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Selective Breeding of Rats for High and Low Motor Activity in a Swim Test: Toward a New Animal Model of Depression

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WEISS, J. M., M. A. CIERPIAL AND C. H. K. WEST. *Selective breeding of rats for high and low motor activity in a swim test: Towards a new animal model of depression.* PHARMACOL BIOCHEM BEHAV **61**(1) 49–66, 1998.—Because low motor activity in a swim test has been found to represent "depression-like" behavior in the rat, Sprague–Dawley (SD) albino rats were selectively bred for low motor activity (low struggling time/high floating time) in a swim test, while others were bred for high motor activity (high struggling time/low floating time). Eighty-four male and 42 female SD rats were initially purchased from Charles-River Breeding Laboratories in 1987, their behavior assessed in a 15-min swim test, and selective breeding carried out by mating those male and female rats that showed either low or high levels of motor activity in the test; results from behavioral testing of the first 18 generations produced by this selective breeding process are reported here. Two rat lines have been obtained, Swim Low-Active (SwLo) and Swim High-Active (SwHi) rats, which differ dramatically in swim-test behavior—SwLo rats show little struggling and much floating, while SwHi rats show the reverse. Activity scores of individual SwLo and SwHi rats now show no overlap. Selective breeding has produced bidirectional changes; that is, SwLo rats are considerably less active than randomly bred Sprague–Dawley albino rats, while SwHi rats are considerably more active than randomly bred rats. Measuring activity of SwLo and SwHi rats in other situations—ambulation in the home cage, open-field activity, exploratory activity in a novel, home cage-like situation, and immobility in the Porsolt swim test—revealed that differences are most pronounced when animals respond to acute challenges; under these conditions, SwHi rats show active, assertive behavior, whereas SwLo rats show a distinct absence of this type of response. When SwLo rats from the 8th to the 11th generations were given antidepressant medication [desipramine, (DMI), a tricyclic, or phenelzine, an MAO inhibitor], chronic but not acute administration of both drugs increased swim-test activity of SwLo rats. Buspirone, an anxiolytic, did not increase activity of SwLo rats. Use of animals selectively bred for high and low activity in the swim test may represent a new tool for studying physiological processes relevant to affective disorders and for testing antidepressant drugs/treatments. © 1998 Elsevier Science Inc.

Swim test Selective breeding SwHi/SwLo rats Motor activity Animal models of depression Desipramine hydrochloride (DMI) Osmotic minipumps

MEASUREMENT of the activity of rats in a tank of water ("swim test") is now widely used in the experimental study of depression. This behavioral response is used both as a screening test for identifying effective antidepressant medications (5,21,22) as well as a method for revealing deficits in active motor behavior in animal models of depression (16,20,24,30). When activity in a swim tank is used as a screening test for antidepressant medication, the usual procedure is to place rats in a tank of water, remove them after 15 min, and then return them to this tank the following day for a 5-min swim test under the influence of the medication. During the initial exposure to the swim tank (i.e., 15-min duration), the animals become inactive; when they are reintroduced to the tank the next day, effective antidepressant medication decreases the inactivity (i.e., immobility) normally shown on this second exposure to the tank. When activity in a swim tank is measured in conjunction with animal models of depression, the usual procedure is to expose animals to some treatment, often a stress-

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ful condition, and then measure activity in a single session in the swim tank. Treatments that induce depressive symptomatology decrease the amount of activity (i.e., decrease active struggling behavior and/or increase floating behavior) in this swim test.

In so far as the procedures described above are useful and valid as a screening technique for effective antidepressant treatments and/or a measure of depression-related symptomatology, low activity of rats in a swim tank is, therefore, a manifestation of physiological processes related to depression. It is important to note that both the screening procedure for antidepressant drugs and the animal models *induce* a low level of swim-test activity in the animal. In the screening procedure for antidepressant medication, this is done by exposing animals to the initial 15-min session in the tank, during which active attempts to escape from the tank are extinguished. As a result, animals begin the drug trial (second exposure to the tank) with a reduced propensity to be active. In the animal models of depression, low swim-test activity is induced by exposing animals to some treatment that is thought to cause a depression-like condition.

What would occur if, rather than inducing low activity in the swim test by an environmental manipulation (such as preexposure to the swim tank or a stressful condition), animals were constitutionally inactive in this test? Would, for example, the responses within the central nervous system (CNS) that mediate inactivity be the same in these animals that are inherently inactive as in animals that are made inactive by preexposure to the swim test or exposure to a highly stressful situation? If so, do animals that are inherently inactive in the swim test possess some of the same CNS characteristics that are present in clinical depression without anything further being done to them? Questions such as these led us to determine if animals could be selectively bred for different levels of activity in the swim test in that such animals might prove useful in the study of depression.

Here we report on the generation of two selectively bred rat lines, Swim High-Active (SwHi) and Swim Low-Active (SwLo) rats, which differ dramatically in their swim test responses. SwHi rats respond in the swim test with active behavior, whereas SwLo rats respond with inactive behavior. This difference is not manifested in all situations; other studies show that it is most pronounced when animals are presented with a threatening or challenging circumstance in which active behavior is appropriate. This report concludes with studies showing that SwLo rats respond to chronic, but not acute, antidepressant drug treatment with increased swim-test activity, indicating that these animals may represent a new, genetically derived, animal model that possesses characteristics relevant to depression.

PART ONE: SELECTIVE BREEDING FOR DIFFERENTIAL MOTOR ACTIVITY

In this part, the derivation of SwLo and SwHi rats, and the behavior of these lines in the swim test, is described.

METHOD

Animals

Throughout all phases of this project, rats were housed in standard 45 × 25 × 15 cm clear polycarbonate cages living directly on bedding in groups of two to four animals per cage. The cages were contained in enclosed, laminar flow racks (Lab Products, Inc., NJ) as designed and described by Riley, Fitzmaurice, and Spackman (25). For breeding, male and female rats were housed together in pairs or two to three females with a single male; females were then removed and relocated into individual cages 20 days later. Standard laboratory rat chow and water was available ad lib for all animals. The colony room was maintained on a 12 h L:12 h D cycle (lights on 0700–1900 h).

Swim Test

The swim test employed was a modification of that initially described by Porsolt (21) and used by Weiss and co-workers as a test of behavioral depression in rodents (30). The swim tank consisted of a Plexiglas cylinder 65 cm tall and 30 cm in diameter. The tank was filled with water (25°C) to a height of 14 cm from the top. Swim tests were conducted in a quiet, darkened testing room with the tank illuminated by a 25 watt light suspended approximately 60 cm above the tank. A cardboard blind covered with black cloth was placed around the swim tank to provide a constant visual environment for all rats. An investigator observed and scored behavior through a 30 × 30 cm hole in the blind.

The swim test was conducted by removing an animal from its home cage in the colony room and bringing it to the adjacent testing room. For all studies, swim tests were carried out between 0800 and 1300 h. A set of "water wings" were then placed onto the animal. These consisted of a lightweight plastic bubble (made from "bubble pack" packing material), which was placed onto the midscapular area of the animal's back and held in place by a 1/2 inch wide strip of adhesive tape attached to the bubble and wrapped around the sternum. This bubble did not appear to restrict movement of the animal in any way while enabling all rats to float without sinking if they ceased movement. With the floatation bubble in place, the animal was dropped into the water from approximately 20 cm above the surface of the water so that the entire body of the rat dipped below the surface of the water on entry into the water. An investigator then observed the rat for a period of 15 min, timing the duration of the two types of motor activity: 1) struggling, which was defined as vigorous movement of all paws with the forepaws breaking the surface of the water (see Fig. 1, left); and 2) floating, which was defined as the animal remaining motionless with no movement of limbs (see Fig. 1, right). As reported previously (30), reliability coefficients for the timing of these categories of behavior by different investigators have always been found to be above 0.90. A measure of overall activity, called an "activity score," was then calculated from these measures by subtracting the total amount of time (in seconds) that the animal spent floating from the total time (in seconds) that the animal spent struggling; a negative activity score thus indicates a low level of active motor behavior in the test (i.e., more time spent floating than struggling) while a positive activity score indicates a high level of active motor behavior (i.e., more time spent struggling than floating). The investigator also noted any instances of diving behavior, recording a "full" dive if the animal submerged its entire body under the water or a "half" dive if the animal submerged only its head.

Selective Breeding

Selective breeding was initiated in July of 1987 with the testing of 84 male and 42 female Sprague–Dawley albino rats obtained from Charles River Breeding Laboratories, Wilm-

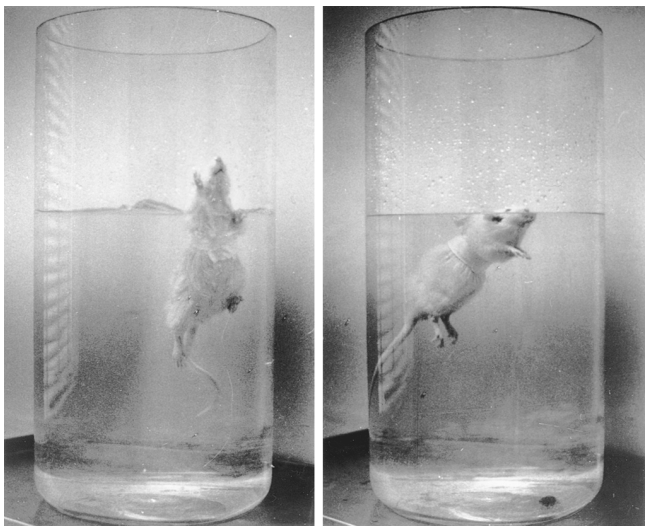

FIG. 1. At left is shown an animal in the swim tank exhibiting struggling behavior indicated by vigorous motor activity with both paws breaking the surface of the water, and at right is shown an animal exhibiting floating behavior indicated by the absence of any movement of limbs in the water.

ington, MA (referred to as Original Stock). Each of these animals was given the 15-min swim test as described above. From this stock, nine "high-active" and six "low-active" male-female pairs were selected and bred to produce the first generation (S1) of SwHi and SwLo rats. High-active breeders were selected for having the best combination of high struggling and low floating times and low-active breeders were selected for having the best combination of low struggling and high floating times. In each successive generation, animals were tested in the swim tank and selective breeding was repeated. Inbreeding was minimized by breeding animals that were least related to one another; brother–sister breeding was never done, and the animals that were bred rarely shared even the same grandparents. Animals were given the swim test when they were 90–120 days of age. Random-bred (or nonselected) animals were also produced in parallel with the SwHi and SwLo rats; these animals, derived from animals obtained from the same original source as those used for selective breeding, were simply bred without having been given the swim test. Generations of nonselected animals were given the swim test only intermittently to determine the swim-test activity of such animals, and were bred without consideration of swim-test scores when testing was done.

Statistical Analysis

For each generation, each measure obtained in the swim test (i.e., time spent struggling, floating, or activity score) was analyzed by a two-way analysis of variance using line (SwHi vs. SwLo) and sex (male vs. female) as main effects. If a significant effect due to line was obtained, post hoc protected *t*-tests were then carried out to compare the SwHi and SwLo rats of each sex in that generation. In the text below, the confidence levels of such comparisons are reported. When randomly bred (nonselected) rats were also analyzed, similar analyses were carried out including this additional line as well; subsequent contrasts between individual groups were then carried out as described above. Diving behavior was evaluated by contrasting the percentage of SwHi and SwLo rats in each generation that showed any diving in the swim test and determining whether the difference between these percentages reached statistical significance by Fisher's Exact Test.

RESULTS AND DISCUSSION

Table 1 shows data relevant to the selective breeding process. This table shows, for the first 18 generations, the number of SwHi and SwLo animals of each sex that were tested in the swim test, the number of male and female animals selected from this number for breeding, and the mean activity score of the animals that were used for breeding.

