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Delayed Effects of Amphetamine or Phencyclidine: Interaction of Food Deprivation, Stress and Dose[1]

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COVENEY, J. R. AND S. B. SPARBER. *Delayed effects of amphetamine or phencyclidine: Interaction of food deprivation, stress and dose*. PHARMACOL BIOCHEM BEHAV **36**(3) 443–449, 1990.—Tritium-labelled phencyclidine (PCP) hydrochloride (12 mg/kg) was injected SC for six consecutive days into two groups of eight male rats maintained at 85% of their initial free-feeding weights. Eight days after the last injection, electric footshock raised fat levels of PCP 28% over nonshocked controls, and lowered blood levels 18%, but did not alter brain levels of the drug significantly. Thus, application of an acute stressor does result in redistribution of tissue stores of phencyclidine as predicted in the literature; however, the direction of the redistributions was to fat, rather than to brain. To explore the relation of a long-term stressor (one that eliminates adipose tissue as a sink for mobilized PCP), exploratory behavior was evaluated in male rats during six days of food deprivation commencing after six daily injections of PCP HCl (2 or 4 mg/kg, SC). Exploratory behavior of the 4 mg/kg dose group was abruptly altered, compared to saline controls, at six days of food deprivation, when the rats' body weights were about 70% of initial weights and when body fat would be severely reduced or depleted. To assess replicability and generalizability of this phenomenon, PCP HCl (4 or 8 mg/kg, SC) or dextroamphetamine sulfate (3.2 or 6.4 mg/kg, SC) was injected into male rats for six days and food deprivation followed afterward for nine consecutive days, or until similar body weight reductions as in the first experiment were achieved. Again, exploratory behavior was altered in comparison to saline controls in phencyclidine-treated rats (at the 4 mg/kg dose level) when rats reached about 70% of initial weights. Behavior of dextroamphetamine-treated animals (at 3.2 mg/kg) was also different from controls, suggesting that interactions between food deprivation (stress) and lipophilic drugs of abuse occur when body fat stores reach a threshold.

Phencyclidine Dextroamphetamine Stress Biodistribution Behavior Rat Footshock

PHENCYCLIDINE is a drug noted for its ability to occasionally produce bizarre and violent behaviors in abusers. Although it is not as widely used among the general population as it was during previous years, its use, expecially among inner city youth and young adults, remains a public health concern. Since the mid-1970s, there have been reports of protracted psychotic episodes of two to four weeks duration after PCP use (1,17). Resurgent physiologic and psychologic toxic signs have also been documented (7,13). Accompanying these reports of protracted or even resurgent clinical signs of intoxication has been the finding of urinary reappearance of PCP in persons not suspected of further use (12). Yago et al. (24) found low levels of PCP in nearly half of psychiatric emergency admissions, an observation attributed to "recent PCP (PCP) exposure with relative clearing of blood and/or remote PCP exposure with slow remobilization from lipid stores (p. 195)."

The findings of James and Scholl (10) and of Misra, Bartolomeo and Pontami (15) lend support for this contention in that the drug was found to distribute preferentially to fat and to remain there for a protracted time. They have suggested that adipose tissue could provide a reservoir from which PCP could be supplied, upon sympathetic activation as a result of stressful stimuli, to prolong or intensify a course of intoxication. However, results from this laboratory (4) did not find an interaction between an acute stressor (electric footshock) and a recent history of PCP exposure in rats, using operant behavior as an index of drug action under circumstances in which dextroamphetamine did show such an interaction. Previously dextroamphetamine had been shown to undergo altered biodistribution after chronic administration and the application of electric footshock as an acute stressor (22).

The failure of PCP to interact with footshock in a manner similar to that of dextroamphetamine could result from several factors, among which are physicochemical differences between the drugs. These differences might alter the direction of PCP flux

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TABLE 1

TISSUE LEVELS OF PHENCYCLIDINE AFTER SUBACUTE TREATMENT AND FOOTSHOCK

Treatment Group	Tissue				
	Brain	Fat	Muscle	Blood	Heart
Nonshocked	5.31 ± 0.26	281 ± 21	19.4 ± 3.8	7.83 ± 0.35	9.69 ± 0.62
Shocked	5.39 ± 0.20	359 ± 21*	15.7 ± 4.2	6.64 ± 0.20*	8.57 ± 0.57

Values are ng PCP/g or ml (mean ± SE) for n = 8.

*Statistically significantly different from nonshocked group by Student's *t*-statistic ($p<0.05$).

during stress-induced redistribution. To clarify this difference between PCP and dextroamphetamine in their ability to interact with footshock, the first experiment described here was designed to determine what redistribution, if any, occurs with tissue stores of PCP during footshock stress. This experiment was based upon the supposition by Misra, Bartolomeo and Pontami (15) that lipid-stored PCP is mobilized by stress. A dose of 12 mg PCP HCl/kg was selected for the first experiment, since the dose that Misra, Bartolomeo and Pontami (15) used in albino rats (25 mg/kg) is 2–5 times greater than behaviorally active doses generally used.

