Pharmacology Biochemistry and Behavior, Vol. 65, No. 3, pp. 389–398, 2000

0091-3057/00/$–see front matter

PII S0091-3057(99)00210-5

Test Conditions Influence the Response to a Drug Challenge in Rodents

ANDREW HARKIN, JOHN P. KELLY, JOHN FRAWLEY, JAMES M. O'DONNELL, AND BRIAN E. LEONARD

Department of Pharmacology, National University of Ireland, Galway, Ireland

Received 7 April 1999; Revised 1 September 1999; Accepted 3 September 1999

HARKIN, A., J. P. KELLY, J. FRAWLEY, J. M. O'DONNELL AND B. E. LEONARD. *Test conditions influence the response to a drug challenge in rodents.* PHARMACOL BIOCHEM BEHAV **65**(3) 389–398, 2000.—These studies were conducted to examine the differential response to a drug challenge under varied experimental test conditions routinely employed to study drug-induced behavioral and neurophysiological responses in rodents. Apomorphine, a nonselective dopamine agonist, was selected due to its biphasic behavioral effects, its ability to induce hypothermia, and to produce distinct changes to dopamine turnover in the rodent brain. From such experiments there is evidence that characterization and detection of apomorphine-induced activity in rodents critically depends upon the test conditions employed. In rats, detection of apomorphine-induced hyperactivity was facilitated by a period of acclimatization to the test conditions. Moreover, test conditions can impact upon other physiological responses to apomorphine such as drug-induced hypothermia. In mice, apomorphine produced qualitatively different responses under novel conditions when compared to those behaviors elicited in the home test cage. Drug-induced gross activity counts were increased in the novel exploratory box only, while measures of stereotypic behavior were similar in both. By contrast, apomorphine-induced locomotion was more prominent in the novel exploratory box. Dopamine turnover ratios (DOPAC:DA and HVA:DA) were found to be lower in those animals exposed to the exploratory box when compared to their home cage counterparts. However, apomorphine-induced reductions in striatal dopamine turnover were detected in both novel and home cage environments. The implications of these findings are discussed with particular emphasis upon conducting psychopharmacological challenge tests in rodents. © 2000 Elsevier Science Inc.

Environment Rats Mice Apomorphine Challenge Locomotor activity Stereotypy Hypothermia Striatum Dopamine turnover

MEASURING spontaneous behaviors is widely applicable to the study of drug action in laboratory animals. The present series of experiments were aimed at a characterization of drug-induced behavioral activity and how the behavioral response to a given drug differs, depending on the experimental design and test environment.

In the present study, a combination of classification by observation and automatic recording was adopted as an appropriate way to study stereotyped and locomotor behaviors produced by the dopamine agonist, apomorphine (12). Apomorphine is used extensively as a reference drug in preclinical pharmacology (14); evoking a complex locomotor and stereotypic response [at low doses ($\leq$0.1 mg/kg) it reduces locomotor activity (6), while at higher doses ($\geq$0.3 mg/kg) it produces hypermotility and stereotypies (29), reducing body temperature (4) and producing distinct neurochemical responses in rodent brain (23). With apomorphine, we were thus able to study a broad spectrum of drug-induced changes under varied experimental conditions. Drug effects were monitored in two species—the mouse and the rat—and in contrasting automated monitoring systems—an automated home-cage activity monitor, and an automated novel exploratory box. Direct comparisons could then be made between the spontaneous behavior of animals in both tests, alone and in response to the psychostimulant, apomorphine. Such studies are of importance and relevance to studies of general activity, exploration or investigatory behavior, where a major difficulty has been to distinguish between activity related to an animals' level of arousal, activity elicited by external environmental stimuli, and drug-induced activity (10).

Test conditions may not only influence behavior of the animals but can also have an effect on physiological parameters

Requests for reprints should be addressed to Andrew Harkin, Ph.D., Department of Pharmacology, National University of Ireland, Galway, Ireland.