Figures 2 and 3 show the swim-test activity of male and female SwHi and SwLo rats for 18 generations. In both figures, struggling is shown in the top panel, floating in the middle panel, and activity score in the bottom panel. In each panel, the results from testing of the original stock of animals are shown as a solid bar at the center of the graph, and the values for successive generations of SwHi rats progress to the right of this solid bar while values for successive generations of the SwLo rats progress to the left of the solid bar.

Figures 2 and 3 show that the swim-test activity of SwHi and SwLo rat lines diverged in opposite directions from the original stock. The divergence in activity occurred rather rapidly in the selective breeding process. Among male animals, significant differences in both struggling time and floating time were evident from the S1 generation on (p at least $<$ 0.05, most comparisons $p < 0.001$). Among female animals, SwHi and SwLo animals differed significantly in floating in the S1 generation, and thereafter differed significantly in struggling as well as floating (p at least < 0.01). Activity Scores of SwHi rats differing significantly from same-sex SwLo rats in the S1 generation and in all generations thereafter (for all tests, $p < 0.001$).

The divergence in the activity of SwHi and SwLo rats became quite dramatic in magnitude, so that by the fifth generation of offspring there was very little overlap in the individual scores of rats from the two lines. To illustrate, Fig. 4 shows the activity scores of all individual animals of the original stock population and of all animals tested from the 7th (S7) and 13th (S13) generations of selective breeding. Also shown are the scores of animals from the seventh and thirteenth generations that were randomly bred (nonselected) in parallel with the SwHi and SwLo rats. For both males and females, the mean score of the SwHi and SwLo rats of the seventh and thirteenth generation not only differed from one another—in fact, individual scores of SwHi and SwLo rats showed no overlap in the 13th generation—but the mean of each of these lines also differed significantly ($p < 0.001$) from that of the nonselected animals of these generations.

Diving behavior of SwHi and SwLo rats is shown in Table 2. SwHi rats showed more diving than did SwLo rats, but a difference in this behavior emerged more gradually (i.e., took more generations to emerge) than did differences in struggling and floating. Consistent differences in diving did not emerge until S8, and significant differences were not seen until S13. But in each generation after S13, significantly more SwHi rats showed diving than did SwLo rats, the latter rarely showing any diving in the later generations.

In summary, the results described above show that it was possible to breed Sprague–Dawley rats for large differences in motor activity in the swim test. After 18 generations of selective breeding, differences were quite robust, with little, if any,

TABLE 1

NUMBER OF MALE AND FEMALE RATS SCREENED IN THE SWIM TEST AND THE NUMBER SELECTED FOR BREEDING IN EACH GENERATION OF SwHi/SwLo SELECTIVE BREEDING

	Original Stock	S1	S2	S3	S4	S5	S6	S7	S8	S9
SwHi										
Males tested	*84	43	14	10	14	16	12	17	15	16
Females tested	*42	56	27	27	24	16	13	24	25	12
Males bred	9	9	3	3	3	3	3	4	5	4
Activity score	−6.8	26.0	14.0	111.3	−26.0	73.7	213.7	211.5	221.4	174.5
(Mean ± SE)	± 45.6	± 33.2	± 4.9	± 9.9	± 109.7	± 24.8	± 46.5	± 44.5	± 17.9	± 24.9
Females bred	9	9	8	9	8	7	7	12	15	12
Activity score	−85.3	67.2	154.8	161.6	59.6	54.7	85.6	176.3	181.1	179.4
(Mean ± SE)	± 67.4	± 0.0	± 34.6	± 16.5	± 49.8	± 30.0	± 26.6	± 18.7	± 17.2	± 2.7
SwLo										
Males tested	*84	45	8	6	14	16	12	22	15	16
Females tested	*42	41	8	36	24	16	20	25	25	19
Males bred	6	3	5	3	3	2	3	4	5	5
Activity score	−478.8	−657.7	−575.0	−656.3	−719.0	−770.5	−589.0	−749.5	−672.2	−707.8
(Mean ± SE)	± 29.7	± 35.2	± 42.3	± 21.7	± 60.3	± 29.5	± 74.6	± 17.4	± 15.7	± 55.8
Females bred	6	3	5	9	8	5	8	12	15	15
Activity score	−277.2	−392.0	−338.0	−662.9	−579.0	−559.6	−576.1	−621.3	−550.7	−452.3
(Mean ± SE)	± 86.8	± 48.5	± 93.3	± 23.2	± 32.5	± 48.4	± 37.0	± 27.9	± 25.2	± 51.6

*Original foundation stock, thus same animals used to select SwHi and SwLo breeders.
Activity Score is for the rats selected as breeders. In generations 13 and 15, SwHi and SwLo females used for breeding were not tested.

overlap in struggling, floating, and activity scores of individual animals of the SwHi and SwLo lines.

PART TWO: OTHER BEHAVIORAL MEASURES IN SwHi AND SwLo RATS

One question concerning the SwHi and SwLo rats is whether the marked differences between the two lines that are described above are specific to the swim-test situation used for selective breeding or represent a pervasive and general difference in the propensity of the two lines of animals to show motor activity. To assess this question, SwHi and SwLo rats of various generations have been compared for motor activity in other situations; measures taken included assessment of (a) daily spontaneous ambulatory activity in the home cage, (b) activity in an open field, and (c) exploratory ambulatory activity in a novel situation similar to the home cage. Also, swim-test activity has been assessed in the standard Porsolt testing situation that utilizes shallower water in the swim tank than was used in the selective breeding process reported here so that the animal's rear feet can touch the bottom of the tank in the Porsolt test. The findings from the testing of various generations of SwHi and SwLo rats in these test environments are described in this part.

METHOD

Animals

Subjects were male and female SwHi and SwLo rats of S5, S13, and S14. In S5 and S14, male rats were 360–400 g and female rats were 240–260 g at the time of testing; in S14, male rats were 550–590 g and female rats were 310–340 g at testing.

Open-Field Test

Open-field ambulatory behavior was assessed in a chamber having a floor measuring 107 × 107 cm (40 × 40 in) with walls 50 cm (19 in) high. The walls of the chamber were painted black and the floor white. Black lines painted on the floor demarcated 25 squares of equal size [each 20 × 20 cm (8 × 8 in)]. The open field was housed in a quiet, darkened testing room and was illuminated by a 25 watt light bulb positioned approximately 90 cm above the center of the floor.

Animals were brought into the testing room and gently placed in the center square of the open field facing away from an observer who sat quietly on a stool positioned above one corner of the field so as to be able to view the entire enclosure. The observer then traced the path of the rat's movements on a map of the open field. The following measures of ambulatory activity were subsequently tabulated: number of outer squares (i.e., those along the walls) entered, number of inner squares (i.e., those not bordered by a wall) entered, and total number of squares entered. An animal was considered to have entered a square when it moved all four limbs across the boundary into that square; smaller movements of turning, spinning, or probing with the head or forelimbs were not counted as entering a square. The observer also counted instances of rearing, noting rearing against a wall (wall rears—rearing with one or two paws placed on the side of the chamber) or rearing without use of the wall for support (free rears). A rearing response consisted of the animal raising up on its hind feet with both forelimbs not in contact with the floor for at least 1 s. Before a new rear could be counted after an animal had reared, the animal had to move its forelimbs across the floor to a new location or it had to maintain contact with the floor for more than 1 s; thus, repeated up-and-down movements separated by only brief forelimb contacts with the

TABLE 1
CONTINUED

	S10	S11	S12	S13	S14	S15	S16	S17	S18
SwHi									
Males tested	15	9	16	19	26	20	12	2	12
Females tested	21	23	24	18	26	4	28	12	12
Males bred	5	5	5	3	9	8	3	2	6
Activity score (Mean ± SE)	196.2 ± 28.9	395.0 ± 66.9	310.8 ± 53.9	250.0 ± 18.9	365.1 ± 61.5	255.9 ± 55.9	277.7 ± 27.7	236.5 ± 40.5	225.2 ± 20.3
Females bred	13	15	11	6	24	10	14	10	10
Activity score (Mean ± SE)	286.0 ± 46.4	282.9 ± 36.6	283.8 ± 49.4	(not tested)	277.9 ± 30.0	(not tested)	461.3 ± 52.5	173.5 ± 19.0	225.0 ± 32.1
SwLo									
Males tested	11	14	11	19	24	20	16	2	12
Females tested	29	31	32	19	32	5	41	10	12
Males bred	5	5	5	3	9	6	6	2	4
Activity score (Mean ± SE)	−786.6 ± 31.3	−609.2 ± 35.5	−739.4 ± 32.0	−804.0 ± 28.2	−672.2 ± 40.5	−757.3 ± 29.0	−787.7 ± 21.0	−825.5 ± 38.5	−777.8 ± 13.9
Females bred	15	15	15	6	24	14	29	8	7
Activity score (Mean ± SE)	−585.9 ± 29.0	−535.5 ± 31.0	−567.5 ± 26.7	(not tested)	−512.3 ± 37.8	(not tested)	−679.8 ± 17.6	−661.1 ± 25.0	−759.4 ± 17.0

floor were not scored as multiple responses. The open-field test lasted 10 min. At the conclusion of the test, the animal was removed and the observer noted whether it had urinated and counted the number of fecal boli deposited. The floor of the open field was then cleaned thoroughly and washed with a dilute acetic acid solution in preparation for the next animal. This measure was made with S5 and S14 rats; 10 SwHi and SwLo rats of each sex were used from each generation.

Home Cage Ambulatory Activity

Ambulatory activity was measured throughout the day in the home cage. This recording was carried out in sound-attenuated rooms each containing 12 individual cages. These cages were of the same size and composition as those used to house animals in the colony room (i.e., standard 45 × 25 × 15 cm clear plastic cages); in these cages, animals also lived directly on bedding. For activity recording, animals were housed individually. Activity was recorded continuously by a photocell assembly (ANA 1219, Riverpoint Electronics) mounted in a frame; the frame contained an array of eight infrared light beams spaced equidistant from one another along the length of the cage. Movement of an animal within its cage resulted in interruptions of the light beams that were monitored electronically. The recording computer recognized any change in the pattern of light beams and responded by recording a new "sentence" in memory (a sentence described the status of the eight light beams, defining whether each was interrupted or uninterrupted). Ambulatory activity was distinguished from repetitive movements in the same location as follows: a unit (or count) of ambulatory activity was recorded if a newly generated sentence in memory included a change in the status of a beam that had not been altered in the previous 4 sentences; for this to occur required the animal to move into the field of a new beam. At the end of a 24-h recording session, the computer provided a printout of ambulatory counts per hour for each rat. Average ambulatory counts per hour were calculated separately for the light and dark portions of the day: night cycle (lights on 0700–1900 h). Animals were disturbed once per day (between 1400–1600 h) to be weighed, at which time food and water bottles were also weighed to obtain measures of daily intake. Measurement of spontaneous home cage activity was initiated on the day that animals were placed into the activity monitoring room; i.e., habituation to the recording cages was not allowed to occur before recording was begun so that a difference in response during the initial days in the apparatus would be detected. This measure was made with S5 and S14 rats; 10 SwHi and SwLo of each sex were used from S5. In S14, 10 SwHi and SwLo female rats and 6 SwHi and SwLo male rats were used.

Exploratory Ambulatory Activity in a Novel Situation Similar to the Home Cage

This measure was made under the same conditions as described directly above except that ambulation was recorded for a period of one hour, and analyzed for each 2-min segment, after the rats were initially introduced into the cages. Thus, animals were placed individually into cages similar to their home cage but containing new, clean bedding, and explored this environment for 1 h, during which ambulatory activity was recorded. This measure was made with S13 rats; 18 SwHi, SwLo, and randomly bred [i.e., nonselected (SwNS)] male rats and 12 SwHi and SwLo female rats were used.

