A Pharmacological Evaluation of a Genetically Predisposed Conditioned Avoidance Response[1] (34368)

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(Introduced by A. M. Burkman)
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In spite of the variety of responses, such as lever-pressing, pole-climbing, hurdle-jumping, and wheel-turning, utilized in conditioned avoidance studies (1–5), little attention has been directed toward examining drug effects on responses based upon behaviors of probable significance to the survival of the species employed. Yet, such behaviors may exhibit a marked resistance to alteration by drugs. For example, although aggressive behavior of albino mice has been shown to be readily suppressed by small doses of chlorpromazine and chlordiazepoxide (1), aggressive behavior of the carnivorous, predatory grasshopper mouse, *Onychomys leucogaster*, may be increased by similar treatment (6). The deer mouse is another species whose natural repertoire includes behaviors suitable for psychopharmacological investigation. Taking advantage of the genetically determined arboreal predisposition of the woodland deer mouse, *Peromyscus maniculatus gracilis*, Wolf *et al.* (7) demonstrated that an arboreal conditioned avoidance response (CAR) was more rapidly acquired, less readily extinguished, and more resistant to suppression by chlorpromazine than was a relatively foreign, terrestrial, conditioned avoidance response.

In the present study, we examined the ability of several central depressants to alter an arboreal and a terrestrial CAR in the prairie deer mouse, *P. m. bairdi*. The *bairdi* is strictly a terrestrial mouse which is physically well suited to a terrestrial habitat (8), and whose terrestrial behavioral patterns are genetically determined (9). In view of the terrestrial predisposition of this mouse, it was hypothesized that a terrestrial CAR would be more rapidly acquired, less readily extinguished, and more resistant to suppression by drugs than would a comparable but "genetically foreign" CAR.

Methods. The apparatus and procedures employed in these studies have been described in general previously (1). Briefly, mice were trained to perform a typical shuttlebox CAR. They were placed in one end of the shuttlebox and, after a 5-sec environmental exposure period, 5 sec of buzzer (CS) was presented, followed by up to 30 sec of footshock (US) while the buzzer remained on. To perform an avoidance (CR) or an escape (UR) response, subjects trained on the terrestrial paradigm were required to remain on a wood "safe area" pan placed at grid level at the far end of the shuttlebox. For arboreal responders, the pan was replaced with a threaded Plexiglas pole. A CR occurred when the mouse remained on the pan or pole with all four limbs off of the grid for a minimum of 5 sec in response to the environmental exposure or CS. A similar behavioral response to the US constituted a UR.

Groups of 12 and 9 adult, naive, male *bairdi* were employed in studying the acquisition and extinction of the terrestrial and arboreal CAR's, respectively. Each mouse was given 10 acquisition trials daily until it performed 10 avoidance trials in 1 day. The day after reaching this 100% avoidance criterion, the mouse was subjected to an extinction procedure differing from the acquisition procedure only in that the shock was deleted. Extinction trials were terminated when the mouse failed to respond during the first 10 sec of each trial on a given day or after 100

[1] Supported by USPHS Grant No. MH-07397-06.

TABLE I. Median Toxic Doses and Times of Peak Drug Effect.

	CPZ	CDP	MPB	PTB
TD_{50}[a]	36.0 (29.5–43.9)[b]	79.5 (69.7–90.6)	175 (153–199)	21.0 (18.8–23.5)
TPE[c]	60	10	20	5

[a] Median toxic dose (mg/kg).
[b] 95% fiducial limits.
[c] Time of peak drug effect (min).

extinction trials (10 days) had passed. During the acquisition and extinction trials, the nature of each response (CR, UR, or no response) was recorded.

The drugs employed, chlorpromazine HCl (CPZ), chlordiazepoxide HCl (CDP), meprobamate (MPB) and sodium pentobarbital (PTB) were prepared as has been described (1), and were administered intraperitoneally. Drug vehicles served as control treatments. Times of peak drug effect (TPE) and median toxic doses (TD_{50}), presented in Table I, were based on roller rod performance as described (10), except that the rate of rotation employed was 4 rpm.

