Psychopharmacological Studies of Some 1-(Chlorophenyl)-2-aminopropanes I

Effects on Appetitive-Controlled Behavior

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A series of 1-phenyl-2-aminopropanes with chloro substitutions on the 2,4, 3,4, and 4 positions on the phenyl ring and their optical isomers were compared with *dl*- and *d*-amphetamine and methamphetamine on a fixed-ratio (FR) procedure. The subjects were rats that had been trained to press a lever 30 times for 0.25-ml. reinforcement of sweetened condensed milk. Each session consisted of 40 reinforcements. With unsubstituted amphetamines, rats showed a slight reduction in response rates with long pauses following reinforcement. This effect was similar to the effect seen with onset of normal satiation. After chloro compounds, the rats produced irregular and slowed response rates with frequent short pauses between and following reinforcement. The minimum effective doses of all compounds used in this study ranged from 1–3.2 mg./Kg. Except for *d*-1-(4-chlorophenyl)-2-aminopropane, the *l*-isomers of chloro-substituted compounds were more active suppressants of FR behavior than the *dl* or *d* compounds.

COMPOUNDS THAT stimulate the central nervous system and also have appetite-suppressing activity, such as amphetamine and methamphetamine, have be enreported to alter performance of trained experimental animals working for food reinforcement on fixed-ratio (FR) schedules (1–3). Specifically, *dl*-, *d*-, and *l*-amphetamine and methamphetamine (4) changed FR behavior in rats by slowing the response rate, increasing the frequency and duration of pauses between and following reinforcements, and inducing temporary cessation of responding following drug administration. Some chloro substituted 1-phenyl-2-aminopropanes in a series of phenylalkylamines studied for central-nervous-system activity by Rathbun (5) exhibited appetite suppression with minimum stimulation. Rathbun estimated the appetite suppressing ED_{50}'s of these compounds in carefully controlled studies on the food intake of hungry rats under the influence of the drugs. Four of these chloro-substituted compounds and their optical isomers were selected for comparison with *dl*- and *d*-amphetamine and methamphetamine on an FR schedule for food reinforcement. The changes in the appetitive controlled behavior of rats after the administration of the drugs is described.

EXPERIMENTAL

Materials.—The compounds and doses used in this investigation as well as the oral ED_{50} doses (5) are listed in Table I. The compounds were dissolved in distilled water and administered subcutaneously.

Method.—Male rats of a Wistar-derived strain, initially weighing 400–450 Gm., were used in this study. With controlled food deprivation their body weights were gradually reduced to 70% of the initial weights. They were trained on an FR-30 schedule in which 30 lever pressing responses were reinforced with 0.25 ml. of a mixture of sweetened condensed milk, water, and homogenized multiple vitamins,[1] in proportions of 1:1:0.01.

The apparatus and procedure have been described in detail elsewhere (4). Briefly, two experimental cages, each 9 × 9 × 7.5 in. were used. Each contained a lever, a modified telephone-type switch (6), a small pilot light over the lever, and a motor-driven dipper for delivery of the reinforcement. The cages were contained in light-proof, sound resistant, ventilated boxes isolated from the control equipment. The procedure, controlled by appropriate electrical relay circuits, was designed to give the rat a reinforcement upon completion of 30 responses. Illumination inside the cage was provided during a session by the small light over the lever. During the 8-second interval that the reinforcement was available to the rat, the light over the lever was extinguished and another small light illuminated the dipper cup.

Each rat was given daily sessions of 40 reinforcements to maintain stable performance. For sessions when drugs were tested, a rat was given ten reinforcements before drug administration as a "warm-up" and to establish that he was working normally that day.

In a previous study (4) the clinically used amphetamines and methamphetamines had been shown to produce maximum suppression of FR responding within 20 to 30 minutes after administration. Therefore, the animal was placed in a small observation cage for 30 minutes following drug administration to allow the drug to be absorbed. The animal was returned to the experimental cage and permitted to work for 40 reinforcements. The compounds were given once to each rat at each dose.

Data.—The data were collected on electrical impulse counters, running-time meters, and cumula-

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[1] Marketed as Homicebrin by Eli Lilly and Co.

