Laboratory Evaluation of Aggressive Behavior of the Grasshopper Mouse (*Onychomys*)

HENRY F. COLE* and HAROLD H. WOLF

Abstract ☐ The genetically predisposed aggressive behavior of two species of grasshopper mouse, *Onychomys leucogaster* and *O. torridus*, was evaluated in an experimental paradigm involving isolation-induced aggression. *O. leucogaster* exhibited considerably more aggression than did *O. torridus*. When aggression was provoked in *Onychomys* by isolation, the behavior could not be suppressed by pentobarbital nor by chlordiazepoxide. Indeed, chlordiazepoxide increased the amount of time spent fighting. Chlorpromazine was able to depress fighting behavior in *torridus* but not in *leucogaster*. The results obtained support the contention that an animal's behavioral predisposition may markedly alter the organism's response to drugs.

Keyphrases ☐ Grasshopper mouse—aggressive behavior ☐ Aggression—genetic predisposition, mice ☐ Chlorpromazine HCl, chlordiazepoxide, Na pentobarbital effect—aggression ☐ Genetic predisposition effect—pharmacological tests

The effects of tranquilizers and other central nervous system depressants on the aggressive behavior of albino mice have been studied by many investigators (1–7). Such investigations reveal that aggressive behavior of most albino mice is mild and relatively unstable. This is not surprising because albino mice have been bred for hundreds of generations for docility and tolerance to high-population densities. Moreover, in laboratory colonies, two major causes of aggression, competition for food and competition for territory, are removed. Under these conditions, a high level of aggressiveness becomes an undesirable trait, to be removed from the population.

In view of the relative docility of albino mice, the authors felt that an aggressive animal might serve as a more appropriate subject for studies on aggression. One such animal is the northern grasshopper mouse, *Onychomys leucogaster*. These grasshopper mice are predatory and carnivorous. In their natural habitat, they stalk and quickly kill other small rodents by gnawing into the base of the victim's brain (8). Ingles (9) estimates that small mammals and insects form 90% of *Onychomys*' natural diet. In the laboratory, the fighting behavior of *O. leucogaster* is readily elicited, quite stable, and easily quantified (10). When given the opportunity, *leucogaster* will quickly and vigorously attack a victim mouse. When given regular opportunities to fight, they may become so adept at expressing this behavior that the victim is killed as quickly as 20 sec. after its exposure to the aggressor (10).

The highly stereotyped and invariable method of killing by gnawing through the base of the skull exhibited by *leucogaster*, as well as the ease with which the behavior is elicited, even in naive subjects several generations removed from the wild, strongly suggests that this behavior is primarily genetically determined rather than learned. Therefore, the authors thought it worthwhile to investigate the genetically predisposed aggressive behavior of these grasshopper mice in an experimental paradigm involving isolation-induced aggression.

Members of a second species, *O. torridus*, the southern grasshopper mouse, were also included in this investigation. Because the authors are unaware of any studies concerned with the behavior of *torridus* under laboratory conditions, they felt valuable behavioral data could be provided by the inclusion of this species.

EXPERIMENTAL

A commonly employed method of producing aggressive behavior in albino mice is to subject them to extended periods of social isolation. Since such isolation-induced aggression has been frequently employed in drug studies (1, 2, 4, 6), the authors also utilized this method in their investigations of the aggressive behavior of *Onychomys*.

The *O. leucogaster* ranged in weight between 24 and 41 g., were derived from a colony previously maintained at the University of Utah College of Pharmacy, and were approximately 6 to 10 generations removed from the wild. The *O. torridus* were first-generation offspring of mice trapped by The Pet Corral, Tucson, Arizona, and weighed 20–32 g. *Onychomys* were segregated according to sex and were housed in groups of at most three mice to a standard 43 × 25 × 12-cm. plastic cage. When it became necessary to employ female subjects, care was taken to ensure that pregnant females were excluded. Purina Lab Chow and water were continually available in the cages. This diet was periodically supplemented with sunflower seeds, wheat, or small quantities of canned dog or cat food. Results were statistically analyzed by means of the following specific procedures; except where otherwise indicated, differences were considered to be significant at $p < 0.05$.

The drugs employed in these studies, chlordiazepoxide HCl (CDP), chlorpromazine HCl (CPZ), and sodium pentobarbital (PTB), were administered intraperitoneally in aqueous solution in such concentration that 1 ml./100 g. body weight contained the appropriate dose. Dosage and time of peak drug effect (TPE) were based on a roller-rod test slightly modified from that previously described (11) in that a speed of 4 r.p.m. was employed. The time at which the greatest number of mice were unable to remain on the rod for at least 30 sec. in any one of three trials (minimal neurological deficit) was taken as the time of peak drug effect (TPE). Employing this TPE, the dose of each drug producing evidence of minimal neurological deficit in 50% of the mice (TD_{50}) was calculated by the method of Litchfield and Wilcoxon (12). All further drug-behavior studies were carried out at the TPE using a dose level of $^1/_2$ TD_{50}. The TPE's and TD_{50}'s for the drugs employed are presented in Table I.