Food deprivation of fasting is another stressor which is relevant to the nonmedical use of psychoactive drugs, including the illicit use of PCP: the anorectic action of PCP has been reported to result in 10 to 35 pounds of body weight loss during binges [(16), p. 8]. Accordingly, further studies reported here were conducted to clarify the relation between this stressor and the behavioral consequences of repeated exposure to PCP in rats.

Investigations have been done to determine the relation of such stress-induced mobilization of pesticide residues to declines in migratory bat populations (8). Results from those studies indicated that brain levels of pesticides remained low until a minimum carcass fat content was reached (6 to 9 days of food deprivation and forced exercise), beyond which brain residue levels rose sharply. Clark and Prouty (3) found analogous results in brown bats fed organochlorine environmental contaminants and then starved to death. In their study, a substantial decrease in carcass lipids had to be reached before abrupt increases in brain levels of these lipophilic organochlorine compounds were observed.

Because of the highly lipophilic nature of PCP (14), an analogous threshold might exist for it (i.e., a history of PCP injections will sensitize rats to food-deprivation stress when all or nearly all of the fat reserves are exhausted). Based upon the results of a pilot experiment (reported herein), this level is reached after about six or more days of food deprivation or a duration of deprivation sufficient to reduce rats to about 70 percent of initial weights (for rats in the 420–430 g range). The purpose of the succeeding experiments was to determine whether behavior of rats would be altered when given repeated PCP injections and then subjected to the consequences of this degree of food deprivation. To do this, the spontaneous motor activity of rats was evaluated daily, during food deprivation, beginning after the last of six daily injections of PCP. Doses of PCP (2–8 mg/kg) used in these experiments were based upon previous studies in this laboratory (5), in which comparable doses of PCP were used to study behavior in rats.

EXPERIMENT 1

Method

Sixteen male Sprague-Dawley rats (350–450 g initially) from Holtzman Co. (Madison, WI) were used in this experiment. They were housed individually in hanging wire mesh cages (20 × 25 × 18 cm) in a temperature-controlled (22 ± 1°C) and humidity-controlled (40–50%) environment under a 12-hour light/dark cycle (on from 0700 hr). They were fed on rations of a standard laboratory rat diet (Purina Rat Chow) to maintain weights at approximately 85% of free-feeding weights, recorded at the beginning of the experiments. Water was available ad lib in the home cages. Rats received tritium-labelled PCP HCl (National Institute on Drug Abuse, Rockville, MD) at a dose level of 12 mg/kg (100 μCi/kg) subcutaneously in the flanks for six consecutive days. On the eighth day after the last injection, the rats were randomly subdivided into two equal groups, one of which was subjected to a moderate footshock stress (2 mA, 0.5 sec duration for 15 random presentations on the average of once every 64 sec) in standard small-animal operant chambers.

Thirty minutes later, rats were decapitated and tissues [brain, blood collected from the trunk into beakers containing 0.1 ml of heparin sodium (1000 U/ml), epididymal fat, abdominal muscle and heart] were taken and stored at −70°C until assayed. Assay of tissues and blood for tritium-labelled PCP was performed by the method of Misra, Pontami and Bartolomeo (15): tissues were spiked with 1 mg PCP HCl carrier and homogenized in four-to-five volumes of 0.1 N HCl. Homogenates were alkalinized with ammonia water, buffered with 20%, w/v, potassium dihydrogen phosphate, and extracted with 15 ml of cyclohexane in an Eberbach mechanical shaker. An aliquot of the cyclohexane extract was acidified with ethereal hydrochloric acid and dried in liquid scintillation counting vials on a microscope slide warmer. The residue was taken up in 0.5 ml of methanol, followed by the addition of beta-ray counting fluor. Tritium was assayed by liquid scintillation spectrometry with a 30% counting efficiency. Extraction of tritium-labelled PCP, added to tissue homogenates, was quantitative.

Data were analyzed by Student's *t*-test, with $\alpha = 0.05$.

Results

Phencyclidine concentrations in assayed tissues and blood are given in Table 1. Brain concentrations of PCP were nearly identical between the shocked and nonshocked groups. There were small differences in muscle, blood and heart concentrations of the drug with the nonshocked group generally showing higher average levels than the shocked group, but only differences in blood concentrations (18% higher levels) in the nonshocked group attained statistical significance. Adipose tissue from rats in the footshocked group had 28% higher average levels of PCP ($p<0.05$) than did adipose tissue from rats not subjected to this stressor. Adipose tissue also appeared to have the highest levels of the drug, of those tissues examined. Muscle appeared to have the next