Seventy-two adult, male, naive *bairdi* were equally divided into three arboreal and three terrestrial groups. One arboreal and one terrestrial group received control and drug treatments at a dose of 0.25 TD_{50}. A second pair of groups received 0.5 TD_{50}, and a third pair, a full TD_{50}. Each mouse within a group was eventually given the same treatments (drug or control) as all other mice of that group, but in a unique sequence according to a random but balanced design.

Drug treatments, administered no more frequently than once every 5 days, were given only to animals exhibiting at least 80% avoidance on the preceding day. Drug trials, carried out at the TPE of the drug given, were modified from those described for the acquisition study by limiting the maximum duration of the US to 10 sec and the number of trials to five. A scoring system was devised to take advantage of the differential levels of performance observed in evaluating drug effects. A mouse performing a secondary CR (11) was assigned 4 points; a CR, 3; a UR, 2; and nonresponders, 1 point. Since drug effects were measured over a series of five trials, a fully trained mouse would receive a behavioral score of 20 points. It should be stressed that all statistical analyses of the drug treatments were performed utilizing appropriate individual control scores. Thus, each animal served as its own control. The statistical tests employed to analyze results, *i.e.*, Friedman's Two-Way Analysis of Variance, the Wilcoxon Matched-Pairs Signed-Ranks test, the Kruskal-Wallace One-Way Analysis of Variance, and the Mann-Whitney *U* test, are fully described by Siegel (12).

Results. The acquisition of the arboreal and terrestrial conditioned behaviors are illustrated in Fig. 1. In this terrestrial mouse,

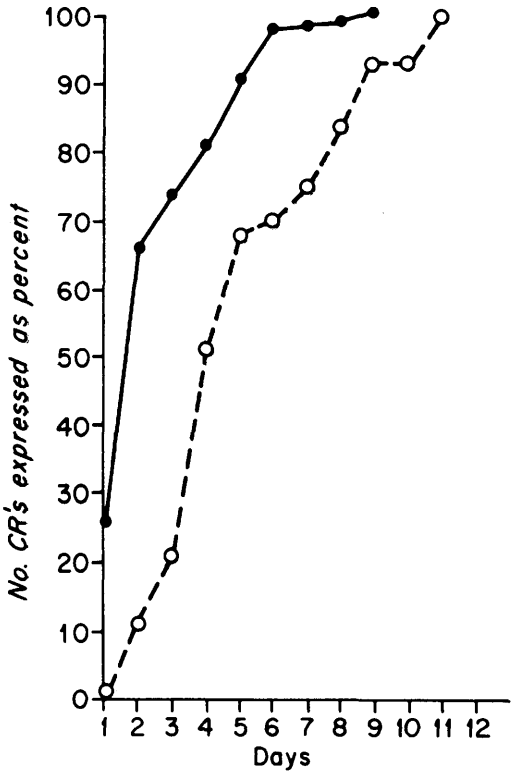

FIG. 1. Acquisition of arboreal and terrestrial CAR. (○), arboreal response; (●), terrestrial response.