Four male and two female adult *leucogaster* and five male and two female adult *torridus* were caged individually and then tested at intervals of every 2 or 3 days until they would consistently attack a small albino victim mouse introduced into the aggressor's cage for a 5-min. period. Isolation periods as short as 3 or 4 days were usually sufficient to evoke this behavior in *leucogaster*, whereas *torridus* required as much as 2 to 3 weeks of isolation. When attack behavior was consistently displayed by *Onychomys*, the drug studies were initiated. Mice were randomly assigned to receive each of four treatments (control, CPZ, CDP, and PTB) in a unique sequence. Mice were injected with the appropriate treatment; at the TPE listed in Table I, a small (10–14 g.) victim albino mouse was placed in the cage of the *Onychomys* for a maximum of 5 min. The latency to the first attack by the aggressor and total amount of time spent engaged in aggressive activity were recorded. Mice not observed to attack the

Table I—Median Toxic Doses and Times of Peak Drug Effect

Species	CPZ		CDP		PTB	
	TD_{50}[a]	TPE[b]	TD_{50}	TPE	TD_{50}	TPE
O. leucogaster	10.0 (6.8–14.6)[c]	60	54.0 (31.8–91.8)	20	22.0 (17.6–27.5)	5
O. torridus	10.2 (5.7–18.4)	60	29.5 (18.4–47.2)	5	14.0 (9.5–20.6)	10

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

victim within the 5-min. test period were assigned a latency of 300 and a fighting duration of 0 sec. If a victim was killed before the end of the 5-min. test period, the time was noted and the victim was removed immediately.

Mice were given one of the four treatments no more than once every 5 days. On the day immediately preceding that on which a treatment (drug or control) was administered, each mouse was injected with distilled water and tested at the TPE of the treatment to be given the following day. Thus, each mouse received five control (distilled water) treatments over the duration of the study. This paradigm allowed each treatment to be compared to the immediately preceding control treatment and, furthermore, allowed an evaluation of the stability of the behavior with respect to time and exposure to the fighting situation. Moreover, this schedule permitted each animal to serve as its own control.

The results, unless otherwise indicated, were analyzed with the Wilcoxon matched-pairs signed-ranks test or with the Friedman two-way analysis of variance (13).

RESULTS

The effects of $^1/_2$ TD_{50} of chlorpromazine, chlordiazepoxide, and pentobarbital and of the combined control treatments on the amount of time spent engaged in aggressive activity after isolation are displayed in Figs. 1 and 2 for *leucogaster* and *torridus*, respectively. None of the drugs significantly depressed fighting time in *leucogaster*. Indeed, CDP significantly increased the duration of fighting in this species. The fighting time of *torridus* was also slightly, but significantly, prolonged by this drug, while CPZ significantly decreased the duration of fighting time of *torridus*.

The time that the victims survived on exposure to either species of *Onychomys* after the various treatments is tabulated in Table II. None of the drugs significantly altered the time either species required to kill a victim. However, as Table III reveals, CPZ did significantly increase the latency of *torridus* to attack a victim.

A statistical analysis of differences in performance between each of the five control treatments for either species revealed no significant changes in latency to attack, survival time of the victim, or fighting time. Thus, levels of aggression remained constant over the entire experimental period. Moreover, a given treatment had no appreciable lasting effect on subsequent fighting performance.

A Mann-Whitney "U" test comparison (13) of control performances of *torridus* and those of *leucogaster* revealed that *torridus* exhibited significantly longer latencies to attack and significantly shorter fighting durations than did similarly treated *leucogaster*. Although no significant difference was found between the survival times of *leucogaster's* and *torridus's* victims, *leucogaster* did kill a significantly ($p < 0.01$) greater number of their victims (58%) than did *torridus* (40%) as determined by the Fisher exact probability test (13).