TABLE I.—DRUGS AND DOSES USED IN FR SUPPRESSION STUDIES

Drug	Doses, mg./Kg.			Oral ED_{50}, mg./Kg.
dl-Amphetamine Sulfate	1.0	2.0	3.2	...
d-Amphetamine Sulfate	1.0	2.0	3.2	2.65
d-Methamphetamine HCl	1.0	2.0	3.2	...
dl-1-(4-Chlorophenyl)-2-aminopropane HCl	3.2	5.0	8.0	2.05
d-1-(4-Chlorophenyl)-2-aminopropane HCl	1.0	2.0	3.2	1.33
l-1-(4-Chlorophenyl)-2-aminopropane HCl	2.0	3.2	5.0	3.35
dl-1-(2,4-Dichlorophenyl)-2-aminopropane HCl	3.2	5.0	8.0	4.11
d-1-(2,4-Dichlorophenyl)-2-aminopropane HCl	3.2	5.0	8.0	10.00
l-1-(2,4-Dichlorophenyl)-2-aminopropane HCl	2.0	3.2	5.0	2.57
dl-1-(3,4-Dichlorophenyl)-2-aminopropane HCl	2.0	5.0	8.0	2.86
d-1-(3,4-Dichlorophenyl)-2-aminopropane HCl	3.2	5.0	8.0	4.10
l-1-(3,4-Dichlorophenyl)-2-aminopropane HCl	2.0	3.2	5.0	1.81
dl-1-(3,4-Dichlorophenyl)-2-methylaminopropane HCl	3.2	5.0	8.0	4.21
d-1-(3,4-Dichlorophenyl)-2-methylaminopropane HCl	3.2	5.0	8.0	5.69
l-1-(3,4-Dichlorophenyl)-2-methylaminopropane HCl	3.2	5.0	8.0	2.65

tive recorders. Two variables of this appetitive controlled FR behavior were studied: the running response rates, *i.e.*, the sustained constant rate (7), and the total time used to complete each session. The response rates of the individual rats were determined as responses per second. Because this investigation was concerned with the running response rate in contrast to the overall response rate (4, 7, 8), pauses in responding of more than 5 seconds were not included in the response-rate calculations. Since each rat acted as its own control, *i.e.*, normal behavior *versus* behavior with drug, the data from each rat were calculated as ratios of response rates with drug to response rates without drug. Also, ratios of total time to complete a session with drug to mean total time to complete a session without drug were calculated.

The cumulative recorders produced direct recordings of responses related to time. Thus, a visual record of behavior was available at any point of an experimental session. In these records vertical movement of the pen is related to the number of