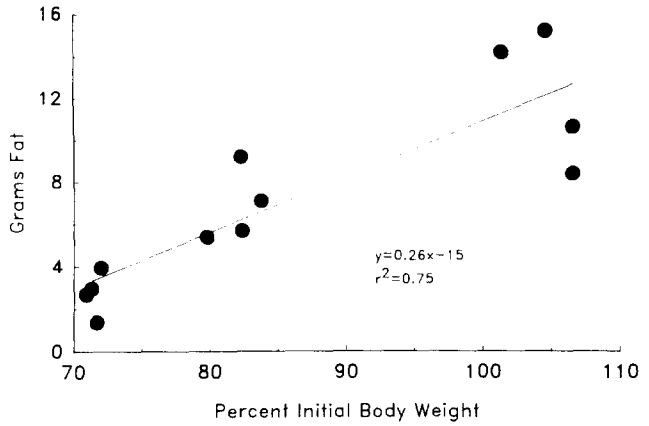

FIG. 1. Fat loss with food deprivation. Sums of perirenal, pericardial and epididymal fat pads were averaged and plotted in relation to the rats' final weights (expressed as a percent of initial weights).

highest levels, one-twentieth to one-fifteenth of those found in fat, and also a great deal of intersubject variation; the other organs and tissues generally had lower levels.

EXPERIMENT 2

Method

In order to gain information regarding the depletion of fat depots during short to intermediate periods of food deprivation of adult rats in the anticipated weight range of those used in further experiments, twelve male Sprague-Dawley rats (368–421 g, Holtzman Co., Madison, WI) were randomly allocated among three equal groups. One group was deprived of food for six consecutive days, a second group was deprived of food for the last three of these six days, and a third group was left with food ad lib. At the end of the six days, all rats were killed, and selected fat pads (perirenal, pericardial and epididymal) were removed and weighed. Sums of these fat pads were averaged and plotted in relation to the rats' final weights (expressed as a percent of initial weights). Statistical assessment of the relations was made by least-squares linear regression.

Results

Free-feeding animals gained about 5% more weight over the six days. After three days of food deprivation, rats lost between 15 and 20% of their initial body weight; by six days of food deprivation, body weights were reduced to about 70% of their initial experimental weights. There was a significant prediction of fat pad mass by body weight change (Fig. 1; coefficient of determination $r^2 = .75$, $p<0.05$). In these latter animals, there was a dramatic reduction in fat pad weights, accompanied by an apparent reduction in the interanimal variability in fat pad weights suggesting a floor effect (i.e., approaching the maximum reduction possible). Much of the mass of the remaining pads in the six-day deprived animals appeared to consist of vasculature and connective tissue, indicating that, should a fat-mobilized drug action become evident, it could be expected to become evident as the body weights of rats (in the 370–420 g range) approach 70% of their starting weights.

EXPERIMENT 3

Method

Forty male Long-Evans rats (405–432 g, Blue Spruce Farms, Altamont, NY) were housed individually in hanging wire mesh cages with food and tap water available ad lib, except as indicated below.

Rats were randomly allocated among three treatment groups in a manner to minimize differences in body weight among groups. Two groups (n = 10, n = 10) received PCP HCl (2 or 4 mg/kg, SC), and one (n = 20) received saline vehicle (0.5 ml/kg) daily for six consecutive days. Immediately after the last injection, food was removed from the cages of all animals. Rats were deprived of food (but with tap water available ad lib) for six consecutive days. During this time body weights were recorded daily. On each of these days, exploratory behavior was assessed during 30-minute sessions. Behavioral sessions were conducted in standard small-animal operant chambers modified to detect spontaneous, unconditioned exploratory or drug-induced motor activities. Solid-state touch circuits were calibrated to detect contacts with an operant lever and with a wall strip, 7.5 cm wide attached to two walls 15 cm above the stainless steel grid floor. Touching either the lever or wall strip completed an electrical circuit to the grid floor with an impedance less than 2.5 megohms. Wall strip contacts were used as an index of rearing (18,21). Lever contacts served as an additional, simultaneous measure of horizontal exploratory behavior. Body weight and behavioral data (square root-transformed wall-strip contacts per 30 minutes or lever-contacts per 30 minutes) were subjected to repeated measures analysis of variance, and planned individual comparisons were made according to the method described by Winer [(23), pp. 170–177] with the Satterthwaite correction [(23), pp. 530–532]. Because the behavioral data were of the nature of counts, square root transformation was selected to stabilize the variances among groups and times [(23), p. 399].

Results

During the injection phase, saline-treated rats gained weight, increasing from 417 to 432 g. Rats injected with the low dose of PCP (2 mg/kg) did not gain weight following the first injection (dropping from 419 to 417 g), but gained weight thereafter at a slower rate than that of saline-injected rats. Animals in the high-dose (4 mg/kg) group lost weight, on average, after the first two daily injections and stabilized thereafter (at about 411 g). Analysis of variance indicated that these differences among groups, with respect to the pattern of weight changes, to be statistically significant, i.e., treatment × days interaction, $F(15,180) = 11.16$, $p<0.05$.