Porsolt Swim Test

These conditions were meant to approximate the original procedure described by Porsolt and colleagues (21–23). The difference between the apparatus used by Porsolt et al. and that employed in the studies reported here (shown in Fig. 1) is that the swim tank used by Porsolt and colleagues contains

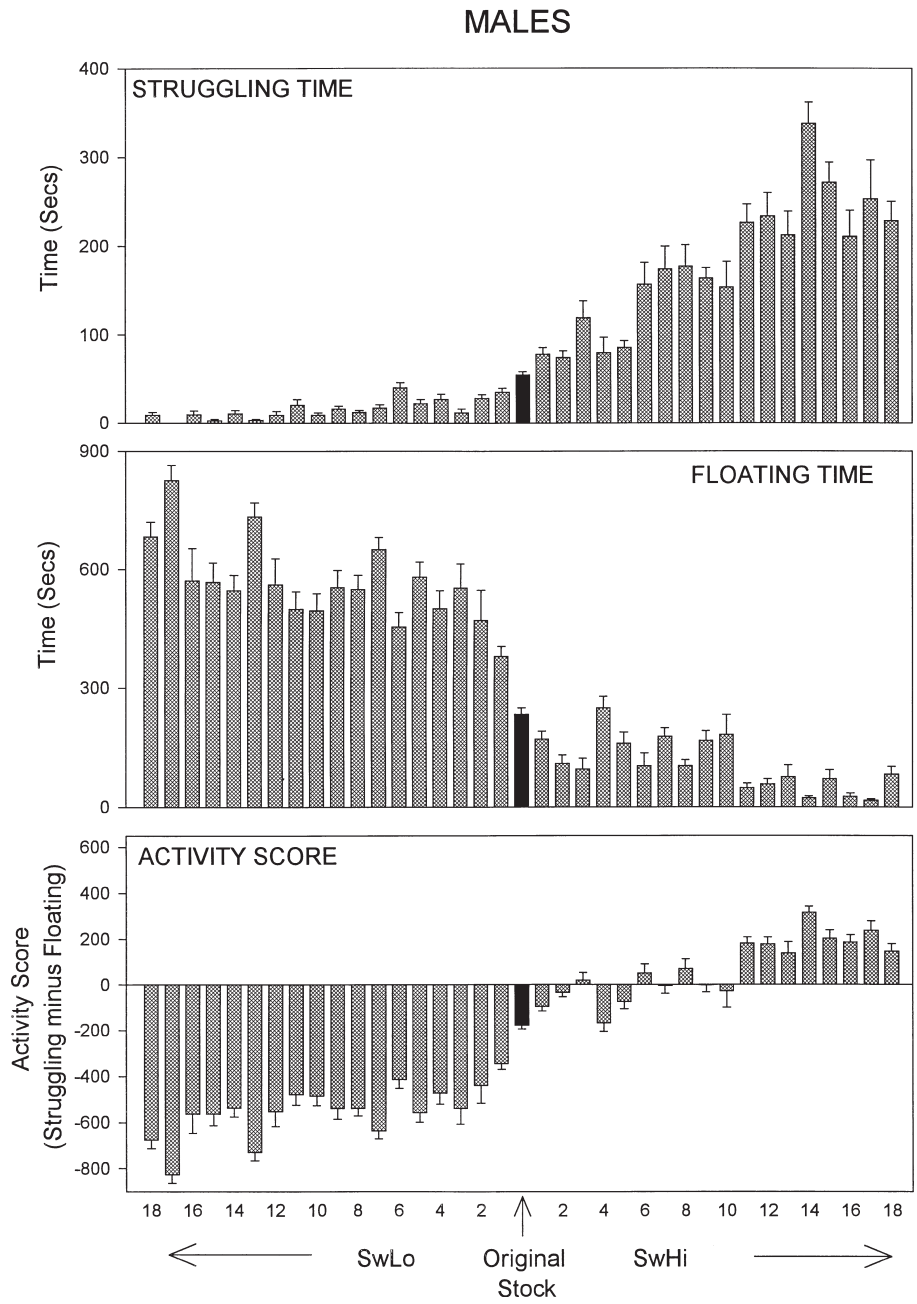

FIG. 2. Swim-test activity of male SwHi and SwLo rats from 18 successive generations of selective breeding. Shown is the mean (and standard error) time (in seconds) spent struggling (top) and floating (middle) in a 15-min swim test and activity score computed by subtracting floating time from struggling time (bottom). Each panel shows the mean for the original stock of animals purchased from Charles River Breeding Laboratory (indicated by the solid bar at the center) and for successive generations (S1–S18) of SwHi rats (progressing to the right) and SwLo rats (progressing to the left). The number of animals from each generation that was tested can be found in Table 1.

less water than does the tank shown in Fig. 1, and a rat in the Porsolt test can, with its body fully extended, reach the bottom of the tank with its rear feet to keep its nose above the surface of the water. The original studies of Porsolt and his colleagues utilized 15 cm of water for testing of 150–180 g rats. To approximate these conditions for larger rats that were used in the present study, the Porsolt test conducted here was carried out with a water depth was 19 cm for male rats (7–8-month-old rats, weighing approximately 560 g) and 17 cm for female rats (same age as males, weighing approximately 325 g). The same tank shown in Fig. 1 was used (without the screen surrounding the tank). Each animal was brought individually into the test room and gently dropped into the water; because the animal was not in danger of sinking in these conditions, no

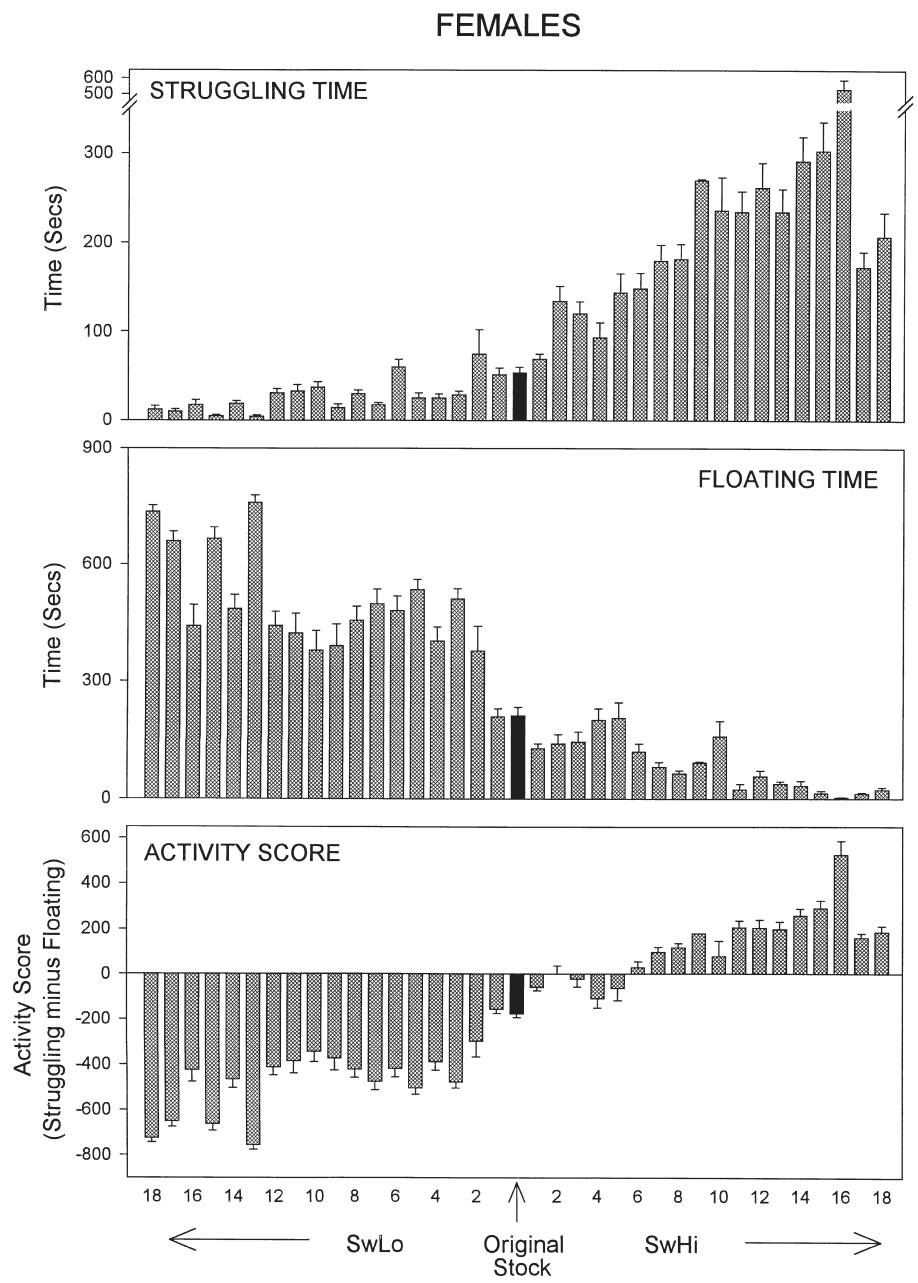

FIG. 3. Swim-test activity of female SwHi and SwLo rats from 18 successive generations of selective breeding. All details as in legend for Fig. 2.

flotation bubble was used. The test was carried out by exposing an animal to the tank for 15 min on day 1 and then, 24 h later, exposing it to the tank again for 5 min, which constituted the usual "test." Time spent immobile was quantified (timed) by an observer during both exposures to the tank. Immobility was defined as the animal remaining motionless in the water (no limb movement) except for small postural adjustments or minor head movements such as occur during sniffing. It was observed that, under these conditions where it is possible for the animal to reach the bottom of the tank with its body fully extended, some of the animals engaged in jumping, apparent attempts to escape from the tank. Jumps were also counted. A jump was scored when the rat elevated itself sufficiently that any part of its hind legs were above the surface of the water; hopping or less pronounced jumping was not counted. At the end of each session, the rat was removed from the cage, dried, and returned to its home cage. This measure was made with S13 rats; 9 SwHi, SwLo, and SwNS male rats and 12 SwHi and SwLo female rats were used.