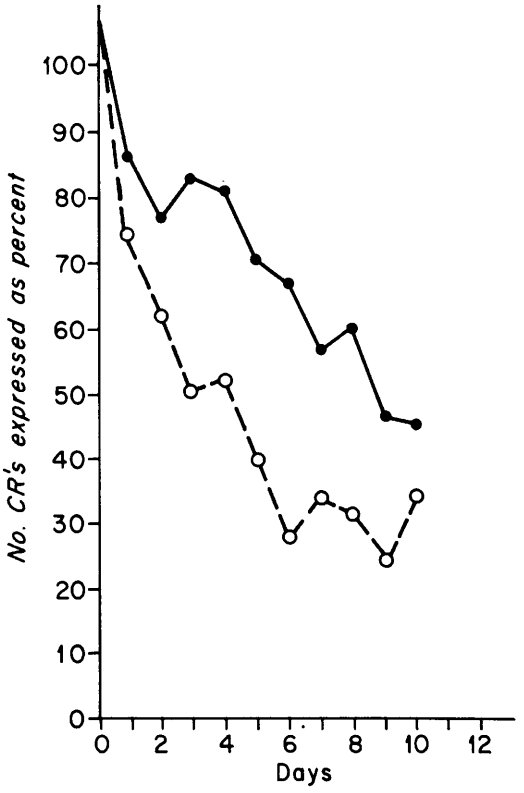

FIG. 2. Extinction of arboreal and terrestrial CAR. (○), arboreal response; (●), terrestrial response.

it is shown that the pan response is more readily acquired than is the pole response. The arboreal group required significantly more trials to reach the 100% avoidance criterion than did the terrestrial group ($p<.01$). Extinction curves for the two conditioned behaviors are presented in Fig. 2. Pan responders exhibited a greater resistance to extinction than did pole responders. The overall number of responses performed by pan responders was significantly greater than that of pole responders ($p<.05$).

The ability of the drug treatments to depress the terrestrial conditioned avoidance responding of *bairdi* is summarized in Table II. The distilled water and the methylcellulose controls did not differ significantly from each other. Mean overall control performance was 17.5. At the lowest dose level employed, none of the treatments depressed the terrestrial conditioned behavior significantly. At a dose level of 0.5 TD_{50}, only CPZ exerted a significant effect, and at 1 TD_{50}, all drugs except CDP significantly interfered with terrestrial performance. Table III displays the effects of drug treatments on the arboreal conditioned response. Arboreal avoidance behavior was markedly suppressed by most of the drug treatments. Indeed, all drug treatments except for the lowest doses of PTB and MPB significantly depressed performance of the pole response. Again, no difference in performance was found between the distilled water and the methylcellulose treated mice. Overall control scores averaged 17.1 and did not differ from those of the terrestrial groups.

Discussion. The results obtained provide support for our contention that a genetically predisposed behavior should be more easily elicited, more resistant to extinction, and less susceptible to alteration by drugs than a more foreign behavior. As the acquisition and extinction curves indicate, the terrestrial CAR was significantly more stable than the arboreal CAR. Although the possibility that the relative ease of performing the terrestrial response may be responsible in part for the greater stability of this behavior cannot be excluded on the basis of these data alone, results obtained in the drug studies lend added support to our hypothesis.

CPZ is known to exert a selective blockade of conditioned avoidance responses while MPB and the barbiturates are nonselective in this respect (1, 2, 4, 5). CDP has also been found capable of suppressing conditioned avoidance behaviors, although less selectively than does CPZ (3, 13). Table III shows that at the 0.25 TD_{50} dose level, CPZ and

TABLE II. Effect of Treatments on Terrestrial Performance.

Dose (TD_{50})	Mean behavioral score					
	CPZ	CDP	MPB	PTB	C_1[a]	C_2[b]
1	12.7[c]	16.5	15.8[d]	14.2[c]	17.2	17.8
0.5	14.1[c]	17.8	17.4	17.8	17.6	17.8
0.25	16.1	17.5	16.9	18.3	17.7	17.3

[a] Distilled water control treatment.
[b] Methylcellulose control treatment.
[c] Significant drug effect ($p<.05$).
[d] Significant drug effect ($p<.025$).

TABLE III. Effect of Treatments on Arboreal Performance.

Dose (TD_{50})	Mean behavioral score					
	CPZ	CDP	MPB	PTB	C_1[a]	C_2[b]
1	10.0[e]	12.6[e]	7.5[e]	6.2[e]	17.1	17.8
0.5	11.2[e]	11.8[e]	14.0[c]	15.8[d]	17.1	16.3
0.25	14.1[e]	15.3[e]	16.8	17.3	17.3	16.8

[a] Distilled water control treatment.
[b] Methylcellulose control treatment.
[c] Significant drug effect ($p < .05$).
[d] Significant drug effect ($p < .025$).
[e] Significant drug effect ($p < .005$).