During the food-deprivation phase all groups lost weight in parallel, their relative averages remaining as they were on the last day of injections. By the sixth day of food deprivation, saline control rats were at 80.3% of their preinjection weights. Because they had gained weight during injection days, after six days of food deprivation they were at 77.5% of their free-feeding weights at the beginning of food deprivation. Likewise, the 2 mg/kg and 4 mg/kg dose groups were at 78.2% and 76.7% of their initial weights, but they were at 75.8% and 74.0%, respectively, of the free-feeding saline control group at the end of injections (i.e., of the weight the drug groups would have been had there been no predeprivation weight loss due to drug treatment).

Lever contacts (Fig. 2A) for all groups declined significantly to nearly the same level on the fifth day of food deprivation, $F(5,180) = 25.25$, $p<0.05$. However, on the sixth day, the high-dose group abruptly increased their average number of lever contacts per 30 minutes above that of the saline controls, $F(1,180) = 8.60$, $p<0.05$, suggestive of a surge in circulating PCP in response to a sufficient loss of adipose at this time.

Changes in the other measure of spontaneous motor activity are presented in Fig. 2B. Rearing (strip contacts) during the food-

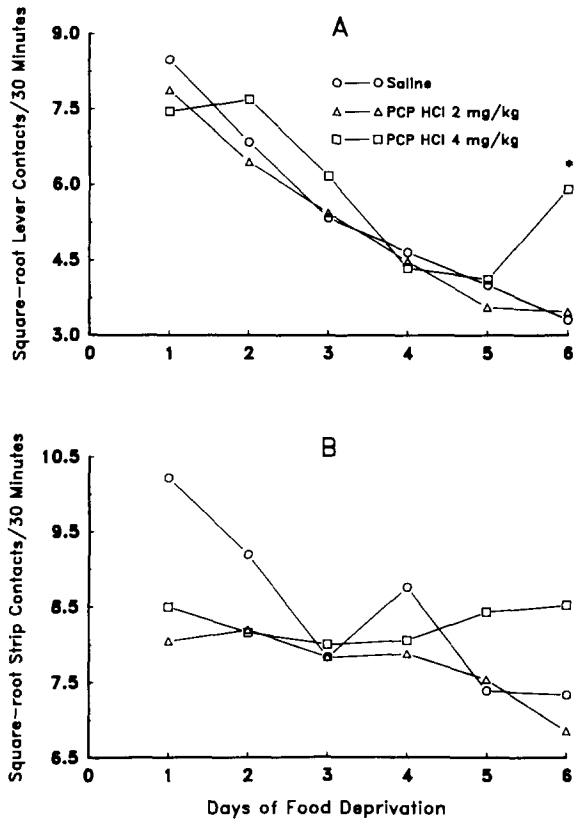

FIG. 2. Horizontal exploratory behavior (A) and rearing (B) during six days of food deprivation in rats injected for six days with saline, or PCP at 2 or 4 mg/kg. Root-transformed contacts to extended lever and to wall strip during the 30-minute unconditioned behavior session during each of six days of food deprivation are plotted for each group. *Indicates statistically significantly different from saline control group ($p<0.05$).

deprivation period (Fig. 2B) varied in a complicated fashion. There appeared to be an interaction between treatment and days of food deprivation, $F(15,180)=1.60$, $p=0.078$. The saline group showed the expected habituation during the repeated exposure to the same chambers, beginning at a relatively high value and declining to nearly half the initial rate of rearing by the sixth day, $F(5,180)=8.46$, $p<0.05$. Rearing activity appeared lower in the two PCP groups during the first session and did not change significantly with time, $F(5,180)<1$ for each treatment group.

EXPERIMENT 4

The previous experiment suggested that, after a period of food-deprivation sufficient to reduce body fat to low levels so as to essentially eliminate it from fat pads, rats with a recent history of repeated PCP injections exhibited altered behavior relative to those with a recent history of saline injections. Because of the ephemeral nature of the effect and the potential clinical relevance of the observation, the reliability of the phenonmenon needed to be determined. Also, a higher dose of PCP was tested in this experiment to examine the dose range in which the observed phenomenon takes place, and an additional, less lipophilic drug, dextroamphetamine, was included to determine the physicochemical and pharmacological range of substances liable to this type of behavioral finding. Finally, because the phencyclidine-treated animals began the food deprivation phase of the previous experiment at a slightly lower weight than saline-injected animals, a pair-fed saline control group was included to determine to what extent initial weight loss contributed to the behavioral response of drug-treated rats to food-deprivation. The pair-fed controls were matched for weight loss of the 4 mg PCP HCl/kg group in order to capitalize on the prior knowledge of drug-induced weight loss with this dose in the previous experiment.

Because rats were somewhat heavier initially in this experiment, food deprivation was carried out for nine days, aiming for terminal weights of about 70% of free-feeding, at which time fat pads are virtually eliminated.

Method

A total of 52 male Long-Evans rats (390–460 g; Blue Spruce Farms) were housed individually in hanging wire mesh cages with food and water available ad lib until experimental manipulations.

Rats were randomly allocated among six treatment groups, in a manner so as to make average body weights among groups as nearly equal as possible. Treatment groups were comprised of between eight and nine subjects each.