DISCUSSION

Under the conditions employed in these studies, the *torridus* appeared to be somewhat less aggressive than *leucogaster*. In the isolated subjects, *torridus* displayed longer latencies to attack, shorter fighting times, and made fewer kills than did *leucogaster*. Moreover, in an unpublished study conducted in this laboratory, it was observed that a conditioned aggressive response which was readily acquired by *leucogaster* could not be reliably reproduced in *torridus*. Studies to date from this laboratory have not provided an apparent explanation for the quantitative discrepancy in aggression exhibited by the two species of *Onychomys*, and the authors are unaware of any studies comparing the aggressive behavior of these two mice either in the laboratory or the field. Nevertheless, it should be emphasized that both species of *Onychomys*, especially when

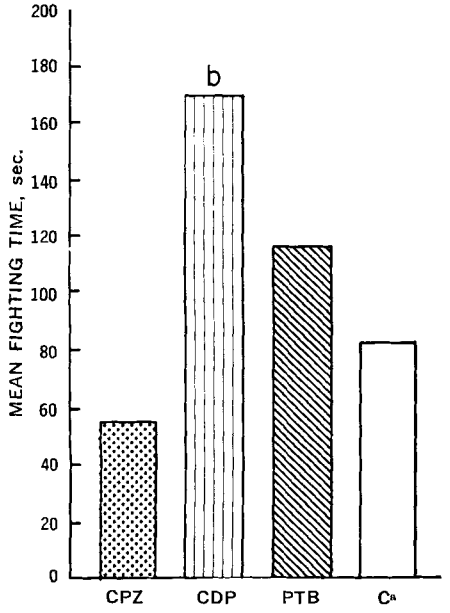

Figure 1—*Effects of drugs on fighting time of* O. leucogaster. *Key: a, combined control treatments; and b, significant drug effect* ($p < 0.05$).

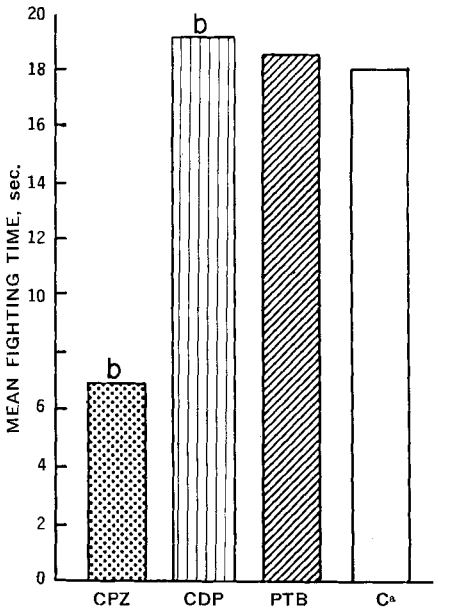

Figure 2—*Effects of drugs on fighting time of* O. torridus. *Key: a, combined control treatments; and b, significant drug effect* ($p < 0.05$).

Table II—Mean Survival Times of Victim Mice, Given in Seconds

Aggressor	CPZ	CDP	PTB	Control
O. leucogaster	156	190	151	178
O. torridus	250	199	146	219

Table III—Mean Latencies to Attack Victim Mice, Given in Seconds

Aggressor	CPZ	CDP	PTB	Control
O. leucogaster	11.0	6.7	5.2	7.2
O. torridus	121.9[a]	34.4	51.7	46.1

[a] Significantly longer than control latencies ($p < 0.05$).

isolated, were found to display a markedly greater degree of aggressive behavior than would albino subjects under similar conditions.

To evoke aggressive behavior in albino subjects, weanlings are typically isolated for periods of a month or longer (1, 7, 14). In contrast, the genetically predisposed aggressive behavior of *torridus* could be evoked in half this time and *leucogaster* would consistently attack victim mice after periods of isolation as short as 3 or 4 days. Moreover, less than 70% of a population of albino mice subjected to extended periods of social isolation can be expected to display even a minimal amount of aggressive behavior (1, 4, 7), while each of the *Onychomys* vigorously and consistently attacked the victim mice after the isolation period. Furthermore, an analysis of the latencies to attack, fighting times, and killing times for each of the five control treatments which every *Onychomys* received revealed no change for either species over the entire course of this study (eight opportunities to display aggressive behavior). In contrast, in a similar experimental paradigm, half of a population of albino mice failed to display a minimal amount of aggressive behavior after as few as six exposures to a victim (1). It should be mentioned also that while albino mice are commonly isolated while relatively young, the *Onychomys* employed in these studies were placed in isolation as adults, a time when isolation is much less effective in producing aggression (14).

The aggressive behavior of albino mice is characterized by sparring and nipping, components of mild rather than severe fighting. The injury to the victim is only superficial and the authors have never observed an albino aggressor kill a victim, even though exposure periods of up to 15 min. were employed (1). On the other hand, *torridus* and *leucogaster* killed 40 and 58%, respectively, of their victims.

Differences between the aggressive behavior of isolated albino mice and *Onychomys* are also apparent with regard to drug effects. For example, CPZ has been shown to depress the isolation-induced aggressive behavior of albino mice in small, nontoxic doses (1–3, 6). In contrast, a dose of CPZ as large as $1/2$ TD_{50} was ineffective in suppressing significantly the isolation-induced aggressive behavior of *leucogaster*. Although the less aggressive *torridus* did respond to CPZ, this effect might be at least partially due to a nonselective depression of the central nervous system, since at this dose the animals appeared to be somewhat sedated. Clark (10) also found the aggressive behavior of *leucogaster* to be quite resistant to suppression by this drug. He further reported that aggression was increased on the day following CPZ treatment. However, the experiments reported here were not designed to investigate such late alterations of behavior.