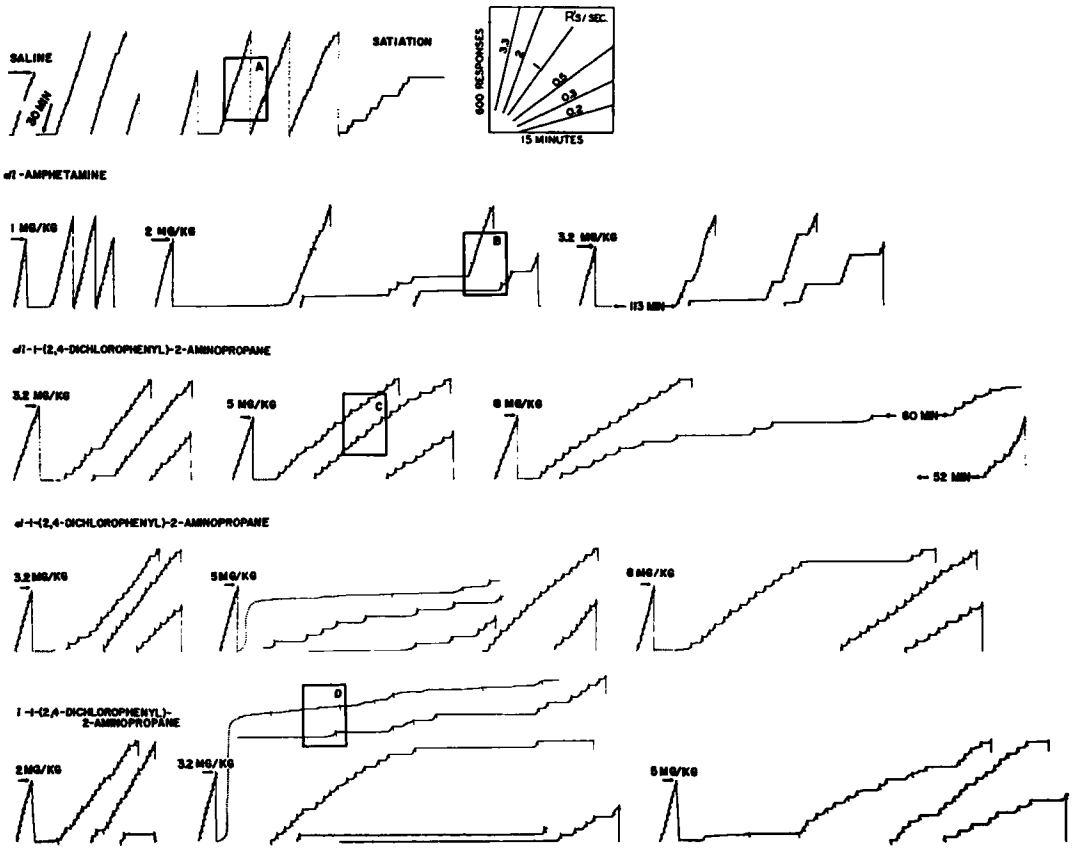

Fig. 1.—Cumulative records of responses of Rat 84 showing patterns of normal behavior and behavior under the influence of the drugs used in this study.

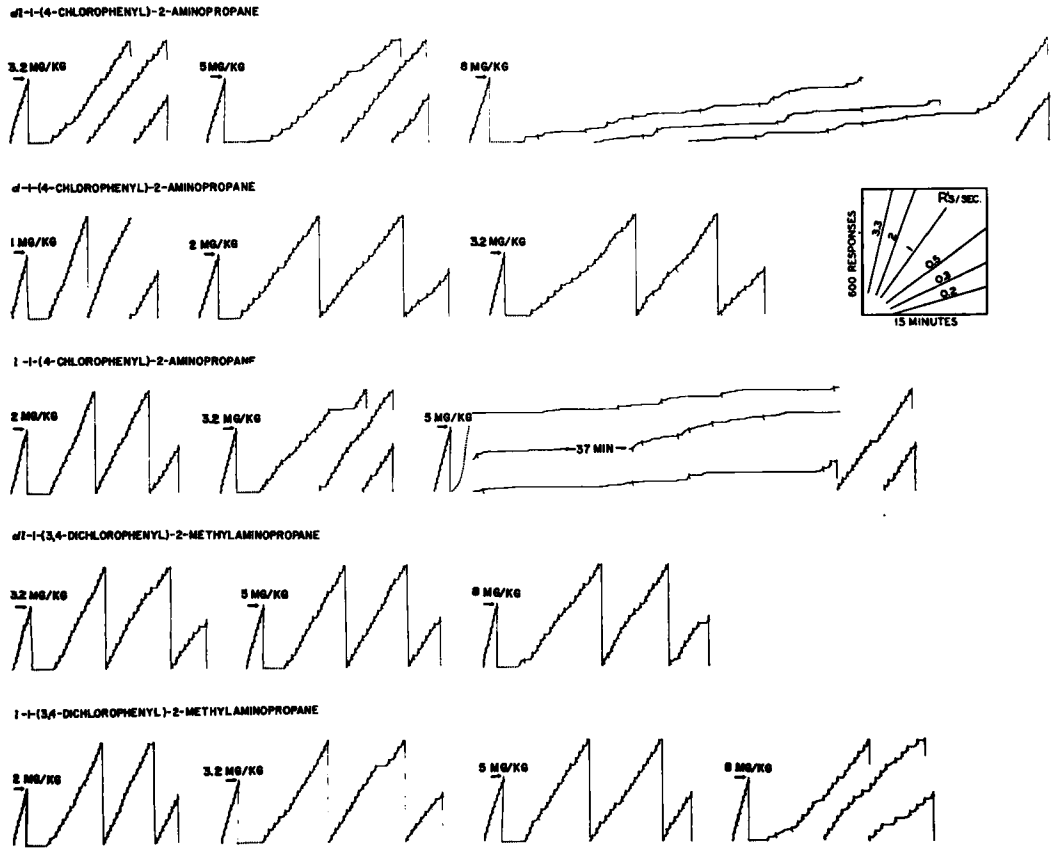

Fig. 2.—Cumulative records of responses of Rat 84 showing patterns of behavior under the influence of drugs used in this study.

responses and horizontal paper movement indicates time. The angle of the line or any segment of the ine is indicative of the individual's response rates. Short vertical marks on the response line show when reinforcements occurred.