Injections of saline or drug solutions were made SC in a volume of 0.5 ml/kg. Injections were made daily for six consecutive days.

The day before the beginning of treatment, food was removed from the hoppers of cages for all rats and replaced with a weighed quantity (50–60 g) of food on the cage floors for all rats. Food consumption was measured on the following day by reweighing the remaining food. For pair-fed rats, the weighed quantity of food placed in the cage each day was limited (to about 15–41 g) to produce weight loss estimated to occur in those rats injected with PCP HCl at the 4 mg/kg dose. Food consumption was measured daily thereafter, throughout the injection phase of the experiment, until the final injection. Immediately after the final injection, food was removed from all rats for the remainder of the experiment. Tap water was available ad lib.

Beginning the following day and continuing for a total of nine consecutive days, rats were placed in small animal operant chambers for 30-minute unconditioned behavior sessions. Lever contacts and contacts made with the wall strip were recorded as described previously. During this time, weights were recorded daily. Behavioral and body weight data were subjected to repeated-measures analysis of variance with the Satterthwaite correction for simple main effects comparisons as described previously, except that, as a preliminary data-reduction step in this experiment, the strip- and lever-contacts were summed prior to transformation and analysis.

Results

As before, free-feeding rats gained weight throughout the injection period, and the 4 mg PCP HCl/kg group lost weight for the first day, then stabilized at a lower level. At the 8 mg/kg level, average body weights were reduced to a greater extent than those at the lower dose and weights continued to decline to a small extent until the fifth injection. The general pattern of stabilization was apparent after the first day in that the greatest drug-related decline in body weight occurred the first day. Average body weight of the pair-fed group fluctuated around that of the 4 mg/kg group. Average body weight for dextroamphetamine-injected animals also declined in a dose-related fashion and appeared to stabilize after repeated injections. At the end of the injection phase, average body weights of the saline-treated/free-feeding group (446 g) and the 4 mg/kg dose group (421 g) in this experiment were somewhat higher than those of the corresponding

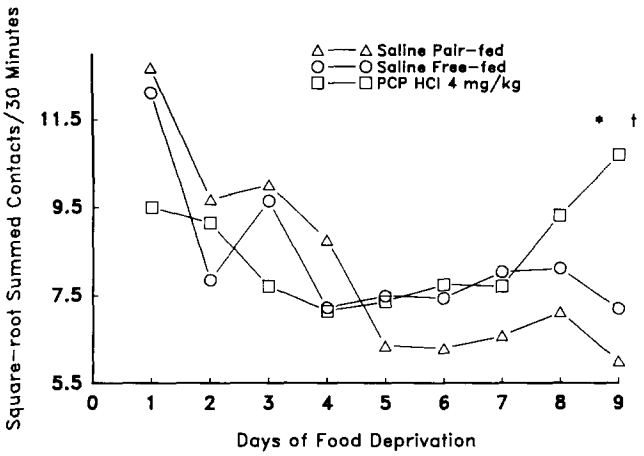

FIG. 3. Unconditioned behaviors during nine days of food deprivation in rats injected for six days with saline or 4 mg PCP/kg. Root-transformed summed lever and wall strip contacts made in daily 30-minute unconditioned behavior sessions for rats given saline and free-fed, for rats given saline and pair-fed to the 4 mg PCP/kg group, and for rats given 4 mg PCP/kg, are plotted for each day of deprivation. *Indicates significantly different from saline/pair-fed control ($p<0.05$). †Indicates significantly different from saline/free-fed control ($p<0.05$).

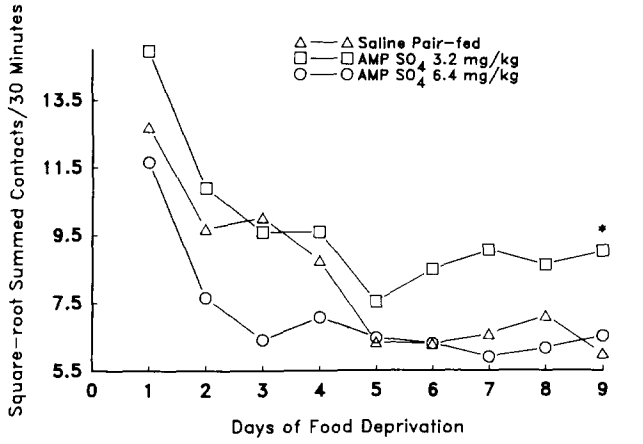

FIG. 4. Unconditioned behaviors by rats during nine days of food deprivation after dextroamphetamine (AMP) or pair-feeding. Root-transformed summed lever and wall strip contacts made in daily 30-minute unconditioned behavior sessions for rats given saline and pair-fed, or dextroamphetamine at 3.4 or 6.8 mg/kg, are plotted. *Indicates significantly different from pair-fed/saline group ($p<0.05$).

groups in Experiment 3.