(11,30). In conjunction with the detection of the behavioral response we examined the effect of test conditions on apomorphine-induced reduction in body temperature, which has been previously documented in rodents (4). Apomorphine produces a dose-dependent reduction in striatal dihydroxyphenylacetic acid (DOPAC) and homovanillic acid (HVA) concentrations in rats (23). In the present experiment, dopamine turnover in response to apomorphine administration was measured in mice following exposure to both novel and home cage test conditions to study whether changes to the animals environment might impact upon striatal dopamine activity, alone and in response to apomorphine.

Behavioral, hypothermic, and neurochemical responses to vehicle and apomorphine challenge were characterized and detected under varied test conditions, and were shown to be influenced by the test environment. Activity measures including locomotion were found to differ significantly between novel and home-cage test conditions. Moreover, apomorphine-induced hypothermia and striatal dopamine turnover were affected by exposure to the different situations. Although there is a great deal of literature published on apomorphine, there is a paucity of data on the influence that test conditions may have on the response to apomorphine or other psychoactive drugs. The interest of this study is that this series of behavioral and neurochemical investigations are made by the same group of investigators in the same laboratory and with well-defined experimental conditions, so that the data presented in the manuscript may be viewed as a second comparative reference set of data on the behavioral effects of apomorphine. In addition, a great deal of emphasis in this study is placed upon how the drug-induced changes differ depending upon the experimental design and test environment. The results outline appropriate conditions in which particular responses to drug challenge may be successfully detected following the administration of central stimulant or depressant drugs. The influence that test conditions and experimental design have in drug challenge tests are often understudied. The importance of assessing the effects of drug challenge under appropriate experimental conditions is emphasized.

METHOD

Subjects

Male Sprague–Dawley rats obtained from Harlan Olac, U.K. (weight on arrival: 230–250 g) and male CD-1 mice obtained from Biological Laboratories (Europe Ltd.), Ballina, Ireland (weight 25–30 g) were used in the present series of studies. The rats were housed four per cage in plastic-bottomed cages (45 × 25 × 20 cm). The mice were housed 10–15 in similar cages to those used for the rats. All animals were allowed free access to food and water except during periods of experimentation. Lighting was controlled on a 12 L:12 D cycle (lights on: 0800 h, lights off: 2000 h); temperature was maintained at 20 ± 2°C, and relative humidity at 50–80%. All experimental testing was conducted between 1200 and 1700 h. All procedures were carried out under the guidelines of the Animal Welfare Committee of the National University of Ireland, Galway, and in accordance with the European Communities Council Directive 1986 (86/806/EEC).

Preparation and Administration of Apomorphine

Apomorphine hydrochloride was freshly prepared prior to the injections. It was dissolved in a vehicle containing 0.89% NaCl with 1 mg/ml ascorbic acid added as an antioxidant. All injections to rats were made in a volume of 1 ml/kg bodyweight. Injections to mice were made in a volume of 10 ml/kg. Control animals were treated with vehicle in the same dose volumes as the test groups. All injections were given subcutaneously.

Measuring Behavior Following a Challenge Injection of Apomorphine in the Home Cage

Home cage activity was measured by placing animals singly in a standard plastic-bottomed cage. Cages were placed individually in a home cage activity monitor. Behavior was monitored automatically with passive infrared sensors mounted above the floor of the test cage. The term activity measured by such techniques incorporates all aspects of activity that could be measured by interruption of a sensor beam. As the beams cross the floor of the chambers, behaviors primarily including exploratory behavior and ambulation are cumulated in activity counts. Separate rating scales were introduced for quantifying apomorphine-induced locomotor activity and stereotyped behaviors so that these behaviors could be assessed independently. The most commonly used measures of stereotyped behaviors are based on time sampling and observational recordings, which have been employed in previous investigations (8,12,24,27). As continuous recording of behavior is difficult over extended periods, especially when the behavior is complex and rapidly changing, time sampling methods with interval recording and momentary sampling provide appropriate estimates of the occurrences and duration of particular behaviors. The following were employed as rating scales to score the stereotyped behavior and locomotor response to a challenge injection of apomorphine.