Statistical Analysis

For each of the measures taken in the studies described above, the statistical significance of the difference between

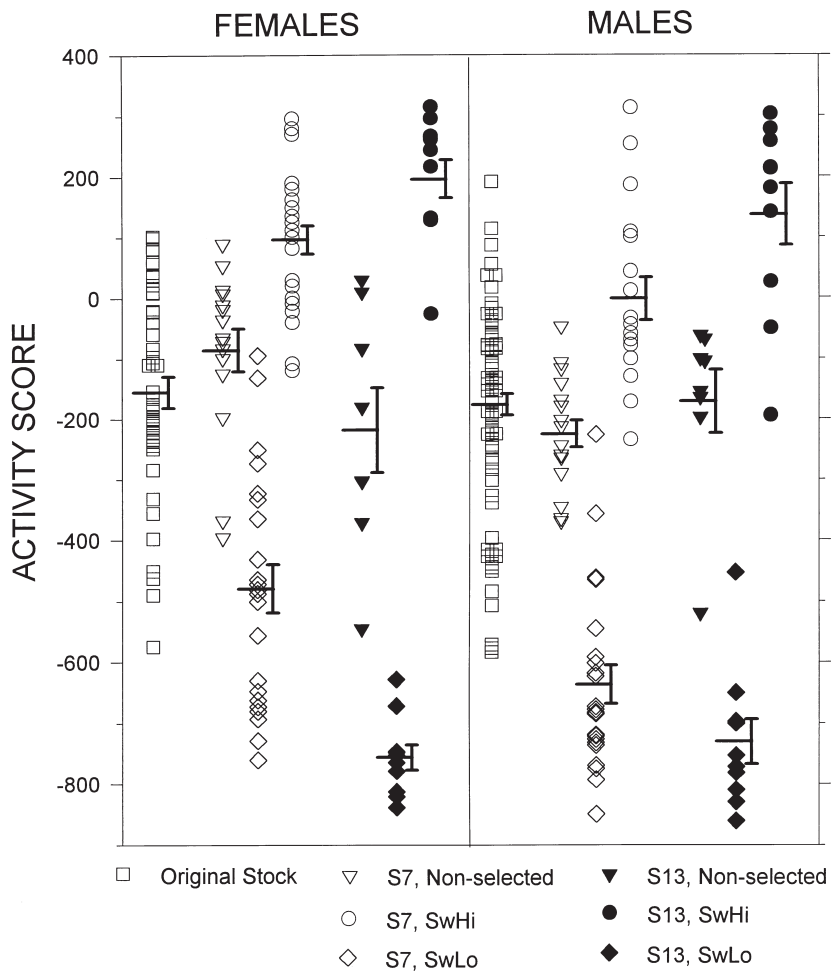

FIG. 4. Individual activity scores of all animals of the original parental stock purchased from Charles River in 1987 (Original Stock) and all SwHi and SwLo animals of the 7th generation (S7) and 13th generation (S13) of offspring. S7 and S13 nonselected animals were randomly bred for the same number of generations as SwHi and SwLo rats. Horizontal lines indicate group means and vertical lines denote ± one standard error.

any two groups (SwHi vs. SwLo or vs. SwNS rats when included) was determined by a *t*-test comparing the two groups.

RESULTS AND DISCUSSION

Daily Spontaneous Ambulatory Activity

Recording of spontaneous ambulatory activity in the home cage throughout the 24 h of the day was carried out on rats of S5 and S14. Measurement was carried out for 10 days. Figures 5 and 6 show the mean activity (average ambulatory counts per hour) observed during the dark and light periods of the day for each of these generations. In S5 (see Fig. 5), no appreciable or significant differences were evident between SwHi and SwLo rats. In S14 (see Fig. 6), however, some significant differences were seen, with female SwHi rats showing more ambulatory activity than female SwLo rats during both dark

TABLE 2

PERCENTAGE OF ANIMALS THAT DISPLAYED DIVING BEHAVIOR (AT LEAST ONE-HALF OR FULL DIVE) DURING A 15-MIN SWIM TEST ACROSS THE FIRST 18 GENERATIONS OF SwHi/SwLo SELECTIVE BREEDING

	OS	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11	S12	S13	S14	S15	S16	S17	S18
SwHi males	32.4	18.6	35.7	30.0	14.3	18.8	66.7	35.3	53.3	37.5	40.0	22.2	6.2	70.0	26.9	70.0	62.5	50.0	75.0
SwLo males		8.9	0.0	33.3	21.4	6.3	41.7	22.7	0.0	0.0	0.0	21.4	0.0	0.0	8.3	5.0	6.3	0	0
SwHi females	19.1	19.16	29.6	29.6	16.7	25.0	30.8	41.7	56.0	50.0	38.1	20.0	29.2	80.0	38.5	50.0	50.0	25.0	50.0
SwLo females		14.6	37.5	27.8	12.5	18.7	50.0	8.3	36.0	10.5	8.30	6.0	9.4	0.0	6.3	0	12.5	0	8.3

OS = original stock purchased in 1987. See Table 1 for number of subjects in each generation.

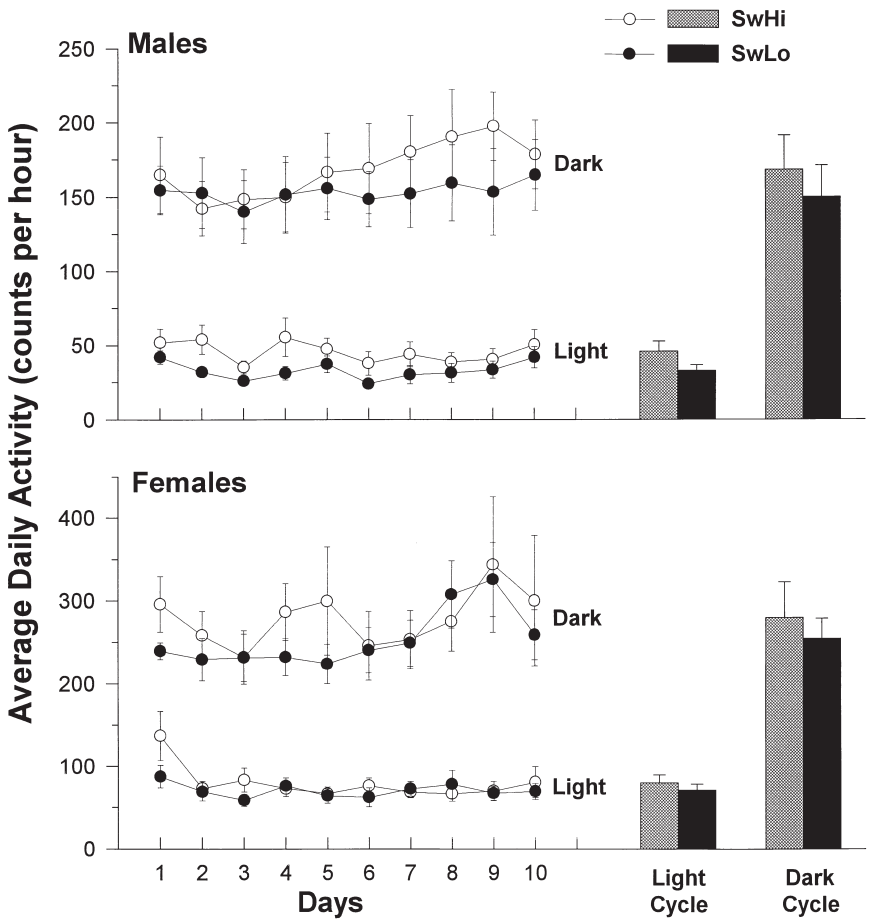

FIG. 5. Daily ambulatory activity in the home cage during the light and dark portions of the day for S5 male and female SwHi and SwLo rats. At left is shown the mean (and standard error) ambulatory counts per hour for each of 10 days of recording, and at right (bar graphs) is shown the average hourly activity for all 10 days. No statistically significant differences were found.

and light periods of the day. For male rats of S14, no difference in activity was seen during the dark, the period of most activity, but a small difference in ambulatory activity was seen during the light period, which was similar to a difference also present in the males of S5.

Activity in the Open Field

Activity in an open field was also assessed on rats of S5 and S14 (this measurement was made on different animals from those used for measurement of daily ambulatory activity). Figures 7 and 8 show the mean number of squares entered during the 10-min test as well as the mean number of rearing responses made. In S5 (see Fig. 7), female SwLo rats showed more activity than female SwHi rats, entering more inner, outer, and total squares of the open field. Female SwLo rats also reared more than SwHi rats. Male SwLo rats of S5 displayed a tendency to show more activity and rearing than SwHi rats, but differences were not statistically significant. In S14 (see Fig. 8), differences were more pronounced, with both male and female SwLo rats entering more squares than SwHi rats and also rearing more often.

Defecation, often measured in the open field, also showed differences. In S5, male SwHi rats defecated significantly more boluses than SwLo rats (mean = 1.6 ± 0.5 vs. 0.3 ± 0.2), but this difference was not evident in the female rats (1.3 ± 0.8 vs. 1.1 ± 0.6). In S14, male SwHi rats again defecated significantly more boluses than male SwLo rats (4.5 ± 0.9 vs. 0.8 ± 0.4), but the difference in female rats again was less pronounced and did not reach statistical significance (1.1 ± 0.7 vs. 0.2 ± 0.2).

Exploratory Ambulatory Activity in a Novel Situation Similar to the Home Cage

Animals from S13 were placed into the same home-cage apparatus as used for recording of daily ambulatory activity but in this case activity was closely monitored for only 1 h; thus, ambulatory activity was measured as animals first explored a novel home cage-like situation. Figure 9 shows mean ambulatory activity for each 2-min interval of the recording period. For both male and female rats, statistically significant differences in ambulatory activity were seen during the first 20–30 min after the animals were placed into the apparatus, with SwHi rats showing more ambulatory activity than SwLo rats [and, for male rats, also more activity than randomly bred (SwNS) rats]. Following 30 mins of exposure to this situation, however, differences in ambulatory activity were no longer seen.

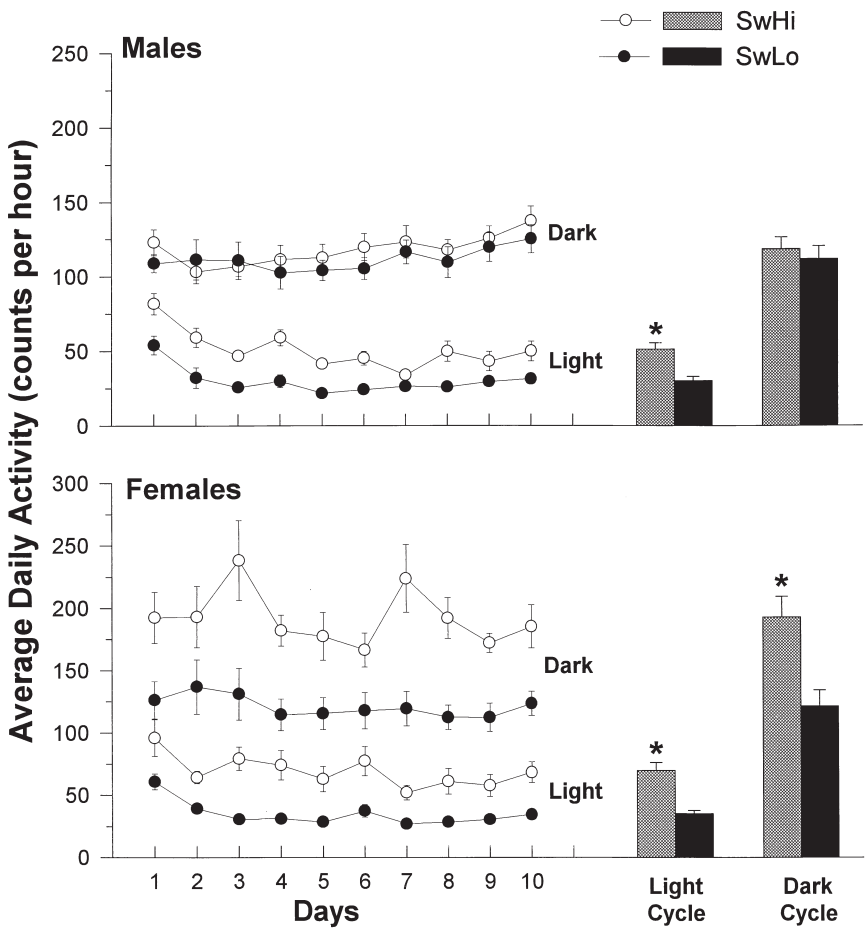

FIG. 6. Daily ambulatory activity in the home cage during the light and dark portions of the day for S14 male and female SwHi and SwLo rats. All other descriptions as in Fig. 5. *Significantly different (p at least < 0.05) from SwLo rats. [Significant differences on individual days (left side of figure) are not noted.]