During the food-deprivation phase of the experiment, body weight reductions followed the pattern seen previously. When body weights in this experiment are expressed as a percent of initial (and as a percent of the saline/free-feeding control group on Day 6 of injection), they were similar to those of Experiment 3 on the corresponding days. On Day 6, the weights of the saline/free-feeding control group were 81.2% of initial (77.9% of last injection day) weights. The 4 mg/kg dose group on that day weighed 76.9% of its initial (preinjection) weight and 73.4% of the saline/free-feeding group's weight on the last day of injection. The 8 mg/kg dose group weighed 76.2% of initial (73.0% of saline/free-feeding group's last injection day) weights. By Day 9 of food deprivation, weight of these groups had fallen to 74.8, 69.9 and 69.7% of initial (71.8, 67.7 and 67.7% of saline/free-feeding group's last injection day) weights, respectively.

Average summed contacts made by saline/pair-fed, saline/free-fed, and 4 mg PCP/kg groups are shown in Fig. 3. Again, there appeared to be an initial difference in spontaneous motor activity after one day of food deprivation [4 mg PCP/kg vs. saline/pair-fed: $F(1,54)=4.76$, $p<0.05$; 4 mg PCP/kg vs. saline/free-fed: $F(1,54)=3.01$, $p<0.10$], although this observation may be more related to a carryover effect of the chronic treatment. As in the last experiment, the saline/free-fed group showed a gradual decline in these unconditioned behaviors. The 4 mg PCP/kg group appeared to begin a gradual increase beginning on Day 7, and continuing to Day 9. The average on Day 9 of food deprivation was significantly different from the saline/pair-fed group, $F(1,54)=10.42$, $p<0.05$, and from the saline/free-feeding group, $F(1,54)=5.45$, $p<0.05$. Contacts made by the 8 mg PCP/kg group were relatively stable and did not differ significantly from either saline control group at any day of food deprivation (data not shown).

Summed contacts for dextroamphetamine groups are shown with those for the saline/pair-fed group in Fig. 4. Dextroamphetamine-treated rats appeared similar to that of saline/pair-fed animals until Day 5, when the average for the 3.2 mg/kg dose group began to rise, attaining significance on Day 9, $F(1,54)=4.05$, $p<0.05$. The high-dose group did not differ significantly from the saline/pair-fed group.

DISCUSSION

Inability of chronic PCP treatment to produce a statistically discernable interaction with footshock (4) may be due to the absence of any increase in brain levels of the drug afforded by footshock stress, because of the analgesic action of PCP, or because adipose tissue binds the drug too tenaciously. Adipose tissue might act initially, shortly after injection, and during the redistribution phase in a sponge-like manner to soak up drug released from other tissues as a result of footshock stress.

The latter possibility was supported by the results reported here: fat had 28% higher concentrations of PCP in the shocked group, indicating that when mobilization occurred, the direction was to fat. It is possible that the shock redirected blood flow to the tissues where the tissue to blood ratio is high. Such a high ratio would permit redistribution of PCP from blood to these tissues faster than to organs where metabolism or excretion of PCP takes place.

The qualitative nature of the redistribution of the more lipophilic PCP appeared to mimic that of other lipophilic compounds in that mobilization was from other tissues to adipose, similar to that reported by Dale *et al.* (6) for highly lipophilic DDT residues in rats after short-term stress (acute intoxication with the pesticide, itself).

Shibata *et al.* (19), in a corroboration of clinical findings, recently demonstrated that electroshock-induced redistribution of the lipophilic antipsychotic drug, haloperidol, occurs in rats after subacute administration. In this case, also, brain levels were unaltered at a time after electrically induced convulsions when plasma levels were altered. They surmised that muscle was the source of the elevated plasma levels of drug, but did not measure fat levels to determine whether adipose tissue is the sink for redistributed haloperidol.

During the course of food deprivation in the latter experiments of the present report, body weights of rats declined to 74–78 percent of the control groups' free-feeding weights. At the end of this time, spontaneous motor activity of the 4 mg PCP/kg dose group increased over that of control groups. Although decreases in

spontaneous motor activity have been recorded with this apparatus after acute administration of PCP, at doses ranging from 2 to 8 mg/kg (5), the increase observed under the present circumstances is not unexpected. First, the dose-response relationship for PCP upon spontaneous locomotor activity is reported to be like that of an inverted "U": increases in measures of spontaneous motor activity are observed after low doses, but decreases are seen as the dose is raised (11). At the low brain concentrations which may have been provided by tissue mobilization in these experiments, decreases in spontaneous motor activity would not have been anticipated. Second, decreases in measures of spontaneous motor activity seen previously (5) after acute administration occurred in rats during their initial exposure to the experimental environment. The direction of PCP's effect upon spontaneous motor activity is influenced by the baseline rate of activity. Increases are normally observed when animals are previously habituated to the environment and the baseline level of activity has fallen to low values (as in the present experiments). Decreases are initially seen, even with low doses, when rats are tested during their first exposure (9) as was the case in the earlier study.

