Data Analysis

For each weekly test, each S's scores were averaged across the 7 postinjection observation times to generate his mean stereotypy score in response to each APO dose. The three mean scores for each S were used in a two-way (group vs dose) analysis of variance with repeated measures [27]. An analysis of variance was performed on each week's data. When a significant between-groups difference was obtained, individual comparisons were then used to determine which chronic drug group(s) were significantly different from the SAL group.

Since the SAL group's mean scores varied from week to week and rose over the course of the experiment as expected [7], in the interests of visual clarity and ease of comparison the results are presented graphically in terms of percent of control. The total of each S's mean stereotypy scores at the three APO doses was converted to a percent of the SAL group's mean total score, and these values were averaged to obtain a mean (± SEM) percent of control figure for each chronic drug group at each weekly test. Variability within the SAL group was assessed in an identical fashion, by converting each SAL S's total score to a percent of the SAL group's mean total score, and from these calculating the SEM.

In order to identify the source of differences between groups in their mean stereotypy scores, time-action data at each APO dose were also calculated, by averaging the scores obtained at each observation time post-injection.

RESULTS

The data are summarized in Fig. 1, which illustrates the extent of hyper- (or hypo-) sensitivity exhibited by each chronic drug group over the course of the experiment. The results of the statistical analyses are presented in Table 1; these F values, with their associated levels of statistical significance, are the substantiating evidence for the descriptive statements which follow.

It is apparent that all the MD groups exhibited hypersensitivity, and that the degree and duration of hypersensitivity appeared to be fairly well related to dosage schedule. The 10–20 group, for example, showed a moderate level of hypersensitivity only on weeks 2–4. The 20–40 group rose to a much higher peak hypersensitivity on Week 4 and gradually declined to control level, whereas the 40–80 group's hypersensitivity rose somewhat more slowly, and, with the exception of the anomolous point on the Week 6 test, declined more gradually as well. In fact, this was the only group which was still hypersensitive on the final test. It is particularly interesting that in all these groups the onset of hypersensitivity was not immediate, and that the two higher-dose groups required a 4-week period to rise to their peak levels. This latter result is consistent with our earlier work [6].

Although the MS groups did not show as clear a pattern relating hypersensitivity to chronic drug dosage, it is apparent that when hypersensitivity was exhibited it occurred

TABLE 1

F VALUES FOR BETWEEN-GROUP DIFFERENCES IN THE ANALYSIS OF VARIANCE (ANOVA) AND SUBSEQUENT INDIVIDUAL COMPARISONS

Week	ANOVA (df=6,80)	Comparison of SAL Group with Groups MD 10–20	MD 20–40	MD 40–80	MS 10–20	MS 20–40	MS 40–80
1	15.1‡	3.0	6.3*	<1.0	<1.0	41.8‡	7.8†
2	17.5‡	7.0†	108.6‡	12.3‡	42.2‡	20.1‡	90.3‡
3	19.7‡	4.9*	62.6‡	25.9‡	<1.0	<1.0	31.6‡
4	34.3‡	1.6	107.0‡	66.3‡	<1.0	2.0	<1.0
5	200.7‡	25.2‡	268.4‡	330.9‡	40.9‡	<1.0	20.1‡
6	37.9‡	36.5‡	27.5‡	6.9*	27.5‡	<1.0	33.8‡
8	10.8‡	1.5	<1.0	42.4‡	1.6	<1.0	4.8*

*$p<0.05$; †$p<.01$; ‡$p<0.001$

F values for within-group differences (dose-effect relation) ranged from 102. to 714.; $df=2, 160$; $p<.001$