Porsolt-Type Swim Test

Animals of S13 were subjected to conditions similar to those used by Porsolt and colleagues (21–23)—in the Porsolt test, animals are initially exposed to the swim tank for 15 min on day 1 and then are tested in the same apparatus for *5* min on the following day; also, the water in the swim tank is usually of a depth that subjects can, when fully extended, touch the floor of the tank with their rear feet. In the standard Porsolt procedure, time that the animal is immobile in the 5-min test on day 2 is reported. Figure 10 shows the mean time that animals were immobile during the initial 15-min exposure to the tank as well as during the 5-min test period 24 h after the initial exposure. Both male and female SwHi and SwLo rats differed in immobility time in the Porsolt procedure, with SwHi rats showing less immobility than SwLo rats. For male animals, randomly bred rats (SwNS) were also tested; in the 5-min test period, SwNS rats showed a mean immobility time that was roughly midway between that of the SwHi and the SwLo rats. Also, it was observed that, in this test where the animals were able to reach the floor of the tank when fully extended, they showed jumping behavior, apparent attempts to escape from the swim tank, and this response was also counted. SwHi rats of both sexes showed much more jumping activity than did SwLo or SwNS rats.

Summarizing the various results described above, these indicate that differences in motor activity of SwHi and SwLo rats depend upon the nature of the testing conditions. Thus, while differences between SwHi and SwLo rats in the swim test used (shown in Fig. 1) were so large that the individual scores of SwHi and SwLo rats showed virtually no overlap by S7, and did not overlap by S13, differences between SwHi and SwLo rats were much less pronounced when motor activity was examined under other circumstances. For example, when ambulatory activity was measured in the home cage across the entire day, ambulation of SwHi and SwLo rats was only moderately different—more ambulation by SwHi rats was clearly evident only in females of the S14 generation, with little difference between males even in that generation. In the open field, the SwLo rats actually showed more ambulation than did SwHi rats, with this difference being significant for both males and females in S14. Although it certainly could be pointed out that ambulation in the open-field test is strongly influenced by fear/anxiety [e.g., (29)], this does not detract from the fact that the conditions under which testing takes place clearly influence whether SwHi and SwLo will show dif-

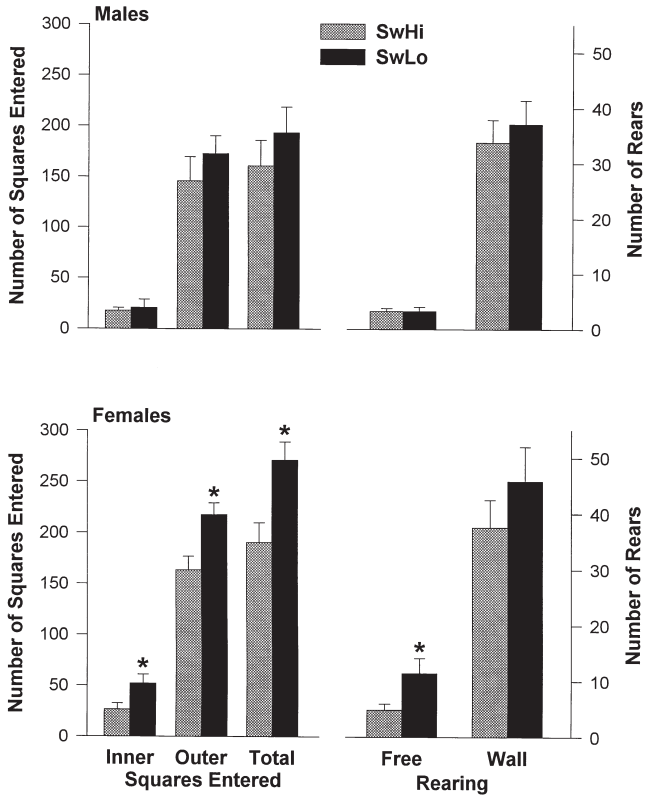

FIG. 7. Open-field ambulatory activity and rearing behavior for S5 male and female SwHi and SwLo rats. At left is shown the mean (and standard error) of squares entered (inner squares, outer squares, and total squares) and at right is shown mean (and standard error) number of rearing responses (free rears and wall rears). *Significantly different (p at least < 0.05) from SwHi rats.

ferences in motor activity. What general characteristics appear to describe circumstances in which SwHi rats show markedly more activity than SwLo rats? In addition to the swim-test situation used throughout this study, SwHi rats showed more motor activity than SwLo rats when one measured 1) immobility and jumping in the Porsolt test, and 2) initial exploratory activity in a home cage-like situation. Thus, in circumstances that tend to elicit, as an appropriate behavioral response, active, assertive motor behavior as a means of coping with and/or dealing with an immediate challenge—such as escaping from a tank of water or examining/exploring a relatively small, enclosed area—one sees large differences between SwHi and SwLo rats. Whereas SwHi rats respond to such conditions with large amounts of active, assertive motor behavior, SwLo rats respond to these conditions with a distinct absence of this type of motor behavior.

In concluding this section, it can be noted that it might be tempting to hypothesize from the results described above that SwHi rats are more emotional/anxious than SwLo rats, and that the differences in motor activity seen in the swim test derive from this. However, in addition to the likelihood that this is an oversimplification, measurements directed at this question have not consistently found that SwHi rats are more emotional/anxious than SwLo rats. In both S5 and S14, an "anxiety assessment" test that measures drinking behavior in a small open field [described by Stout and Weiss (28)] was carried out with male and female SwHi and SwLo rats. That the SwHi rats were more anxiety prone than the SwLo rats was indicated by results for animals of S5 (SwHi rats took longer to initiate drinking and drank less in the small open field than SwLo rats), but similar findings occurred only in the male rats of S14, with female rats of S14 tending to show the opposite—female SwHi rats of S14 drank sooner, consumed more water, and defecated less in this test than did female SwLo rats of that generation. Also, stress-induced elevation of plasma corticosterone, an indicant of emotionality and/or anxiety, has been assessed in various generations (S4, S5, S13). In early generations, SwHi rats showed somewhat larger elevations than did SwLo rats, but in the most extensive (and recent) study that examined this measure, no differences at all were seen between SwHi and SwLo rats of S13 (31). In summary, although SwHi rats have shown the tendency to be more emotional and/or anxious than SwLo rats in various generations, differences between SwHi and SwLo rats in this regard clearly wax and wane across different generations, and thus differences between SwHi and SwLo rats along an "emotionality/anxiety" continuum are variable and not well correlated with differences in swim-test motor activity.

PART THREE: EFFECT OF ANTIDEPRESSANTS ON SWIM-TEST ACTIVITY OF SwLo RATS

In the widely used screening technique for antidepressants developed by Porsolt and colleagues (21), rats are placed into the swim tank in which they become increasingly inactive over the course of a 15-min exposure to the tank. When these animals are again placed into the tank on the following day and their activity is measured in a 5-min test, they are more inactive than they were the day before. Effective antidepressant medication given between the first and second exposure to the water tank attenuates the inactivity that animals otherwise show on the second exposure. Thus, effective antidepressant treatments increase activity in the Porsolt swim test.

The studies described in this part were conducted to determine whether effective antidepressant drugs would also increase swim-test activity of the SwLo animals that were bred to show low activity in the swim test. Because their basal level of activity is very low, these animals were not preexposed to the swim tank, but instead, were tested on their first exposure to the tank. This would seem to have certain advantages over the procedure described in the previous paragraph. For example, these conditions readily permit drugs to be administered for prolonged periods prior to the test (i.e., 2–4 weeks), thereby allowing one to duplicate treatment periods that are clinically effective when antidepressant drugs are given to humans if this is desired. The first two studies reported below tested effects of desipramine hydrochloride (DMI), widely known to be effective in treating human depression [see review (14)], when this drug was administered orally (first study) or by constant infusion via SC minipump (second study). The third study tested effects of a different type of antidepressant, a monoamine oxidase inhibitor (phenelzine), and, for comparison, the anxiolytic buspirone (2,10) as well as DMI.

METHOD

Animals

Subjects were male SwLo rats of S8, S11, and S12 which weighed 400–500 g at the time experiments were conducted.

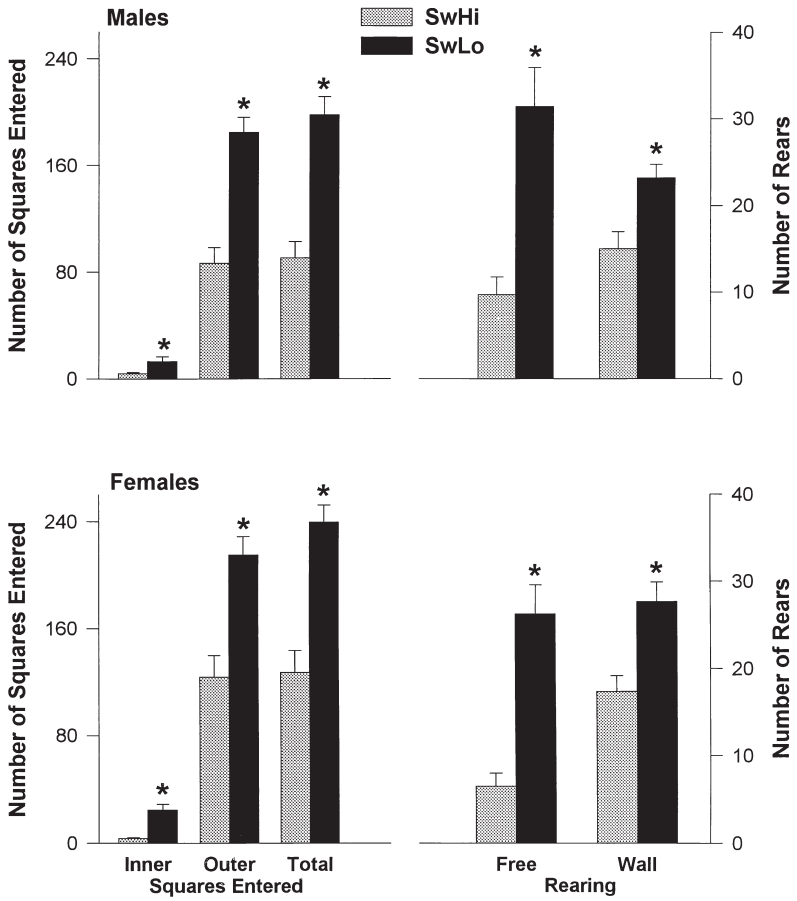

FIG. 8. Open-field ambulatory activity and rearing behavior for S14 male and female SwHi and SwLo rats. All other descriptions as in Fig. 7.

Antidepressant Drug Administration and Swim Test

In the first study, SwLo rats of S8 were given desipramine hydrochloride (DMI; Sigma) in their drinking water. The target dose was 10 mg/kg/day. Because animals varied their water intake, the concentration of DMI in the drinking water of each animal was adjusted every other day based on intake of that animal during the preceding 2-day period to maintain intake of 10 mg/kg/day. Following 1, 14, or 28 days of drug intake, animals were given the swim test and activity measured as described previously. The animals tested after 1 day or 14 days of drug treatment were given the swim test once; animals that received drug for 28 days before the test had drug treatment terminated after this test (i.e., subsequently received normal tap water) and were then retested 3 weeks later to determine whether any effect would remit when the drug was removed. A group of control animals was given standard tap water to drink; these animals were simply removed from the home cage and given the swim test. These animals were also retested 3 weeks after the initial test for comparison to the animals that were given drug for 28 days. Ten SwLo rats were used in each of the four groups used in the study.