CDP, but not MPB or PTB, significantly depressed the conditioned pole response. With larger doses, all drug treatments interfered with this behavior. With one important exception, the same general relation is seen to hold for the pan response. As illustrated in Table II, 0.5 TD_{50} of CPZ significantly depressed the pan response, whereas twice as much MPB and PTB were required to suppress this behavior. However, CDP, even at a dose of full TD_{50}, failed to suppress the terrestrial behavior. Thus, in spite of a considerable degree of neuromuscular impairment, CDP-treated *bairdi* continued to perform the genetically predisposed response.

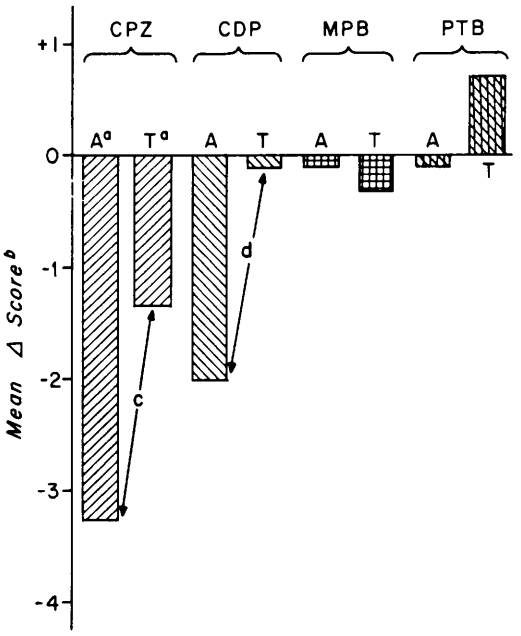

FIG. 3. Changes in behavioral score produced by 0.25 TD50 of the drugs indicated: (a) "A" indicates change in arboreal, and "T" in terrestrial behavioral score; (b) mean of differences of individual subject's drug and control scores; (c) difference is significant at $p < .025$; and (d) difference is significant at $p < .01$.

These results not only confirm that CPZ and CDP are able to interfere selectively with performance of certain conditioned avoidance responses, but also demonstrate that these drugs are more selective in suppressing a relatively foreign response than an avoidance response based on a genetically predisposed behavior. Indeed, if the mean difference in each animal's drug vs. control score is compared (Fig. 3), it is evident that at 0.25 TD_{50}, CPZ and CDP suppress the relatively foreign arboreal behavior to a significantly greater degree than the genetically predisposed response. It seems unlikely that the differential drug effects displayed in Fig. 3 are a reflection of the difficulty associated with the performance of the pole response *per se* since control behavioral scores did not differ for arboreal vs. terrestrial groups and equivalent fractions of equitoxic doses of drugs were employed throughout. Moreover, at 0.25 TD_{50}, none of the drugs produced observable neuromuscular impairment.

These results support and extend our previous contention (7) that the phenotypical propensity which underlies an organism's behavior may quantitatively alter the animal's response to drugs.

Summary. In *Peromyscus maniculatus bairdi*, a genetically predisposed terrestrial CAR was found to be more readily acquired, more resistant to extinction, and less susceptible to alteration by drugs than was a relatively foreign arboreal CAR. Subtoxic doses of CPZ and CDP were more selective in suppressing arboreal rather than terrestrial behavior, whereas similar equitoxic doses of MPB and PTB did not significantly depress either behavior. These results demonstrate that responses based upon behaviors included in the subject's natural repertoire are more stable and less readily altered by drugs than are responses relatively foreign to the subject's genetic predispostion.

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Received June 26, 1969. P.S.E.B.M., 1969, Vol. 132.