Comparison of Anticonvulsant and Psychopharmacologic Drugs

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Four anticonvulsant drugs were compared with seven psychopharmacologic agents by a battery of tests designed to detect possible tranquilizing activity. The drugs tested were found to have only one property in common, the ability to prolong hexobarbital sleep time. Five psychopharmacologic agents, chlorpromazine, promazine, trifluprom azine, reserpine, and hydroxyzine, blocked conditioned avoidance responses and reduced amphetamine toxicity in aggregated mice, whereas the two sedative anticonvulsants, phenobarbital and trimethadione, and two psychopharmacologic agents, meprobamate and phenaglycodol, were inactive by these tests. The two nonsedative anticonvulsants, diphenylhydantoin and phenacemide, markedly reduced amphetamine toxicity in aggregated mice, which suggests that these two anticonvulsant drugs may have mild tranquilizing properties.

IN PREVIOUS reports from our laboratories, the anticonvulsant activity of several psychopharmacologic drugs with tranquilizing properties were compared with those of conventional antiepileptic agents (1, 2). In the present report, the tranquilizing activities of four antiepileptic drugs are compared with those of some clinically useful psychopharmacologic agents. This study was prompted by the fact that some conventional anticonvulsant drugs not only control epileptic seizures, but also may exert a salutary effect on the behavioral disturbances which often accompany convulsant disorders (3). Furthermore, anticonvulsant drugs have been used successfully in the treatment of nonepileptic, mentally ill patients (3–7). These observations suggest that anticonvulsant drugs may possess some central properties which are common to those of the psychopharmacologic agents. The experiments described in this communication were designed to test this hypothesis.

METHODS

Adult male albino mice (CF No. 1 strain) obtained from the Carworth Farms were used as experimental animals. They were allowed free access to food and water except during the experimental test period. All drugs were given orally in aqueous solution, except that phenacemide, trimethadione, meprobamate, and phenaglycodol were given as suspensions in 6% acacia solution. The mean neurotoxic dose (TD_{50}) and the time of peak activity were determined for each drug as described elsewhere (2). Equitoxic doses (multiples of the TD_{50}) were given to groups of animals as described below, and the animals subjected to the particular test at the time of peak drug activity. The various drugs examined, their times of peak activity, and their TD_{50}s are listed in Table I.

TABLE I.—TIME OF PEAK EFFECT AND NEUROTOXIC DOSES (TD_{50}) OF SOME ANTICONVULSANT AND PSYCHOPHARMACOLOGIC DRUGS IN MICE

Drug	Time of Peak Drug Effect, min.	Neurotoxic Dose[a] (TD_{50})
Diphenylhydantoin sodium (Dilantin sodium)	180	84 (74–95)
Phenacemide (Phenurone)	45	660 (545–799)
Phenobarbital sodium	180	70 (58–84)
Trimethadione (Tridione)	120	1150 (1055–1253)
Meprobamate (Equanil; Miltown)	30	228 (196–262)
Phenaglycodol (Ultran)	60	170 (119–241)
Hydroxyzine hydrochloride (Atarax hydrochloride)	30	490 (434–553)
Reserpine	240	16.5 (11.6–23.4)
Chlorpromazine hydrochloride (Thorazine hydrochloride)	90	15.7 (10.6–23.2)
Promazine hydrochloride (Sparine hydrochloride)	60	23.5 (15.0–36.9)
Trifluprom azine hydrochloride (Vesprin)	90	7.8 (5.3–11.5)

[a] Values expressed in mg./Kg. with 95% fiducial limits in parentheses.

Hexobarbital Potentiation.—The drugs were administered to groups of 8 mice each. At the time of peak drug effect, hexobarbital sodium (100 mg./Kg.) was injected intravenously. The sleeping time, in minutes, was measured from the end of the injection until the animal regained its righting reflex. The per cent increase in the sleep time of the drug-treated group over that of the corresponding control group run concurrently was calculated and the results were analyzed by the *t* test.