In the second study, SwLo rats of S11 received DMI via a subcutaneously implanted Alzet minipump. Minipumps were used to avoid repeated handling and injection of animals to administer drug chronically. Daily injection of vehicle in studies of antidepressant drugs is apparently a stressful procedure, as this has been observed to produce, in comparison to noninjected controls, (a) elevated brain norepinephrine release as measured by microdialysis (E. Abercrombie, unpublished data; personal communication), and (b) increased stress-sensitive tumor growth (11). Drug was dissolved in 0.9% saline, loaded into minipumps (Model 2ML1, Alza Corp., Palo Alto, CA), and these minipumps were implanted subcutaneously in the rear flank under pentobarbital anesthesia. Animals received either 2.5, 5.0, or 10.0 mg/kg/day and were then given the swim test 1, 7, 14, or 28 days later. In that the Alzet 2ML1 minipump delivers 10 μl/h for 7 days, animals that received drug for more than 7 days (i.e., 14- and 28-day administration) were reanesthetized after each 7-day interval, the indwelling pump removed, and a new pump inserted at that time. As in the first study, different animals were tested at each time point, so that a total of twelve different groups received DMI in this study. Control animals were implanted with minipumps containing only the 0.9% saline vehicle, and a separate group of control animals was tested at each time point described above. Eight SwLo rats were used in each of the 16 groups of this study.

In a third study, SwLo animals of S12 were given buspirone (20 mg/kg/day) or phenelzine (10 mg/kg/day) via minipump while additional animals also received DMI (10 mg/kg/day) for comparison. Control animals again received 0.9% saline

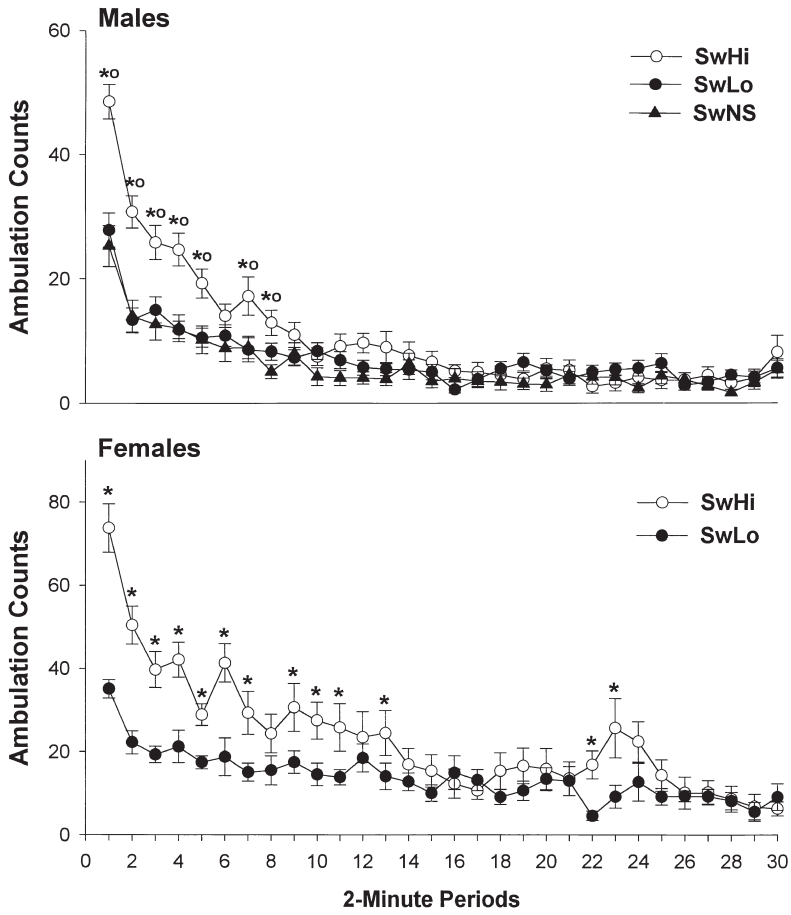

FIG. 9. Exploratory ambulatory activity in a novel home cage-like situation for S13 male (SwHi, SwLo, and SwNS) and female (SwHi and SwLo) rats. Recording was carried out for 1 h and the mean (and standard error) for each 2-min period is shown. *Significantly different (p at least < 0.05) from SwLo rats. ○ = significantly different (p at least < 0.05) from SwNS rats.

via minipump. Groups of animals were given the swim test 1, 14, and 28 days after drug administration was begun. As in the previous study, Alzet 2ML1 minipumps were used, and thus pumps were replaced every 7 days for the period of drug administration. As previously, different groups of animals were tested at each time point. Eight SwLo rats were used in each of the 12 groups in this study, except for the control groups tested at 14 and 28 days which were made up of 11 rats each.

Statistical Analysis

Each of the measures taken in the swim test was analyzed by an analysis of variance: for the first study (oral intake of DMI), one-way analyses were used; for the second study, two-way analyses were used, with drug dose (vehicle, 2.5, 5.0, and 10.0 mg/kg DMI) and days of treatment prior to testing (1,7, 14,30) as main effects; and for the third study, two-way analyses were used, with drug (vehicle, DMI, buspirone, phenelzine) and days of treatment prior to testing (1,14,30) as main effects. When a significant main effect of either drug duration (first study), drug dose (second study), or drug type (third study) was found, protected t-tests were then used to compare each of the drug-treated groups with the vehicle-treated animals; confidence levels for such comparisons are reported in the text and figures. For the second and third studies, these comparisons were conducted within the same duration of drug administration, so that each drug-treated group was compared with the vehicle-treated animals of the same duration of administration prior to testing. Comparison of the two groups on the retest (first study) was done by simple t-test.

RESULTS AND DISCUSSION

Figure 11 shows the results in the swim test after SwLo rats had received DMI (10 mg/kg/day) in their drinking water for 1, 14, or 28 days prior to testing. Acute treatment with DMI (1 day) had no effect on swim-test activity of these rats (struggling or floating); however, prolonged treatment (14 or 28 days) significantly increased struggling of these animals in the swim test. Floating was somewhat decreased after 14 and 28 days of DMI treatment, but this change did not reach statistical significance. Activity score was significantly increased following 28 days of treatment with DMI. When animals that had received DMI in their drinking water for 28 days were no longer given the drug and were then retested in the swim tank three weeks later, these animals reverted to showing a low

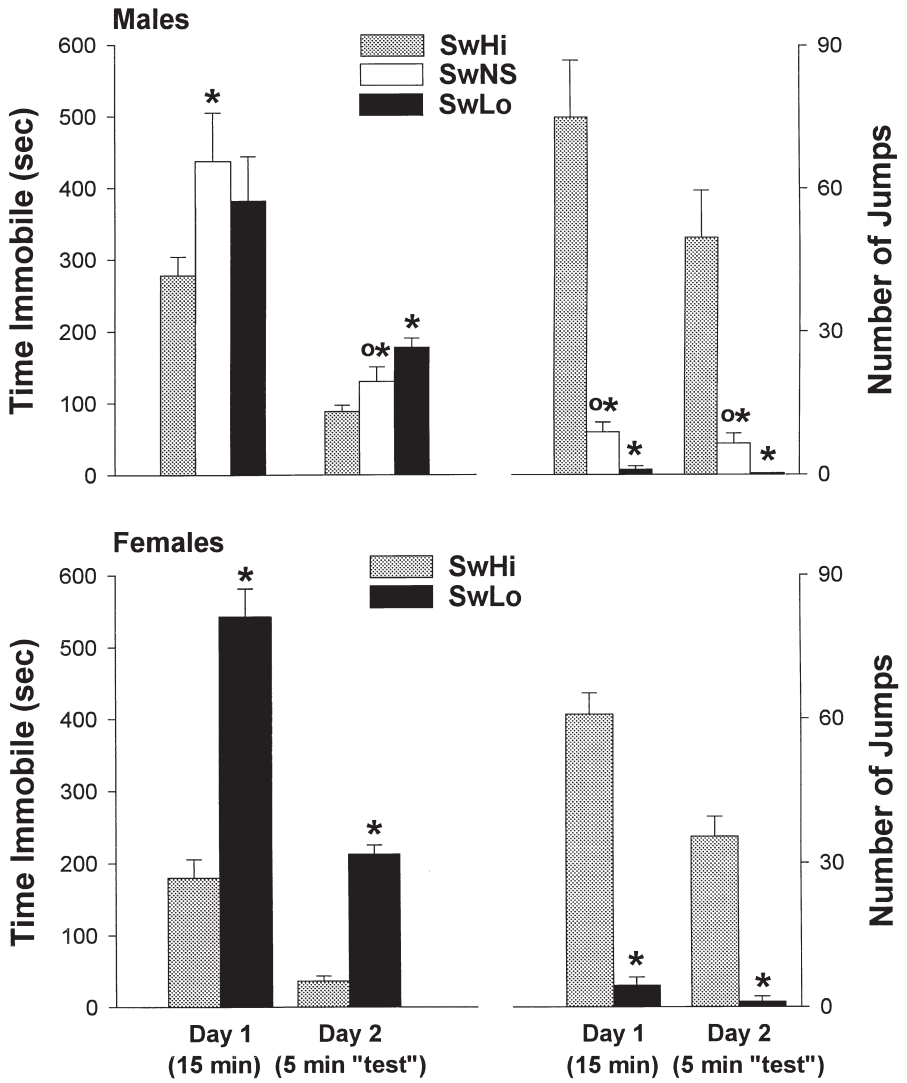

FIG. 10. Activity in a Porsolt-type test shown by S13 male (SwHi, SwLo, and SwNS) and female (SwHi, SwLo) rats. All animals were placed in this swim tank for 15 min on day 1 and returned to the swim tank for a 5-min "test" on day 2. At left is shown mean (and standard error) immobility time (in seconds) on both days of exposure to the tank, and at right is shown mean (and standard error) number of jumps observed. *Significantly different (at least $p < 0.05$) from SwHi rats; ○ = significantly different (p at least < 0.05) from SwLo rats.

level of motor activity in the swim test. Control animals (i.e., animals that received tap water) showed a similar low level of activity as that seen on their first test when retested after 3 weeks.

Figure 12 shows results from testing of SwLo rats that had been given DMI via osmotic minipumps (2.5, 5.0, or 10.0 mg/kg/day) for 1, 7, 14 or 28 days prior to testing. As seen with DMI given in the drinking water, acute treatment (1 day) produced no effect, and this was seen regardless of dose. Nor did 7 days of treatment significantly affect swim-test activity, although a small increase in struggling was seen in the 10.0 mg/kg group at this time. However, treatment with DMI for 14 and 28 days significantly increased struggling behavior, which was seen in animals given either 5.0 or 10.0 mg/kg/day. The 2.5 mg/kg dose was ineffective at all test days.

Figure 13 shows the results from testing animals that were given phenelzine, buspirone, or DMI. As was found previously, DMI increased struggling behavior of SwLo rats when given for 14 or 28 days. Phenelzine also increased swim-test activity of these animals, but did so in a manner different from DMI—when given for 28 days, phenelzine markedly reduced their floating behavior while having little effect on struggling. As a result of the reduced floating from 28 days of phenelzine administration, activity scores were also significantly increased by phenelzine at this time point. Buspirone, in contrast to DMI and phenelzine, decreased swim-test activity. Buspirone significantly increased floating after 14 days of administration, producing significantly lower activity scores at this time point as well. Buspirone also decreased struggling behavior when analyzed across the three durations of administration.