RESULTS

The data from this behavioral study of chloro-substituted 1-phenyl-2-aminopropanes, as recorded on the cumulative response recorders, demonstrated that these compounds influence appetitive-controlled FR behavior differently than do *dl*- and *d*-amphetamine and methamphetamine. In general, the deschloro drugs influenced the behavior by causing only a slight depression of response rates with a marked increase in the frequency and duration of the post-reinforcement pauses. At the higher doses some rats also failed to resume responding immediately upon being returned to the cage after the 30-minute period following drug administration.

Figures 1 and 2 present typical cumulative response curves of one rat showing a session with saline; a session during which an unlimited number of reinforcements was made available to the rat (satiation); the effects of *dl*-amphetamine; and the effects of eight of the chloro-substituted compounds. With *dl*-amphetamine there was little variation in the response rate when the rat was working. This rate appeared constant from dose to dose. Shown

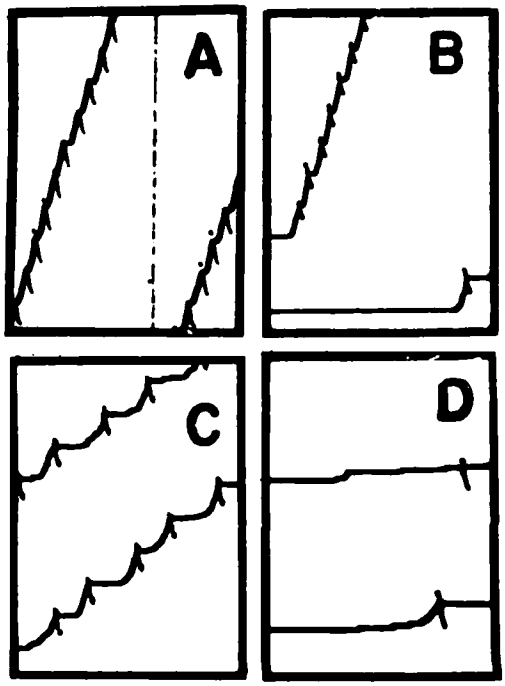

Fig. 3.—Enlarged segments of cumulative record from Fig. 1.

TABLE II.—MEAN RESPONSE-RATE RATIOS

Compound			Rats, No.	Low Dose	Middle Dose	High Dose	Slope, ± S.E.	P
Phenyl Substituent	Amino Substituent	Isomer						
H	H	*dl*	7	0.73	0.64	0.68	−0.025 ± 0.061	0.70
H	H	*d*	7	0.62	0.66	0.66	+0.016 ± 0.039	0.70
H	CH_3	*d*	7	0.84	0.81	0.68	−0.081 ± 0.082	0.45
4-Cl	H	*dl*	5	0.76	0.50	0.32	−0.218 ± 0.079	0.05
4-Cl	H	*d*	5	0.91	0.39	0.38	−0.264 ± 0.037	<0.01
4-Cl	H	*l*	5	0.88	0.56	0.42	−0.229 ± 0.028	<0.01
2,4-diCl	H	*dl*	5	0.74	0.54	0.37	−0.182 ± 0.050	<0.02
2,4-diCl	H	*d*	7	0.68	0.51	0.44	−0.119 ± 0.029	<0.01
2,4-diCl	H	*l*	7	1.03	0.38	0.35	−0.338 ± 0.049	<0.001
3,4-diCl	H	*dl*	5	0.93	0.65	0.55	−0.187 ± 0.081	<0.10
3,4-diCl	H	*d*	5	0.84	0.63	0.74	−0.049 ± 0.035	0.25
3,4-diCl	H	*l*	5	1.03	0.76	0.71	−0.163 ± 0.037	<0.02
3,4-diCl	CH_3	*dl*	6	0.88	0.83	0.68	−0.098 ± 0.024	0 01
3,4-diCl	CH_3	*d*	6	0.75	1.05	0.63	−0.155 ± 0.077	0.10
3,4-diCl	CH_3	*l*	7	0.86	0.68	0.56	−0.151 ± 0.048	<0.05

here are the long pauses that occasionally followed a reinforcement and protracted periods of time when no response occurred following return to the experimental cage.