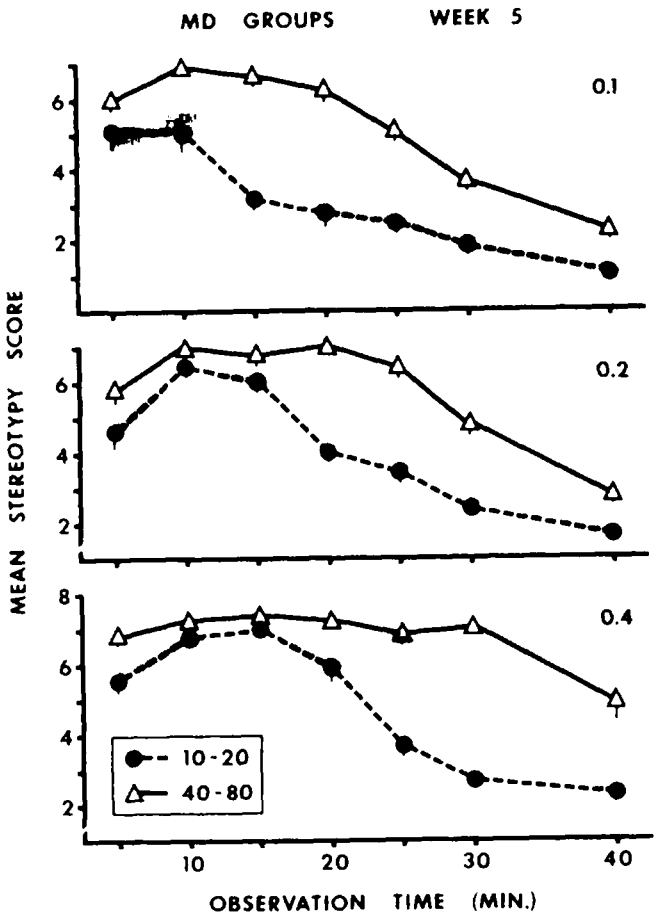

FIG. 2. Mean (± SEM) stereotypy scores at each observation time following APO administration at the Week 5 test. APO dose is indicated in the upper right corner of each panel. The two MD groups are identified in the outlined inset. The grey areas represent the mean ± SEM of the SAL group. In cases in which vertical SEM lines for the MD groups are not shown, the SEM was so small that the line would not have extended past the symbol for the mean value.

much earlier following the termination of treatment than was the case with the MD groups. For example, only in the highest dose group did hypersensitivity persist until the Week 3 test. In the 20–40 group, hypersensitivity during the first test which disappeared by Week 5 replicated our earlier finding [6].

It is norteworthy that several groups showed significant hyposensitivity at Weeks 5 and 6. Although this phenomenon occurred in two of the MS groups, it was observed in only the lowest dose MD group. It is possible that the other MD groups would have exhibited the same effect following hypersensitivity; unfortunately the experiment had to be terminated before this could be determined due to the investigators' relocation to another institution.

In order to determine whether deviations from control levels in mean stereotypy scores were due to changes in the maximal scores or the duration of stereotypy, time-action data were examined in every instance of significant hyper- or hyposensitivity. The data of two MD groups on Week 5 are convenient illustrations of the results obtained in all these cases. As can be seen in Fig. 2, both factors contributed to hypersensitivity, in that the Ss achieved higher peak scores and persisted longer in their stereotypies. The elevation of maximal scores was most obvious at 0.1 and 0.2 mg/kg APO; at 0.4 mg/kg a ceiling effect came into play, since the top score on the rating scale was 8.

Hyposensitivity, on the other hand, was attributable solely to a shortened duration of action, since the maximal scores achieved were in or above the control range. This effect was noticeable at all APO doses, but was most pronounced at 0.4 mg/kg, when an abrupt decline in the scores of hyposensitive Ss began 15 min before the control animals showed any meaningful dwindling of stereotyped behavior.

DISCUSSION

This study not only confirms previous reports [4, 5, 6, 8, 9, 10, 11, 12, 16, 20] that narcotics can induce dopaminergic hypersensitivity, but represents the first systematic investigation of the time course for the retention or expression of hypersensitivity following treatment. The results can be interpreted with reference to three subjects: evidence concerning the underlying mechanism responsible for the develop-

ment of hypersensitivity, the different durations of action of MS and MD, and the retention of MD in brain following chronic treatment.