Stereotypy scale [see (8,12,24,27)]. 0 — inactive, asleep; 1 — awake, stationary; 2 — locomotion with some sniffing, rearing, or grooming. Sniffing was defined as rhythmic movement of the snout and head along the cage wall or floor, accompanied by rapid movement of vibrissae. Rearing was defined as when the animal raised both fore paws from the floor of the test area. Grooming is often a discontinuous process broken at intervals by locomotion and exploratory activity. It consisted of initial head and snout grooming with the front paws followed by burrowing of the snout into the body; 3 — continuous sniffing and rearing over a wide area; 4 — continuous sniffing in one location; 5 — chewing, biting/gnawing, licking, and repetitive head movements in more than one location. Chewing was defined as jaw movement not directed at any stimulus. Gnawing was defined as where the wire of the cage, edge of the Perspex or the bedding on the floor was gripped between the teeth. Licking was defined as tongue protrusion against the cage floor or wall; 6 — continual compulsive biting, chewing, or licking and repetitive head movement in one location and without interruption.

Stereotypies that occurred regularly and were not interrupted for more than 5 s were regarded as continuous.

Locomotion scale [see (27)]. 0 — inactive, asleep; 1 — awake, stationary; 2 — normal (some locomotion with some sniffing, rearing, and grooming); 3 — rapid and continuous locomotion; and 4 — rapid locomotion with leaping, jumping, or rapid darting around the perimeter of the test cage.

Animals were observed for 30 s and scored once every 10 min.

Measuring Behavior Following a Single Injection of Apomorphine in a Novel Exploratory Box

The exploratory box apparatus consisted of four circular Perspex arenas (50 cm in diameter), which provide an annular

area (12.5 cm wide, 35 cm high) for exploration. Three infrared photoswitches consisting of separate beam transmitters and receivers were mounted symmetrically (60°) outside the arena. An infrared beam was transmitted throughout the arena. The annular area was thus crossed by six beams. Upon beam interruption, the activity was registered as a single count. The software allowed for clockwise and anticlockwise locomotion. On the day of testing the animals were placed in one of four test arenas and their activity monitored over the given period (7). Illumination was provided by a 60-W bulb mounted 1 m above the floor of the apparatus.

The same scales as those employed to score the stereotyped behavior and locomotor response to a challenge injection of apomorphine in the home cage were used for monitoring behavior in the exploratory box. Animals were observed for 30 s and scored once every 10 min.

Hypothermic Response to Apomorphine

The colonic temperature was recorded by means of a digital thermometer. The probe was inserted 2 cm into the colon of the rat. Temperatures were recorded prior to and 30 min following an injection of apomorphine. For these studies, the time selected was based on previous dose–response experiments. Thirty minutes postchallenge produces an intensity of response that allows us to measure peak effects in the response to a single injection of apomorphine.

Animal Decapitation and Brain Dissection

Mice were sacrificed by decapitation 40 min following the acute apomorphine challenge. This time point was chosen as it overlaps with peak behavioral effects of apomorphine. The brains were rapidly removed, and the striatum was dissected on an ice cold plate using a sharp dissection blade, weighed, homogenized by sonication in 1.0 ml elution buffer (pH 2.8) containing 2 ng/50 μl *N*-methyl-dopamine as an internal standard and stored at −20°C prior to assay.

Determination of Brain Biogenic Amine Concentrations

Concentrations of dopamine, dihydroxyphenylacetic acid, and homovanillic acid were measured by high-performance liquid chromatography with electrochemical detection (25). The mobile phase contained 0.1 M citric acid, 0.1 M sodium dihydrogen phosphate, 1.4 mM octane-1–sulfonic acid, 0.1 mM ethylendiaminetetra–acetic acid, and 9% v/v methanol. The pH of the mobile phase was adjusted to 2.8 with concentrated NaOH. The retention times of biogenic amines varied between 5 to 40 min on an LI Chrosorb RP-18 column. The flow rate of the mobile phase through the column was 1 ml/min at a pressure of approximately 200 bar. The column oven was maintained at 30°C. All standards were purchased from Sigma Chemical Company (Poole, Dorset, UK).

Behavioral, hypothermic, and neurochemical responses to vehicle and apomorphine challenge were characterized and detected under varied test conditions as outlined below:

Study 1. Characterization of apomorphine-induced psychomotor activity in the home cage in rats.