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Conditioned Response Test.—A shuttle box (20 in. by 4 in. by 11 in.) was divided into two compartments (one black and one white) of equal length and width, by a barrier formed by elevating the smooth white floor of the white compartment 2 in. The black compartment had walls 9 in. high and a grid floor composed of stainless steel rods (1/8 in. diameter, placed 1/4 in. apart) through which an electric shock (60-cycle a.c.; 50 v., delivered through a grid scrambler) could be applied to the feet of the mouse. An electric buzzer was placed beneath the grid floor of the black compartment and a 25-w. light bulb with a reflector was placed above it. Mice were placed individually in the black compartment. After 15-seconds exposure to the environment, the buzzer and light were activated for 10 seconds. An animal could avoid the electric shock that would follow by escaping to the white compartment (conditioned response, CR). Each mouse was further conditioned until it would escape to the white compartment when placed in the box (secondary conditioned response, SCR). Three end points were used to determine drug activity: abolition of (*a*) the SCR (exposure to 15 seconds of the environment), (*b*) the CR (exposure to 15 seconds of environment alone, and 10 seconds of buzzer and light), and (*c*) the unconditioned response (UR, exposure to 15 seconds of environment alone, 10 seconds of buzzer and light, and, finally, 5 seconds of shock associated with the buzzer and light). The response of each mouse was graded according to an arbitrary point system: a mouse exhibiting a SCR was awarded 3 points; a CR, 2 points; a UR, 1 point; nonresponders to 5 seconds of shock were assigned 0 points. Drug activity was determined in groups of 8 mice. Each mouse was given a block of 4 consecutive trials at 1-minute intervals (a fully conditioned mouse would score 12 points in this control response) and then administered the drug to be tested. At selected times after drug administration, the mice were given another block of trials and the responses recorded. A control group was always tested concurrently with the drug-treated groups to insure that the SCR and CR were not extinguished spontaneously, despite the fact that preliminary experiments indicated that there were no significant differences in the total scores of a group of 8 control mice given successive blocks of trials over a 4-hour period. The data were statistically analyzed by the Walsh test (8).

Amphetamine Toxicity Tests.—The drugs were initially tested by the 3-mice-per-cage test described by Lasagna and McCann (9), except that an intraperitoneal dose of 97.5 mg./Kg. of amphetamine, equivalent to 3 times the LD_{50} determined in aggregated mice, was employed as previously reported (10). This test was so severe that even reserpine, a drug known to have tranquilizing properties, was ineffective in nontoxic doses. Therefore, amphetamine toxicity was also studied in aggregated mice by a modification of the 10-mice-per-cage test described by Burn and Hobbs (11). Fifty mice were randomly divided into 5 groups of 10 mice each. A different drug at a selected dose level was administered to each of four groups, and the requisite volume of vehicle was given to the fifth group, which served as a control. Thirty minutes before the time of peak effect for each drug (15 minutes for drugs with a time of peak effect of 30 minutes), each animal was given 25 mg./Kg. of amphetamine sulfate intraperitoneally. Each group of mice was then placed in a clear plastic cage (27 cm. by 17 cm. by 12 cm.) with a screen bottom. Twenty-four hours later the number of dead mice in each box was recorded. The above procedure was repeated until 20 mice (2 groups of 10 mice each) had received the selected dose of each drug. The data were statistically analyzed by the Chi-square method, and drug activity was expressed as a ratio (% of deaths in control group/% of deaths in drug-treated group).

RESULTS

The effects of some anticonvulsant and psychopharmacologic drugs on hexobarbital sleep time in mice are summarized in Table II. As indicated in the table, all of the agents significantly increased hexobarbital sleep time. The increase in sleep time ranged from 40% for meprobamate to 240% for diphenylhydantoin.

TABLE II.—EFFECTS OF SOME ANTICONVULSANT AND PSYCHOPHARMACOLOGIC DRUGS ON HEXOBARBITAL SLEEP TIME IN MICE

Drug	Dose mg./Kg.	Dose Multiples TD_{50}	Increase in Sleep Time,[a] %
Diphenylhydantoin	42	1/2	240
Phenacemide	330	1/2	177
Phenobarbital	35	1/2	53
Trimethadione	575	1/2	81
Meprobamate	114	1/2	40
Phenaglycodol	85	1/2	207
Hydroxyzine	122	1/4	117
Reserpine	8.2	1/2	55
Chlorpromazine	7.8	1/2	184
Promazine	11.7	1/2	71
Trifluprom azine	3.9	1/2	168

[a] All values in this column are significantly different from controls ($P < 0.05$).

The effects of these same drugs on conditioned responses in mice are shown in Table III; these effects are expressed as activity ratios (total points in predrug blocks of trials/total points in postdrug blocks of trials). Ideally, a drug that blocks only the secondary conditioned response (SCR) in all animals has an activity ratio of 1.5 (12/8), whereas a drug that blocks the conditioned response (CR) in all animals has a ratio of 3.0 (12/4). Ratios above 3.0 indicate that the unconditioned response (UR) is also blocked in some animals. The four anticonvulsant drugs (diphenylhydantoin, phenacemide, phenobarbital, and trimethadione) did not significantly suppress either the SCR or the CR even after doses up to the TD_{50}. Meprobamate and phenaglycodol, in doses equivalent to the TD_{50}, partially blocked the SCR, although only the activity ratio of meprobamate, 1.28, was significant. Hydroxyzine at 1/2 the TD_{50} blocked the SCR in all animals, whereas 8.2 mg./Kg. of reserpine (1/2 TD_{50}) abolished the CR in all animals and blocked the UR in some animals, as indicated by an activity ratio of 3.39. Chlorpromazine exhibited greater activity as the dose level was increased. At dose levels of 3.9 and 7.8 mg./Kg., the SCR and the CR were successively