CDP has been reported to possess unique taming properties (15) and to be effective in calming aggressive or vicious albino mice and rats, dogs, cats, and monkeys (5, 15). The authors have found it effective in suppressing the isolation-induced aggressive behavior in albino mice in doses as low as $1/8$ TD_{50} (1). However, in both *leucogaster* and *torridus*, a much higher dose of CDP was not only ineffective in suppressing aggression, but indeed significantly increased the amount of time the aggressor spent engaged in fighting activities. This increase in fighting time could not be attributed to a decrease in fighting efficiency due to neurotoxicity, since both the latency to attack and survival time of the victims remained essentially unchanged.

Pentobarbital was not found to alter significantly the aggressive behavior of either species of *Onychomys*. Similarly, this nonselective central depressant is ineffective in altering the aggressive behavior of albino mice except in severely debilitating doses (1, 3, 4, 6).

It should be reemphasized that *Onychomys* are predatory rodents. In their natural habitat, survival probably depends in large part on the ability of these mice to track and kill victims for food. Moreover, this behavior appears to be genetically predetermined since individuals several generations removed from the wild will readily attack and kill victims in their typical stereotyped pattern. Taken in light of these factors, the experimental results reported here demonstrate how markedly the aggression of albino subjects differs from that of *Onychomys*. The relatively mild, difficult to elicit, and unstable aggressive behavior of albino subjects contrasts sharply with the genetically predisposed, predatory nature of *Onychomys*. Thus, the results the authors have reported support and extend their earlier contention (16) that the genetic predisposition underlying an animal's behavior may qualitatively and quantitatively influence the results obtained in pharmacological and behavioral investigations and must not be ignored.

REFERENCES

(1) H. F. Cole and H. H. Wolf, *Psychopharmacologia*, **8,** 389 (1966).

(2) J. P. DaVanzo, M. Daugherty, R. Ruckhart, and L. Kang, *ibid.*, **9,** 210(1966).

(3) W. D. Gray, A. C. Osterberg, and C. E. Rauh, *Arch. Int. Pharmacodyn. Ther.*, **134,** 198(1961).

(4) P. A. Janssen, A. H. Jageneau, and C. J. E. Niemegeers, *J. Pharmacol. Exp. Ther.*, **129,** 471(1960).

(5) L. O. Randall, *Dis. Nerv. Syst.*, **22,** Suppl. 2, 7(1961).

(6) H. C. Y. Yen, R. L. Stanger, and N. Millman, *J. Pharmacol. Exp. Ther.*, **122,** 85A(1958).

(7) H. C. Y. Yen, R. L. Stanger, and N. Millman, *Arch. Int. Pharmacodyn. Ther.*, **123,** 179(1959).

(8) V. V. Baily and C. C. Sperry, *U. S. Dep. Agr. Tech. Bull.*, **145,** 1(1929).

(9) L. G. Ingles, "Mammals of the Pacific States," Stanford University Press, Stanford, Calif., 1965.

(10) L. D. Clark, *Amer. J. Psychiat.*, **119,** 336(1962).

(11) T. R. Rudy and H. H. Wolf, *J. Pharmacol. Exp. Ther.*, **156,** 397(1967).

(12) J. T. Litchfield, Jr., and F. Wilcoxon, *ibid.*, **96,** 99(1949).

(13) S. Siegel, "Nonparametric Statistics for the Behavioral Sciences," McGraw-Hill, New York, N. Y., 1956.

(14) J. P. Scott and E. Fredericson, *Physiol. Zool.*, **24,** 273(1951).

(15) L. O. Randall, W. Schallek, G. A. Heise, E. F. Keith, and R. E. Bagdon, *J. Pharmacol. Exp. Ther.*, **129,** 163(1960).

(16) H. H. Wolf, E. A. Swinyard, and L. D. Clark, *Psychopharmacologia*, **3,** 438(1962).

ACKNOWLEDGMENTS AND ADDRESSES

Received November 12, 1969, from the *Division of Pharmacology, College of Pharmacy, Ohio State University, Columbus, OH 43210*

Accepted for publication January 28, 1970.

Abstracted in part from a thesis presented by H. F. C. in partial fulfillment of Doctor of Philosophy degree requirements.

This investigation was supported in part by grants from the National Institute of Mental Health (MH-07397-05 and 2-F1-MH-23, 869-05), U. S. Public Health Service.

Preliminary report: *Fed. Proc.* **27,** 438(1968).

* Present address: College of Pharmacy, University of Missouri, Kansas City, MO 64110