Hypersensitivity is thought to result from prolonged blockade of dopamine receptors in the striatum; upon withdrawal of the antagonist, dopaminergic stimulation produces exaggerated stereotyped behaviors [15,23]. The most common blocking drugs are neuroleptics such as chlorpromazine and haloperidol, which induce hypersensitivity in rodents [15, 23, 24], monkeys [8, 10, 13], and man, the condition termed tardive dyskinesia [25]. Recent evidence indicates that a schedule of chronic haloperidol administration sufficient to produce behavioral hypersensitivity in rats increases the number of dopamine receptor sites in the striatum without altering their affinity [3].

Although there is some evidence to the contrary [2], a body of data indicates that narcotic analgesics may also act as dopamine blockers [1, 18, 19, 21, 22], suggesting that this quality may explain their ability to induce dopaminergic hypersensitivity. If one makes the reasonable assumption that the more continuously a blockade is imposed the greater will be the extent of hypersensitivity produced by that blockade, then the relative superiority of MD over MS in inducing hypersensitivity can be attributed to MD's longer duration of action [14], since there would be less opportunity for recovery from blockade between injections. The fact that the degree of hypersensitivity was positively related to chronic MD dose could be explained by the same argument.

An intriguing finding was the gradual emergence of behavioral hypersensitivity in the MD groups following treatment. We propose that this reflects the interplay of two factors, an existing state of hypersensitivity which is progressively unmasked as MD is gradually released from receptor sites in the brain. Even after a single injection of 10 mg/kg, appreciable amounts of MD remain bound to brain protein for up to three weeks [17]. We have administered (^{3}H)-methadone to guinea pigs on the same schedule as the MD 20–40 group in the present study, and sacrificed the Ss 6 hr, 1 week, or 5 weeks post-treatment. Not only was approximately 98% of the methadone bound, but half of the bound radioactivity which was present 6 hr after the last injection was present at 1 week and a third was still present at 5 weeks (Niehoff, Connamacher and Carlson, submitted for publication). Thus, the persistence of MD in brain, presumably at receptor sites, may provide a slowly-dissipating partial blockade which masks the existence of a hypersensitive state. The slower rate of emergence of hypersensitivity after higher chronic MD doses is consistent with this formulation.

We hypothesize that MS's shorter duration of action [14] makes it a less efficient and continuous blocking agent, and thus less capable of inducing hypersensitivity. For the same reason, hypersensitivity is manifested earlier after the termination of chronic drug treatment. We have not yet investigated the retention of (^{3}H)-morphine in brain following chronic treatment, but we would predict more rapid clearance than is the case with MD, on the basis of the present behavioral results.

As for the mechanism underlying hypersensitivity, the time-action data point to one or possibly two factors. Since peak scores were elevated, it is likely that there had been some alteration at the cellular level similar to the increased numbers of receptors seen after haloperidol [3]. Although this could account for the prolonged duration of stereotypy, it is also possible that a reduced rate of metabolism of APO was responsible. We are presently investigating these alternatives.

To our knowledge, this is the first report of dopaminergic hyposensitivity as a result of narcotic treatment, although we have seen the identical phenomenon 5 weeks after chronic haloperidol treatment [6]. The time-action data strongly suggest that the metabolism or clearance of APO is enhanced during these periods, such that the behavioral effect is terminated more rapidly. It remains obscure why this should occur, and occur only after a period of hypersensitivity. Due to the necessary but premature termination of this experiment, we could not determine whether hyposensitivity would have been exhibited eventually by the two higher-dose MD groups. The transitions between different degrees of sensitivity, following these and other narcotics with different durations of action, should be studied in long-term experiments.

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