With the chloro-substituted compounds, the rat's response rate appeared irregularly depressed. Pauses between and after reinforcement became more frequent and were shorter than those with the unsubstituted amphetamines. These phenomena present an overall "scalloped" effect on the cumulative records. The depression of rate appeared more pronounced with the larger doses. A detailed comparison of segments of the records showing the differences between normal running rates, rates with *dl*-amphetamine, and rates with *dl*-1-(2,4-dichlorophenyl)-2-aminopropane is presented in Fig. 3.

The effects of the compounds on the response-rate ratios are shown in Table II. The average response-rate ratios of individual rats were computed for each dose of each drug. The slope values are relative, based on three equally spaced doses and do not represent numerically the dose-response slopes (*cf.*, Table I for actual doses used). They serve the purpose of indicating whether or not there was a change in the rate ratio and whether or not the change was significant. With *dl*- and *d*-amphetamine and methamphetamine, the rats produced lower than normal rates but showed little change from dose to dose. The chloro-substituted compounds, with the exception of *d*-1-(3,4-dichlorophenyl)-2-aminopropane, caused the rats to work at rates that decreased significantly as the doses were increased.

As shown in Table III, the length of time to complete the 40-reinforcement sessions with all of the drugs studied was longer than normal. The means and standard errors of the time ratios are presented for each dose of each drug. In general, the mean time to complete a session increased as the dose was increased. The increases appeared greatest with *dl*- and *d*-amphetamine, methamphetamine, the 4-chlorophenyl, and the 2,4-dichlorophenyl compounds. The variability among animals was high as evidenced by the rather large standard error terms.

DISCUSSION

In animals trained on FR procedures for food reinforcement the response rate normally will remain unchanged once it is established. The development of post-reinforcement pauses, however, is dependent upon several controllable variables (7, 8). If the size of the FR is increased, *e.g.*, from 30 to 100 responses, post-reinforcement pauses become pronounced and in some instances the rat stops responding entirely. If the size of the reinforcement is decreased, the post-reinforcement pauses become more frequent and of greater duration. The state of

TABLE III.—MEAN TOTAL TIME RATIOS

Compound			Low Dose	Middle Dose	High Dose
Phenyl Substituent	Amino Substituent	Isomer	Mean Ratio, ± S.E.	Mean Ratio, ± S.E.	Mean Ratio, ± S.E.
H	H	*dl*	1.74 ± 0.33	3.56 ± 0.80	6.83 ± 2.03
H	H	*d*	2.27 ± 0.37	10.32 ± 2.56	20.05 ± 4.64
H	CH_3	*d*	4.46 ± 1.82	9.33 ± 2.89	16.54 ± 4.76
4-Cl	H	*dl*	1.64 ± 0.26	8.72 ± 3.07	10.28 ± 3.34
4-Cl	H	*d*	1.20 ± 0.16	6.04 ± 2.35	11.30 ± 6.04
4-Cl	H	*l*	1.24 ± 0.13	2.17 ± 0.16	8.40 ± 2.66
2,4-diCl	H	*dl*	1.77 ± 0.10	2.79 ± 0.10	17.76 ± 4.10
2,4-diCl	H	*d*	4.09 ± 1.99	6.59 ± 2.30	12.24 ± 5.31
2,4-diCl	H	*l*	1.27 ± 0.18	11.59 ± 4.39	13.14 ± 4.20
3,4-diCl	H	*dl*	1.39 ± 0.22	2.28 ± 0.71	3.65 ± 1.30
3,4-diCl	H	*d*	1.81 ± 0.75	3.76 ± 1.99	2.25 ± 0.90
3,4-diCl	H	*l*	1.03 ± 0.88	3.07 ± 1.63	3.63 ± 2.19
3,4-diCl	CH_3	*dl*	1.42 ± 0.30	1.52 ± 0.38	2.74 ± 1.00
3,4-diCl	CH_3	*d*	1.68 ± 0.45	1.04 ± 0.12	1.77 ± 0.21
3,4-diCl	CH_3	*l*	1.35 ± 0.21	1.94 ± 0.43	3.89 ± 1.20