TABLE III.—EFFECTS OF SOME ANTICONVULSANT AND PSYCHOPHARMACOLOGIC DRUGS ON CONDITIONED RESPONSES IN MICE

Drug	Dose mg./Kg.	Dose Multiples TD_{50}	Activity Ratio[a]
Diphenylhydantoin	42	1/2	1.18
	84	1	1.21
Phenacemide	660	1	1.02
	1,320	2	1.02
Phenobarbital	70	1	1.08
	140	2	1.15
Trimethadione	575	1/2	0.96
	1,150	1	1.03
Meprobamate	114	1/2	0.99
	228	1	1.28[b]
Phenaglycodol	170	1	1.22
Hydroxyzine	122	1/4	1.32[b]
	245	1/2	1.64[b]
Reserpine	8.2	1/2	3.39[b]
Chlorpromazine	3.9	1/4	1.35[b]
	7.8	1/2	1.96[b]
	15.7	1	3.10[b]
Promazine	11.7	1/2	1.26[b]
Triflupromazine	3.9	1/2	2.58[b]

[a] Total points in predrug block of trials/total points in postdrug block of trials. [b] Significantly different from controls ($P < 0.05$).

TABLE IV.—EFFECTS OF SOME ANTICONVULSANT AND PSYCHOPHARMACOLOGIC DRUGS ON AMPHETAMINE TOXICITY IN AGGREGATED MICE

Drug	Aggregation Toxicity Test: 3 per Cage (ED_{50})[a]	10 per Cage: Dose mg./Kg.	Multiples (TD_{50})	Ratio[b]
Diphenylhydantoin	>200	42	1/2	7.0[c]
		84	1	4.7[c]
Phenacemide	>1,000	330	1/2	7.0[c]
		660	1	7.0[c]
Phenobarbital	>75	70	1	1.2
Trimethadione	>1,000	575	1/2	0.8
		1,150	1	1.2
Meprobamate	>1,000	228	1	0.8
Phenaglycodol	>1,000	170	1	0.9
Hydroxyzine	>400	175	1/3	2.8[c]
Reserpine	>80	2.0	1/8	4.7[c]
Chlorpromazine	2.0 (1.3–3.2)	2.0	1/8	2.3[c]
Promazine	8.0 (5.0–12.8)	8.0	1/3	2.8[c]
Triflupromazine	3.8 (2.4–6.1)	3.8	1/2	3.5[c]

[a] Values expressed in mg./Kg. with 95% fiducial limits in parentheses. [b] % of deaths in control group/% of deaths in drug-treated group. [c] Significantly different from controls ($P < 0.05$).

suppressed (activity ratios, 1.35 and 1.96, respectively). At the TD_{50} (15.7 mg./Kg.), the drug absolished the CR and occasionally blocked the UR, as indicated by a ratio of 3.10. Trifluprom azine was more active and promazine was less active than chlorpromazine at dose levels equivalent to 1/2 the TD_{50}; the ratios were 2.58, 1.26, and 1.96, respectively.

The effects of some anticonvulsant and psychopharmacologic drugs on amphetamine toxicity in aggregated mice are summarized in Table IV. It is evident that the drugs were less effective against amphetamine toxicity in the 3-mice-per-cage test than in the 10-mice-per-cage test. Except for the three phenothiazine derivatives, all the drugs were ineffective in nontoxic doses by the former test, whereas only phenobarbital, trimethadione, meprobamate, and phenaglycodol were ineffective as measured by the latter test. Thus, chlorpromazine, promazine, and trifluprom azine effectively reduced amphetamine toxicity in both situations, whereas diphenylhydantoin, phenacemide, hydroxyzine, and reserpine effectively reduced the toxicity of this substance only in the less severe 10-mice-per-cage test.