Study 2. Detection of apomorphine-induced psychomotor activity in different test environments in rats: (A) Animals were singly housed and allowed to acclimatize to the test conditions overnight; (B) animals were tested in a novel cage (fresh bedding); and (C) animals were tested in a novel exploratory box.

Study 3. Effect of exposure to different test conditions on apomorphine-induced hypothermia in the rat: (A) in animals housed in their home cage; (B) in animals exposed to a novel test cage (fresh bedding); (C) in animals exposed to the exploratory box.

Study 4. Effect of apomorphine on activity in rats and mice in the home cage.

Study 5. Effect of apomorphine on activity in mice in two contrasting test environments, the home cage, and exploratory box.

Study 6. Effect of test conditions on apomorphine-induced changes to dopamine concentrations and turnover in mouse striatum. We allowed for 2 h of exposure in the home cage and exploratory boxes before apomorphine administration. Such an approach was also adopted in the behavioral studies to allow for the effects of apomorphine to be detected. In rodents behavioral effects of apomorphine (1–30 mg/kg) peak after 45 min. Striatal dopamine metabolism was determined at this point.

Statistical Analysis of Data

Data were initially analyzed using a one-way or two-way analysis of variance (ANOVA) where drug treatment and test condition or species were the first and second factors. Individual groups differences were assessed with the Fishers least significant difference (LSD) multiple range test. Data were deemed significant when $p < 0.05$.

RESULTS

Study 1: Characterization of Apomorphine-Induced Psychomotor Activity in the Home Cage in Rats (Fig. 1)

In the immediate few minutes following apomorphine administration, rats sat hunched, motionless, and sedated for several minutes following drug administration. Within approximately 5 min of drug administration hyperactivity and stereotypies were observed in all of the animals that received apomorphine (>0.3 mg/kg) and continued for approximately 1–2 h, varying with the dose of the drug. The effects were more pronounced and persistent with the higher doses.

Behavioral changes observed following treatment with the higher doses of apomorphine (>0.3 mg/kg) consisted of repetitive sniffing at the floor and walls of the cages (different from usual exploratory sniffing, which is more irregular and not continuous), burrowing movements, chewing, rearing, and head-down posture. Short periods of grooming were also evident. At doses ⩾1 mg/kg the animals displayed sudden jerking movements and bursts of running around the perimeter of the cage. At higher doses (⩾3 mg/kg) the animals were observed licking and gnawing, with bedding material between their teeth.

Study 2: Detection of Apomorphine-Induced Psychomotor Activity in Different Test Environments in Rats (Table 1)

ANOVA of home cage activity following a period of acclimatization to the test conditions displayed significant effects of apomorphine, $F(6, 35) = 7.86$, $p < 0.001$, time $F(6, 35) = 6.93$, $p < 0.001$, and a drug × time interaction, $F(24, 140) = 3.6$, $p < 0.001$. Post hoc comparisons revealed that there was no acclimatization effects as animals had already acclimatized to the test environment. Hypoactivity with the 0.03 mg/kg dose of apomorphine was not detected at any of the time intervals measured. Significantly higher counts were found with the 1, 3, and 10 mg/kg doses in the first and second intervals measured. The hyperactive effect of the 1- and 10-mg/kg dose was also significant in the final three time intervals.