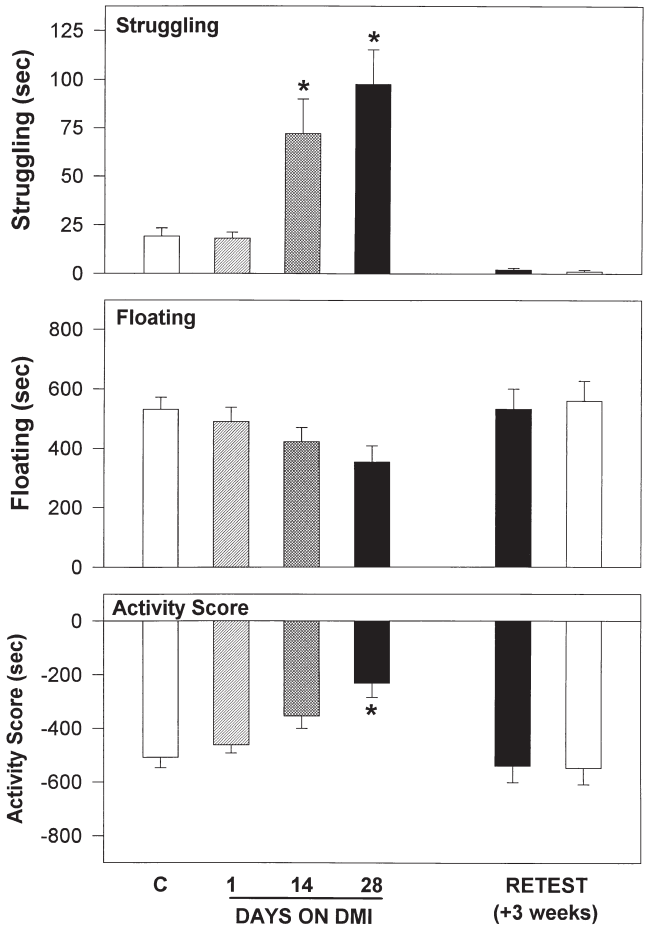

FIG. 11. Time (in seconds) spent struggling (top) and floating (middle) and activity score (bottom) in the swim test for S8 male SwLo rats that had been given DMI (approximately 10 mg/kg/day) in their drinking water for 1, 14, or 28 days prior to the swim test. Control animals (C) received regular tap water at all times prior to the test. Also shown are results from a retest of the control animals and those given tap water after having been tested following receipt of DMI for 28 days; the retest was given 3 weeks after first test. Means and standard errors are shown. *Significantly different (p at least < 0.05) from control group.

GENERAL DISCUSSION

Consideration of the Physical Characteristics of Swim High-Active and Swim Low-Active Rats

The results presented above show that it was possible to successfully breed Sprague–Dawley rats for high or low activity in a swim test; these lines have been named Swim High-Active (SwHi) and Swim Low-Active (SwLo). Moreover, findings described in Part Three above, and also reviewed below, indicate that these animals may be potentially useful for studying processes underlying depression. However, before this discussion proceeds further, a basic question needs to be answered relating to the general physical properties of the two lines of animals, and whether there are important differences between these lines in this regard. After all, one possibility by which animals that show different amounts of motor activity might be generated through selective breeding would be to inadvertently produce animals that show progressively larger differences in robustness and health with each successive generation. Specifically, perhaps SwLo rats are feeble in relation to the SwHi rats, and the large differences in swim-test activity that occurred were based on the breeding of small/weak rats vs. large/strong rats.

Over the years that the breeding program described above has continued, a variety of measures have been taken and tests have been conducted that indicate that the differences in swim-test activity seen in SwHi and SwLo rats are not due to superficial characteristics such as the physical size or robustness/health of the animals. For instance, although body weight of animals was often noted, it was carefully monitored in conjunction with several experiments. In the S5 and S14 generations, body weight and food/water intake were measured over the course of the study that assessed daily spontaneous ambulatory activity of SwHi and SwLo rats in the home cage, the results of which were reported in Part Two. In the S5 generation, over the 10-day period of the study (i.e., age 70 to 80 days for the rats), the mean ($\pm$ standard error) for body weight (in grams) was as follows: for female SwHi—(day 1) 254.6 $\pm$ 8.5 to (day 10) 268.8 $\pm$ 7.6; SwLo—(day 1) 253.2 $\pm$ 9.2 to (day 10) 269.4 $\pm$ 9.5; for male SwHi—(day 1) 362.0 $\pm$ 18.8 to (day 10) 387.8 $\pm$ 7.4; SwLo—(day 1) 366.8 $\pm$ 11.9 to (day 10) 397.0 $\pm$ 15.0. Also, these SwHi and SwLo rats did not differ in the amount of food eaten daily during the study (although it was found that SwHi rats did drink somewhat more water than SwLo rats). Similar findings were observed in S14. Thus, size, weight gain, and food intake for SwHi and SwLo rats were quite similar in the S5 and S14 generations through the period that ambulatory activity of these rats was studied. SwHi and SwLo rats have also been found to be similar in weight at older ages, and at full growth. In S16 and S17, a large study involving delivery of antidepressant drugs via minipump to nearly 400 male rats was undertaken (32). At approximately 100 days of age when animals were implanted with the minipumps, the mean body weight of these animals was: for male SwHi—463.7 $\pm$ 5.4 and SwLo—455.8 $\pm$ 3.1. In S13, approximately 40 rats of each line underwent behavioral tests described in Part Two when the animals were 7.0–8.5 months of age, essentially full growth. Mean body weights at that time were: for female SwHi—324.5 $\pm$ 3.8 and SwLo—325.7 $\pm$ 4.2; for male SwHi 571.5 $\pm$ 7.6 and SwLo 562.2 $\pm$ 5.2. In summary, measurement of body weight and food intake of different generations of SwHi and SwLo rats indicates no differences in the physical size of these animals that might account for the differences in motor activity that are seen.

The behavioral measures reported in Part Two also seem relevant to this discussion. That certain types of activity—daily spontaneous ambulation in the home cage and open-field ambulation and rearing—are either similar for SwHi and SwLo animals or, in the case of open-field ambulation and rearing, actually greater for SwLo rats than SwHi rats argues against the presence of significant differences in robustness/strength/health in the two lines. The fact that differences in motor activity shown by SwHi and SwLo rats are specific to particular testing situations, taken together with the body weight and intake data summarized in the previous paragraph, indicates that differences in activity shown by the two lines are mediated by specific physiological differences—such as differences in brain systems—rather than by any differences in body size, robustness, or general health of the animals.

Regarding Other Projects That Have Used Selective Breeding Techniques

The program reported here is not the first to use selective breeding to attempt to develop animal populations that may be

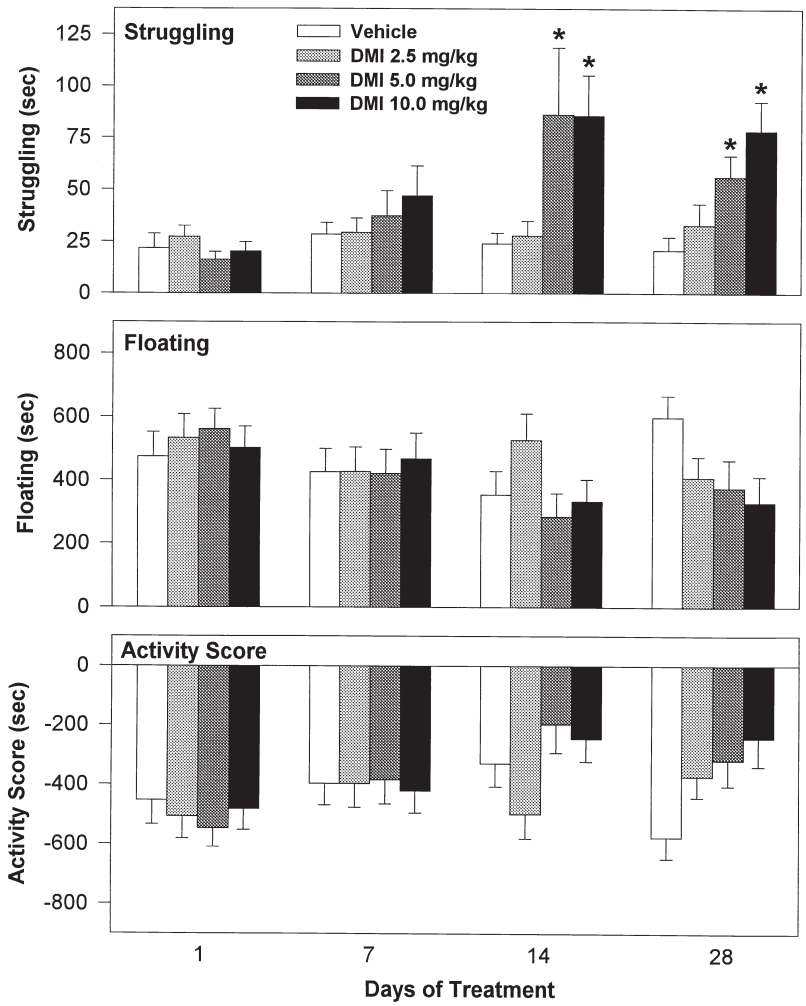

FIG. 12. Time (in seconds) spent struggling (top) and floating (middle) and activity score (bottom) for S11 male SwLo rats that had been given vehicle or DMI (2.5, 5.0, or 10.0 mg/kg) via subcutaneously implanted minipumps for 1, 7, 14, or 28 days prior to the swim test. Means and standard errors are shown. *Significantly different (p at least $<$ 0.05) from the vehicle-treated animals tested at same day of treatment.

relevant to depression. Overstreet (17) has developed an animal model of depression by selectively breeding animals for sensitivity to cholinergic agonists. These Flinders-sensitive (or S-line) rats, which respond to muscarinic agonists with behavioral depression, also show immobility in a forced swim test when compared to Flinders-resistant (or R-line) rats (17,18). Overstreet (19) has provided considerable data to support the contention that the Flinders-sensitive line of rats constitutes a model of depression. Henn and colleagues (13) have also selectively bred rats that may be relevant to depression; they bred rats on the basis of their showing poor acquisition of an active shock-escape response after having been exposed to uncontrollable shock, a phenomenon that has been called "learned helplessness." For a similar purpose, our group has also selectively bred animals that were either susceptible or resistant to reduction of motor activity after exposure to uncontrollable shock (26). It will be of interest to see whether any of these breeding programs generates animals that make genuine contributions either to our knowledge of the pathophysiology of depression or to the development of new therapies for depression.