A history of repeated exposure to PCP altered unconditioned behaviors in rats subjected to food deprivation for six days. This period of food deprivation had been shown to reduce or deplete body fat depots in rats of this weight range. That evidence of behavioral toxicity occurred when fat depots were greatly diminished, if not depleted, is consistent with the hypothesis that mobilized PCP can be made more available to sites of action when potential sinks for the drug's redistribution are removed. Since these rats were not down to 70% of their free-feeding weights, it is possible that maximal interactive effects of mobilized drug and stress were not attained.

Altered behavior observed in the third experiment in rats injected with PCP HCl at the 4 mg/kg dose were also observed in Experiment 4. Alterations in behavior were observed in the third experiment at six days of food deprivation, when the rats were at 74% of their free-feeding weights (432 g), as estimated from saline controls. In the slightly heavier rats, behavioral alterations were not observed until nine days of food deprivation had lowered body weights to 67% of their estimated free-feeding weights (446 g). As rats gain weight, body fat increases as a proportion of body weight. With the slightly heavier rats used in the replication, it is understandable that a greater degree of weight loss would need to occur in order to achieve the same leanness obtained in the earlier experiment with six days of deprivation. In any event, in both cases, behavioral alterations were seen only when weights were in the neighborhood of the 71–72% of free-feeding weight range which had been shown earlier (see Fig. 1) to be associated with depletion or dramatically reduced body fat depots. These results are consistent with the hypothesis that interactions can be expected with stored lipophilic compounds when body fat stores are exhausted to a critical level, beneath which they can no longer store drug or act as a sink for mobilized drug. No direct evidence for mobilization was presented here, and mobilization may play a minor or insignificant role in any observed changes. Underlying neural changes which correspond to this degree of deprivation could be responsible for sensitizing the rats to residual low levels of PCP remaining in brain, or to protracted effects of earlier changes induced by repeated administration of the drug. Based upon the results of the pair-fed group, the small degree of weight loss during the injection does not account for the changes in behavior seen after the greater weight loss produced during food deprivation.

Behaviors altered as a result of food deprivation in rats with a history of repeated PCP injection were likewise affected in rats with a history of repeated injections of another lipophilic drug, dextroamphetamine. This result lends credence to the earlier findings in that this phenomenon is replicable and appears to be general among lipophilic drugs with some shared neurochemical and behavioral actions. Dextroamphetamine has previously been shown to interact with a stressor (footshock), the result of which was manifest not only in terms of its distribution (22), but also in terms of behavioral toxicity (4).

The reason for the failure of the high doses of the two drugs to produce similar changes remains unknown, but it may reflect patterns of changes in spontaneous motor activity seen acutely after these drugs. That is, no effect may be seen at very low doses, increases may be observed at low to moderate doses, and decreases may be obtained at the highest doses. The latter effect may result from precedence of competing drug effects, which are incompatible with spontaneous motor activity. Other mechanisms might have prevented the high-dose groups from demonstrating effects similar to those of the lower doses, for example, the amount of PCP or dextroamphetamine cumulated after chronic treatment at the higher doses was not as much because there was relatively less fat, or sufficient tolerance developed after the high doses so that any drug released on Day 9 produced no apparent effect.

Since there were no nondeprived groups in these experiments, a true interaction between food deprivation and a history of repeated drug injection cannot be demonstrated. That is, similar changes in spontaneous motor activity could have occurred in rats with such a history but without food deprivation, for example, as a result of a drug withdrawal syndrome for PCP (2). However, the PCP withdrawal syndrome reported (2) was observed to begin much sooner after the cessation of drug treatment than was observed in the present experiments. In rats, signs of withdrawal begin four hours after termination, subside to a great extent by 48 hours and involve a reduction in spontaneous motor activity, not an increase as was seen here (20). For the present experiments, alterations in spontaneous motor activity appeared not to be a function of time since the last injection. Alterations occurred six days after termination of drug treatment in the third experiment and after eight to nine days in the fourth. In both instances, percent of free-feeding weights (and, by inference, fat pad loss) were approximately equivalent.

It does not appear intuitively logical that adults of normal weight and in good health would fast for six to nine days shortly after using PCP or dextroamphetamine. Therefore, results demonstrated in these studies might be questioned as to their clinical bearing. However, it must be remembered that subacute or chronic abuse of these drugs induce anorexia and behaviors incompatible with normal weight and good health. Moreover, toxic delirium or coma lasting many days has been reported for PCP intoxication. During this period, caloric intake cannot normally be guaranteed. The resulting chronic fat mobilization could easily contribute drug stores to protract or aggravate the adverse drug response, as has been surmised. In addition, chronic drug users, either unmotivated to maintain good diet by lifestyle, or unable to do so by anorexic drug actions possessed by PCP (and amphetamine), might not have that much fat reserve to begin with. As a result, they would not need many days of complete starvation (or of partial starvation associated with poor diet) to achieve the depletion of fat stores associated here with altered behavior responses after repeated drug exposure.