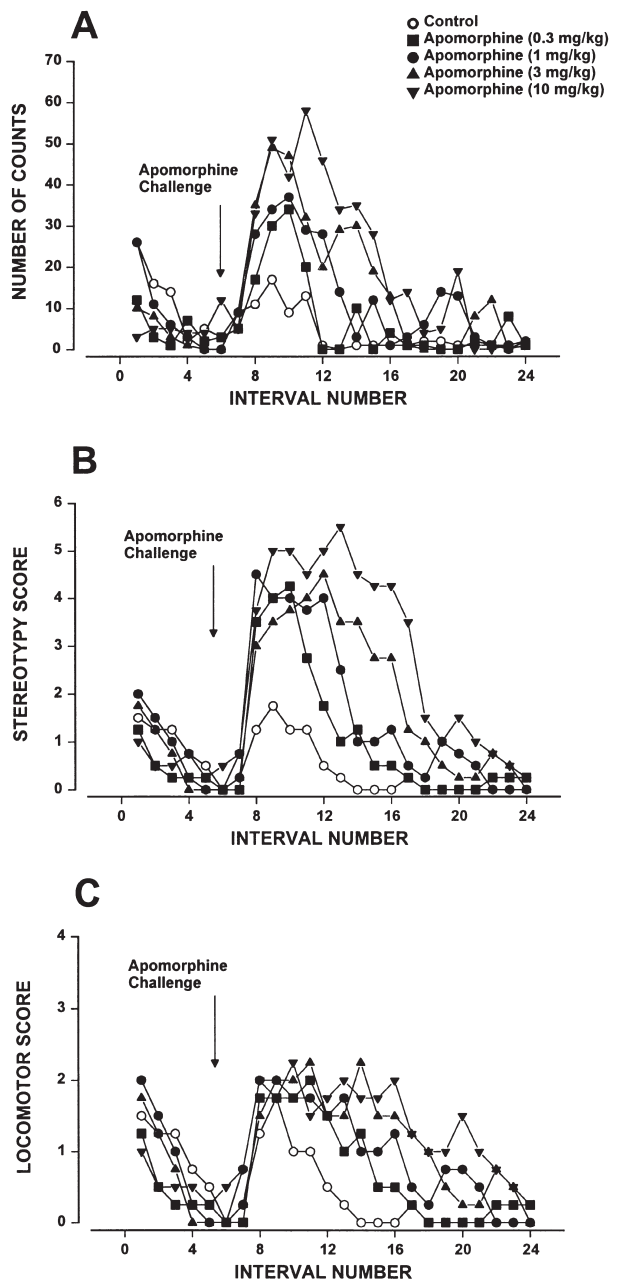

FIG. 1. Effect of apomorphine on home cage activity in rats. Animals were placed singly in a standard plastic bottomed cage with fresh bedding. Behavior was measured automatically in the home-cage activity monitor with passive infrared sensors mounted above the floor of the test cage in combination with observational recording for 1 h pre- and 3 h postchallenge with 0.3, 1, 3, and 10 mg/kg apomorphine SC. Data is represented as mean score for four animals for 24 intervals of 10 min.

ANOVA of activity upon exposure to a novel test cage displayed significant effects of apomorphine, $F(6, 35) = 7.19$, $p = 0.001$, time, $F(4, 140) = 3.09$, $p < 0.05$, and a drug $\times$ time interaction, $F(24, 140) = 3.6$, $p < 0.001$. Post hoc comparisons reveal that activity in the vehicle-treated control group reduced gradually over the 25-min test period. Activity counts in the final 4 $\times$ 5-min intervals were significantly lower than the number of counts over the first 5 min of the test period. Hypoactivity with the 0.1 and 0.03 mg/kg doses of apomorphine were significant at the first two and three time intervals measured, respectively. Significantly higher counts were found with the 0.3, 1, 3, and 10 mg/kg doses in the final time intervals measured.

ANOVA of activity in the exploratory box showed effects of apomorphine, $F(6, 35) = 4.32$, $p = 0.003$, and time, $F(4, 140) = 6.82$, $p < 0.001$. There was no interaction between drug and time, $F(24, 140) = 0.59$, $p = 0.93$. Activity in the control group reduced gradually over the 25-min test period. Activity counts in the third and fourth intervals were significantly lower than the number of counts over the first 5 min of the test period. Hypoactivity with the 0.03 mg/kg dose of apomorphine was significant at the first, second, and final time intervals. Significantly higher counts were found with the 0.3, 1, 3, and 10 mg/kg doses in the third and fourth intervals measured. The effect of 1, 3, and 10 mg/kg apomorphine reached significance in other intervals also (Table 1).