In the selective breeding study reported above, it is noteworthy that the breeding process produced animals that showed extreme responses in both directions. When 7th and 13th generation SwHi and SwLo rats were compared with nonselected, randomly bred rats of the same generation, it was evident that the swim-test activity of both SwHi and SwLo rats had moved away, in opposite directions, from that of the original population. Perhaps the most widely known experiment in selective breeding for a behavioral characteristic is that which produced the Maudsley Reactive and Nonreactive strains (6, 7); these strains were bred for differential defecation in the open field, a measure thought to reveal differences in "emotionality." The Maudsley strains are labeled "Reactive" and "Nonreactive," but in this case only one strain, the Nonreactives, actually differ significantly from the original population (4). It can be noted that, in contrast to the Maudsley strains, both the SwHi and SwLo lines described here each differ markedly from nonselected animals; moreover, two lines each now contain extreme individuals whose scores fall outside the range seen in a nonselected population. Similar bidirectional

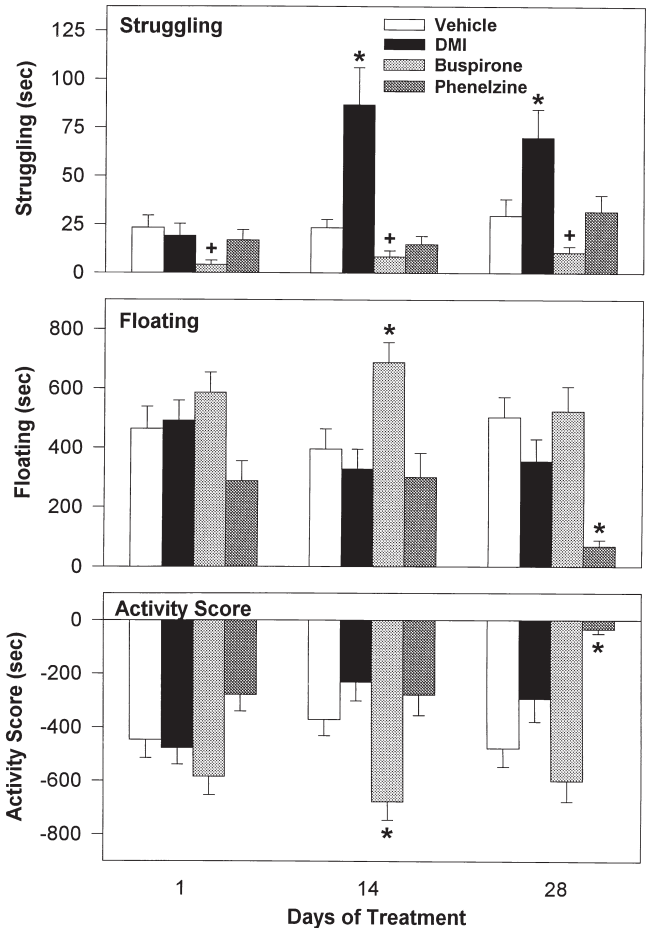

FIG. 13. Time (in seconds) spent struggling (top) and floating (middle) and activity score (bottom) in the swim test for S10–S12 male SwLo rats that had been given vehicle, DMI (10 mg/kg/day), buspirone (20 mg/kg/day), or phenelzine (10 mg/kg/day) via subcutaneously implanted minipumps for 1, 14, or 28 days prior to the swim test. Means and standard errors are shown. *Significantly different (p at least < 0.05) from the vehicle-treated animals tested at same day of treatment. $^{+}$Significant difference ($p < 0.05$) of the buspirone-treated condition from the vehicle-treated condition across all durations of administration.

differences as that reported here have been found in other selective breeding projects using the rat (3) and the mouse (8).

Concerning Effects of Antidepressant Drugs on SwLo Rats

Treatment of SwLo rats with DMI had a marked effect when the drug was administered for 2 or 4 weeks, but DMI had no detectable effect when given for only 1 day and very little effect after 7 days. This delayed response is similar to the time course of recovery from depression that occurs with use of this drug clinically; the drug does not produce a therapeutic effect immediately, but often requires 2 weeks of administration for this to occur (12,27). The time course of response to DMI in SwLo rats resembles more closely what is seen in clinical depression than what has been observed when DMI is screened in the standard Porsolt test. In the Porsolt procedure, the response indicative of an effective drug (decreased inactivity) becomes larger in magnitude after DMI has been administered repeatedly on a daily basis (15); however, a significant response to this drug is seen after three injections given in a 24-h period [the originally used regimen (21,23)] or even after a single administration (1), which does not fit the clinical picture of the therapeutic action of the drug. On the other hand, it should not be considered that the ability of the commonly used Porsolt test to detect antidepressant medication on acute administration of the drug is a drawback of that test, as it could be argued that the utility of any screening technique is based on the ease and rapidity with which it will detect effective antidepressant medication. Rather, what seems evident from the present results is that the use of selectively bred SwLo rats in the swim test more effectively reproduces the typical clinical response seen in human patients, at least in terms of time of onset of therapeutic change, than does the Porsolt procedure. In this sense, the use of SwLo rats in the swim test appears to be more satisfactory as a homologous model of depression (i.e., a model that tries to reproduce the characteristics of human depression) than is the randomly bred rat in the standard Porsolt test, whereas the later retains distinct advantages as a method for rapidly and inexpensively screening for potential antidepressant medication.

In addition to DMI, the MAO inhibitor phenelzine also had a positive therapeutic effect on swim-test activity when administered chronically, which again parallels therapeutic efficacy seen in patients. Most interestingly, phenelzine had its major effect on floating behavior—the drug decreased floating—rather than on struggling behavior as did DMI. Thus, the therapeutic effect of the MAO inhibitor was manifested primarily on a different aspect of behavior than was affected by the tricyclic DMI. Using a similar swim test to that described by Weiss et al. (30) (i.e., deeper water than that utilized in the typical Porsolt swim test), Detke et al. (9) also reported that different types of antidepressant medication can affect different aspects of swim-test behavior, which can be measured when deeper water is used in the swim tank. As would be expected, Detke et al. carried out their studies with normal, randomly bred Sprague–Dawley rats; thus, their results indicate that the ability to differentiate between different types of antidepressant drugs by examining different aspects of behavior in the swim test does not necessarily depend upon the use of selectively bred animals as were employed in the studies described here. It remains to be determined what the contribution of SwLo or SwHi rats will be to the analysis and therapy of depression. Toward this end, the next article (32) reports the results from the assessment of several antidepressant drugs in SwHi and SwLo rats.

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REFERENCES

1. Araki, H.; Kawashima, K.; Aihara, H.: The difference in the site of actions of tricyclic antidepressants and methamphetamine on the duration of the immobility in the behavioral despair test. Jpn. J. Pharmacol. 35:67–72; 1984.

2. Barrett, J. E.; Vanover, K. E.: 5-HT receptors as targets for the development of novel anxiolytic drugs: Models, mechanisms and future directions. Psychopharmacology (Berlin) 112:1–12; 1993.
3. Bignami, G.: Selection for high rates and low rates of avoidance conditioning in the rat. Anim. Behav. 13:221–227; 1965.
4. Blizard, D. A.: The Maudsley Reactive and Nonreactive strains: A North American perspective. Behav. Genet. 11:469–489; 1981.
5. Borsini, F.; Meli, A.: Is the forced swim test a suitable model for revealing antidepressant activity? Psychopharmacology (Berlin) 94:147–160; 1988.
6. Broadhurst, P. L.: Experiments in psychogenetics. In: Eysenck, H. J., ed. Experiments in personality, psychogenetics and psychopharmacology, vol. 1. London: Routeledge and Kegan Paul; 1960:3–102.
7. Broadhurst, P. L.: The Maudsley reactive and nonreactive strains of rats: A survey. Behav. Genet. 5:299–319; 1975.
8. DeFries, J. C.; Gervais, M. C.; Thomas, E. A.: Response to 30 generations of selection for open-field activity in laboratory mice. Behav. Genet. 8:3–13; 1978.
9. Detke, M. J.; Rickels, M.; Lucki, I.: Active behaviors in the rat forced swimming test differentially produced by serotonergic and noradrenergic antidepressants. Psychopharmacology (Berlin) 121: 66–72; 1995.
10. Eison, A. S.; Temple, D. L., Jr.: Buspirone: Review of its pharmacology and current perspectives on its mechanism of action. Am. J. Med. 80:1–9; 1986.
11. Garabal, M. F.; Nunez, M. J.; Balboa, J. L.; Suarez, J. A.; Belmonte, A.: Effects of alprazolam on the development of MTV-induced mammary tumors in female mice under stress. Cancer Lett. 62:185–189; 1991.
12. Heller, A.; Zahourek, R.; Whittington, H. G.: Effectiveness of antidepressant drugs: A triple-blind study comparing imipramine, desipramine and placebo. Am. J. Psychiatry 127:1092–1095; 1971.
13. Henn, F. A.; Johnson, J.; Edwards, E.; Anderson, D.: Melancholia in rodents: Neurobiology and pharmacology. Psychopharmacol. Bull. 21:443–446; 1985.
14. Janowsky, D. S.; Byerley, B.: Desipramine: An overview. J. Clin. Psychiatry 45:3–7; 1984.
15. Miyauchi, T.; Kitada, Y.; Satoh, S.: Effects of acutely and chronically administered antidepressants on the brain regional 3-methoxy-4-hydroxyphenylethyleneglycol sulphate in the forced swimming rat. Life Sci. 29:1921–1928; 1981.
16. Naitoh, H.; Nomura, S.; Kunimi, Y.; Yamaoka, K.: "Swimming-induced head twitching" in rats in the forced swimming test induced by overcrowding stress: A new marker in the animal model of depression? Keio J. Med. 41:221–224; 1992.
17. Overstreet, D. H.: Selective breeding for increased cholinergic function: Development of a new animal model of depression. Biol. Psychiatry 21:49–58; 1986.
18. Overstreet, D. H.; Janowsky, D. S.; Gillin, J. C.; Shiromani, P. J.; Sutin, E. L.: Stress-induced immobility in rats with cholinergic supersensitivity. Biol. Psychiatry 21:657–664; 1986.
19. Overstreet, D. H.: The Flinders sensitive line rats: A genetic animal model of depression. Neurosci. Biobehav. Rev. 17:51–68; 1993.
20. Plaznik, A.; Tamborska, E.; Hauptmann, M.; Bidzinski, A.; Kostowski, W.: Brain neurotransmitter systems mediating behavioral deficits produced by inescapable shock treatments in rats. Brain Res. 447:122–132; 1988.
21. Porsolt, R. D.; LePichon, M.; Jalfre, M.: Depression: A new animal model sensitive to antidepressant treatments. Nature 266:730–732; 1977.
22. Porsolt, R. D.; Anton, G.; Blavet, N.; Jalfre, M.: Behavioral despair in rats: A new model sensitive to antidepressant treatments. Eur. J. Pharmacol. 47:379–391; 1978.
23. Porsolt, R. D.; Bertin, A.; Blavet, N.; Deniel, M.; Jalfre, M.: Immobility induced by forced swimming in rats: Effects of agents which modify central catecholamine and serotonin activity. Eur. J. Pharmacol. 57:201–210; 1979.
24. Prince, C. R.; Anisman, H.: Acute and chronic stress effects on performance in a forced-swim task. Behav. Neural Biol. 42:99–119; 1984.
25. Riley, V.; Fitzmaurice, M. A.; Spackman, D. H.: Psychoneuroimmunologic factors in neoplasia: Studies in animals. In: Ader, R., ed. Psychoneuroimmunology. New York: Academic Press; 1981: 31–47.
26. Scott, P. A.; Cierpial, M. A.; Kilts, C. D.; Weiss, J. M.: Susceptibility and resistance of rats to stress-induced decreases in swim-test activity: A selective breeding study. Brain Res. 725:217–230; 1996.
27. Stewart, J. A.; Mitchell, P. H.: A comparative trial of desipramine and nortriptyline in depression. Br. J. Psychiatry 114:469–471; 1968.
28. Stout, J. C.; Weiss, J. M.: An animal model for measuring behavioral responses to anxiogenic and anxiolytic manipulations. Pharmacol. Biochem. Behav. 47:459–465; 1994.
29. Walsh, R. N.; Cummins, R. A.: The open-field test: A critical review. Psychol. Bull. 83:482–504; 1976.
30. Weiss, J. M.; Goodman, P. A.; Losito, B. G.; Corrigan, S.; Charry, J. M.; Bailey, W. H.: Behavioral depression produced by an uncontrollable stressor: Relationship to norepinephrine, dopamine, and serotonin levels in various brain regions. Brain Res. Rev. 3:167–205; 1981.
31. West, C. H. K.; McCurdy, P. M.; Bonsall, R. W.; Weiss, J. M.: Behavioral and biochemical responses to stress in rats selectively bred for swim test activity. Soc. Neurosci. Abstr. 19:1243; 1993.
32. West, C. H. K.; Weiss, J. M.: Effects of antidepressant drugs on animals bred for low activity in the swim test. Pharmacol. Biochem. Behav. 61:67–79; 1998.