DISCUSSION

Three tests were employed to measure the tranquilizing activity of the drugs selected for study; namely, hexobarbital potentiation, a conditioned avoidance response, and protection against amphetaminetoxicity in aggregated mice. It is well known that many diverse agents, including tranquilizers, sedatives, anesthetics, antihistaminics, and other substances, increase the duration of sleep induced by barbiturates (12). However, this is one of few tests which gives positive results with all drugs claimed to have tranquilizing properties (12). Although conditioned responses are extensively used to evaluate psychotropic substances, the ability to alter conditioned behavior in animals is not limited to tranquilizing agents. For example, morphine, 5-hydroxytryptamine, mescaline, and iproniazid have been shown to block the secondary conditioned response (SCR) and/or conditioned response (CR) (13–15). On the other hand, there have been no reports which indicate that agents other than those with tranquilizing activity can reduce amphetamine toxicity in aggregated mice in nontoxic doses. These facts emphasize that no single method can be relied upon to detect the tranquilizing properties of a drug. The above battery of tests was selected because it appeared broad enough to include all agents with tranquilizing properties and selective enough to delineate drugs with mild tranquilizing activity from those with potent activity.

The data presented indicate that the anticonvulsant and psychopharmacologic drugs studied may be divided into 3 distinct groups: (I) phenobarbital, trimethadione, meprobamate, and phenaglycodol, (II) diphenylhydantoin and phenacemide, and (III) hydroxyzine, reserpine, chlorpromazine, promazine, and trifluprom azine. The drugs in group I are reported to have similar profiles of action (1, 2, 16–18) and all possess sedative-anticonvulsant properties. They are characterized herein by having activity in only one test, hexobarbital potentiation. Thus, this group of drugs probably possesses little tranquilizing activity. The drugs in Group II are nonsedative, anticonvulsant agents and are characterized by ability to potentiate hexobarbital and to reduce amphetamine toxicity in aggregated mice. Thus, the two drugs in this group, diphenylhydantoin and phenacemide, appear to have some properties in common with the drugs in Group III, the tranquilizers, which are characterized by giving positive results in all three tests employed. Of the drugs in Group III, chlorpromazine, promazine, and trifluprom azine appear to be more potent tranquilizers than hydroxyzine and reserpine because they are the only agents

effective by the 3-mice-per-cage amphetamine toxicity test.

The published data on the effects of psychopharmacologic drugs on conditioned avoidance responses in rats are in agreement with those presented herein for mice. In rats, chlorpromazine and reserpine block the SCR and CR (13, 14, 19), whereas hydroxyzine blocks only the SCR (14). Trifluprom azine is more potent (20) and promazine is less potent (14) than chlorpromazine when compared for ability to block the CR in rats. Meprobamate and the barbiturates have little effect on the CR as long as the rats are capable of responding to the UR (electric shock) (13, 16, 19). Because these drugs also have essentially the same profiles of action against conditioned avoidance responses in mice, this species appears to be as useful as rats for evaluating drugs by this technique. In addition, the data reported by Lasagna and McCann (9) and Burn and Hobbs (11) on the effects of psychopharmacologic drugs on amphetamine toxicity in aggregated mice are in agreement with those presented herein. This technique appears to be a useful one for discerning tranquilizing properties in candidate psychotropic substances.

The data indicate that two of the anticonvulsant drugs studied, diphenylhydantoin and phenacemide, may have mild tranquilizing activity. In similarity to the tranquilizers (Group III, above) they lack hypnotic activity and may improve mood and behavior in epileptic patients (3, 21). Clinical reports indicate that diphenylhydantoin also calms disturbed nonepileptic psychotic patients (3–6), conditions in which the phenothiazines are effective (22). Phenacemide may improve behavioral disorders; but, in certain cases, it also causes adverse psychic changes (3, 21, 23). On the basis of these observations, it would appear that diphenylhydantoin and phenacemide do indeed possess some tranquilizing properties and that this property should be investigated more fully in human patients.

SUMMARY

Four anticonvulsant drugs and seven psychopharmacologic agents were tested in mice for ability to increase hexobarbital sleep time, to abolish a conditioned avoidance response (CAR), and to reduce amphetamine toxicity in mice aggregated 3 per cage (more severe test) and 10 per cage (less severe test). All drugs significantly increased hexobarbital sleep time. Chlorpromazine, promazine, trifluprom azine, reserpine, and hydroxyzine blocked the conditioned response and/or secondary conditioned response. Meprobamate, phenaglycodol, and the anticonvulsant drugs (diphenylhydantoin, phenacemide, phenobarbital, and trimethadione) had little effect on CAR in nontoxic doses. Only the phenothiazine derivatives were effective by the 3-per-cage amphetamine toxicity test. The phenothiazines, reserpine, and hydroxyzine were active in the 10-per-cage test; two nonsedative-anticonvulsant drugs, diphenylhydantoin and phenacemide, were also effective by this test, which suggests that they may have some tranquilizing properties. The sedative-anticonvulsant drugs (phenobarbital, trimethadione, meprobamate, and phenaglycodol) were inactive by both amphetamine toxicity tests. The significance of these findings is discussed.

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