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REFERENCES

1. Allen, R. M.; Young, S. J. Phencyclidine-induced psychosis. Am. J. Psychiatry 135:1081–1084; 1978.
2. Balster, R. L.; Chait, L. D. The behavioral effects of phencyclidine in animals. In: Petersen, R. C.; Stollman, R. C., eds. Phencyclidine (PCP) abuse: An appraisal. Washington, DC: National Institute on Drug Abuse; 1978:53–65.
3. Clark, D. R., Jr.; Prouty, R. M. Experimental feeding of DDE and PCB to female big brown bats (*Eptesicus fuscus*). J. Toxicol. Environ. Health 2:917–928; 1977.
4. Coveney, J. R.; Burklund, K. E. A.; Sparber, S. B. Differential interaction between stressors and chronic amphetamine or phencyclidine upon operant behavior in the rat. Life Sci. 31:1171–1179; 1982.
5. Coveney, J. R.; Sparber, S. B. Phencyclidine retards autoshaping at a dose which does not suppress the required response. Pharmacol. Biochem. Behav. 16:937–942; 1982.
6. Dale, W. E.; Gaines, T. B.; Hayes, W. J., Jr.; Pearce, G. W. Poisoning by DDT: Relation between clinical signs and concentration in rat brain. Science 142:1474–1476; 1963.
7. Eastman, J. W.; Cohen, S. N. Hypertensive crisis and death associated with phencyclidine poisoning. JAMA 231:1270–127; 1975.
8. Geluso, K. N.; Altenbach, J. S.; Wilson, D. E. Bat mortality: Pesticide poisoning and migratory stress. Science 194:184–186; 1976.
9. Greenberg, B. D.; Segal, D. S. Behavioral pharmacology of phencyclidine. Soc. Neurosci. Abstr. 7:261; 1981.
10. James, S. H.; Schnoll, S. H. Phencyclidine: Tissue distribution in the rat. Clin. Toxicol. 9:573–582; 1976.
11. Kesner, R. P.; Hardy, J. D.; Calder, L. D. Phencyclidine and Behavior: I. Sensory-motor function, activity level, taste aversion and water intake. Pharmacol. Biochem. Behav. 15:7–13; 1981.
12. Khajawall, A. M.; Simpson, G. M. Peculiarities of phencyclidine urinary excretion and monitoring. J. Toxicol. Clin. Toxicol. 19: 835–842; 1982/1983.
13. McMahon, B.; Ambre, J.; Ellis, J. Hypertension during recovery from phencyclidine intoxication. Clin. Toxicol. 12:37–40; 1978.
14. Maayani, S.; Gross, S. Partition coefficient, pKa and water solubility of phencyclidine. Fed. Proc. 39:302; 1980.
15. Misra, A. L.; Pontani, R. B.; Bartolomeo, J. Persistence of phencyclidine (PCP) and metabolites in brain and adipose tissue and implications for long-lasting behavioral effects. Res. Commun. Chem. Pathol. Pharmacol. 24:431–445; 1979.
16. Petersen, R. C.; Stillman, R. C. eds. Phencyclidine: An overview. Phencyclidine (PCP abuse: An appraisal. Washington, DC: National Institute on Drug Abuse; 1978:1–17.
17. Rainey, J. M.; Crowder, M. K. Prolonged psychosis attributed to phencyclidine: Report of three cases. Am. J. Psychiatry 132:1076–1078; 1975.
18. Segal, D. S.; Mandell, A. J. Long-term administration of d-amphetamine: Progressive augmentation of motor activity and stereotypy. Pharmacol. Biochem. Behav. 2:249–255; 1974.
19. Shibata, M.; Aoba, A.; Kitani, K.; Kanai, S.; Sato, Y.; Kakita, Y.; Yamaguchi, N.; Hasagawa, K. Biodistribution of haloperidol after electroshock: experimental evidence. Life Sci. 44:749–53; 1989.
20. Spain, J. W.; Klingman, G. I. Continuous intravenous infusion of phencyclidine in unrestrained rats results in the rapid induction of tolerance and physical dependence. J. Pharmacol. Exp. Ther. 234: 415–424; 1985.
21. Sparber, S. B. Use of learned behavior in testing for neurotoxity. In: Gryder, R. M.; Frankos, V. H., eds. Effects of foods and drugs on the development and function of the nervous system: Methods for predicting toxicity. Proceedings of the Fifth FDA Science Symposium. Rockville, MD: U.S. Department of Health and Human Services; 1980:49–61.
22. Sparber, S. B.; Nagasawa, S.; Burklund, K. E. A. A mobilizable pool of d-amphetamine in adipose after daily administration to rats. Res. Commun. Chem. Pathol. Pharmacol. 18:423–431; 1977.
23. Winer, B. J. Statistical principles in experimental design, 2nd ed. New York: McGraw-Hill; 1971.
24. Yago, K. B.; Pitts, F. N., Jr.; Burgoyne, R. W.; Aniline, O.; Yago, L. S.; Pitts, A. F. The urban epidemic of phencyclidine (PCP) use: Clinical and laboratory evidence from a public psychiatric hospital emergency service. J. Clin. Psychiatry 42:193–196; 1981.