deprivation and/or satiation of the animal influences the character of the post-reinforcement pause, *i.e.*, the more satiated the animal the longer the pauses. Generally, the response rates remain relatively unchanged despite the length of the post-reinforcement pause. The phenomenon of pausing has been termed "fixed-ratio strain" (4). Rats with stable FR behavior showing little or no ratio-strain responded with frequent and sometimes very long pauses when under the influence of amphetamine compounds. The overall effect of the amphetamines in this situation was considered to be a lowering of the threshold for fixed-ratio straining that resembled, in many respects, a premature onset of satiation.

The chloro-substituted compounds used in this study depressed the rats' FR behavior by causing an irregular decline in response rate that appeared different from the depression seen with the unsubstituted amphetamines as shown in Fig. 3. The cumulative record data indicate numerous relatively short pauses occurred with the slowed response rates. Even segments of the cumulative records that appear as long pauses, on closer inspection, reveal the presence of occasional short bursts of 2–3 or more lever presses. This random, intermittent responding is illustrated in part *D* of Fig. 3. At the doses used, the 4-chlorophenyl and 2,4-dichlorophenyl compounds produced the most marked depression of the FR behavior in regard to response rate and prolongation of total time to complete sessions.

Except for the 1-(4-chlorophenyl)-2-aminopropane compounds, the *l* isomers of the chloro compounds appeared more potent than the *dl* and *d* members. This observation suggests that the presence of dichloro substitutions on the phenyl ring reverses the relative potency of the isomers seen with *d*- and *l*-amphetamine as reported in a previous study (4). Monochloro substitution on the 4 position seemingly failed to cause this reversal since the *d*-1-(4-chlorophenyl) compound appeared more potent than either of the *dl* or *l* compounds.

ADDENDUM

None of the new compounds tested could be considered a clinically useful drug.[2] In one double blind study with 16 moderately obese subjects, 20 and 30 mg. of *l*-1-(2,4-dichlorophenyl)-2-aminopropane was compared with placebo and a marketed anorectic drug. When the patients received the chlorinated compound they reported poor appetite control. Several patients mentioned a sense of agitation associated with inability to work effectively; others complained of nausea, nightmares, irritability, and lack of tolerance for the usual difficulties at work and at home. In general, both the patient and his family disapproved of the experimental compound. The placebo and known drug were well tolerated and the active known compound could be shown to cause significant weight loss in a 1-week period. No patient preferred the chlorinated compound but 15 preferred the active drug to placebo. It would appear that the pseudo-satiation-type of suppression of the FR preformance seen after the rats were given amphetamine is consistent with suppression of appetite clinically. The decrease of rate with the scalloped appearance resulting from frequent short pauses seems to indicate a different, and probably undesirable, type of clinical activity.

REFERENCES

(1) Morse, W. H., and Herrnstein, R. J., *Ann. N. Y. Acad. Sci.*, **65**, 303(1956).
(2) Dews, P. B., *ibid.*, **65**, 268(1956).
(3) Kelleher, R. T., and Cook, L., *J. Exp. Anal. Behav.*, **2**, 267(1959); (Abstract).
(4) Owen, J. E., Jr., *ibid.*, **4**, 293(1960).
(5) Rathbun, R. C., unpublished data.
(6) Verhave, T., *J. Exp. Anal. Behav.*, **1**, 220(1958).
(7) Ferster, C. B., and Skinner, B. F., "Schedules of Reinforcement," 1st ed., Appleton-Century-Crofts, New York, N. Y., 1957, pp. 39–77.
(8) Sidman, M., and Stebbins, W. C., *J. Comp. Physiol. Psychol.*, **7**, 114(1954).

[2] A preliminary trial of some of these appetite suppressants was conducted by Dr. S. M. Chernish of the Department of Clinical Research, Eli Lilly and Co.