Study 3: Effect of Exposure to Different test Conditions on Apomorphine-Induced Hypothermia in the Rat (Table 2)

In animals housed in their home cage there was no difference in temperatures prior to apomorphine administration, $F(8, 36) = 0.67$, $p = 0.713$. ANOVA of the temperatures taken 30 min following apomorphine administration showed an effect of challenge, $F(8, 36) = 2.86$, $p = 0.014$. Post hoc comparisons revealed that apomorphine (1, 3, 10, and 30 mg/kg SC) reduced body temperature ($p < 0.05$). ANOVA of temperature change 30 min following apomorphine treatment showed an effect of challenge, $F(8, 36) = 3.13$, $p = 0.009$. Post hoc comparisons revealed that apomorphine (0.3, 1, 3, 10, and 30 mg/kg) reduced body temperature ($p < 0.05$).

In animals prior to exposure to a novel test cage there was no difference in temperatures prior to apomorphine administration, $F(6, 35) = 0,73$, $p = 0.632$. ANOVA of the temperatures taken 30 min following apomorphine administration showed no effect of challenge, $F(6, 35) = 0.73$, $p = 0.632$. Similarly, ANOVA of the temperature change 30 min following apomorphine treatment showed no effect of challenge, $F(6, 35) = 0.87$, $p = 0.53$.

In animals prior to exposure to the exploratory box there was no difference in temperature prior to apomorphine administration, $F(6, 35) = 0.31$, $p = 0.925$. ANOVA of the temperatures taken 30 min following apomorphine administration showed an effect of challenge, $F(6, 35) = 4.33$, $p = 0.002$. Post hoc comparisons revealed that apomorphine (0.1, 0.3, 1, 3, and 10 mg/kg SC) reduced body temperature ($p < 0.05$). ANOVA of temperature change 30 min following apomorphine treatment showed an effect of challenge, $F(6, 35) = 3.02$, $p = 0.018$). Post hoc comparisons revealed that apomorphine (0.1, 0.3, 3, and 10 mg/kg) reduced body temperature ($p < 0.05$) (Table 2).

Study 4: Effect of Apomorphine on Activity in Rats and Mice in the Home Cage (Fig. 2)

Mice receiving apomorphine behaved in a similar fashion to rats (see Study 1).

ANOVA of home-cage activity (automated counts) prior to apomorphine administration showed an effect of species, $F(1, 24) = 99.50$, $p < 0.001$. Post hoc comparisons revealed that mice had higher counts (cumulative count over 60 min) than rats during the acclimatization stage prior to drug challenge ($p < 0.01$). ANOVA of activity (cumulative score

TABLE 1

EFFECT OF APOMORPHINE ON ACTIVITY OF THE RAT UNDER DIFFERENT EXPERIMENTAL CONDITIONS

	Minute Interval				
Group	1–5	6–10	11–15	16–20	21–25
Home-cage activity following a period of acclimatization to the test conditions					
Control	9 ± 4	4 ± 2	5 ± 3	10 ± 8	2 ± 1
0.03 mg/kg	9 ± 3	5 ± 3	2 ± 2	1 ± 1	1 ± 1
0.1 mg/kg	4 ± 3	3 ± 2	1 ± 1	1 ± 1	0 ± 0
0.3 mg/kg	7 ± 3	7 ± 4	6 ± 5	4 ± 3	2 ± 2
1 mg/kg	25 ± 9*	29 ± 8†	25 ± 10†	19 ± 4	16 ± 2*
3 mg/kg	30 ± 9†	21 ± 5†	12 ± 4	13 ± 5	13 ± 6
10 mg/kg	56 ± 13†	46 ± 8†	40 ± 12†	30 ± 13†	23 ± 13†
Home-cage activity under novel test conditions					
Control	42 ± 4	24 ± 7§	16 ± 7§	9 ± 5§	8 ± 5§
0.03 mg/kg	22 ± 9†	9 ± 4*	3 ± 3*	1 ± 1	7 ± 3
0.1 mg/kg	4 ± 4†	8 ± 3†	6 ± 3	3 ± 2	3 ± 2
0.3 mg/kg	31 ± 10	23 ± 14	20 ± 12	19 ± 12	20 ± 8*
1 mg/kg	46 ± 11	49 ± 10†	66 ± 4†	66 ± 4†	52 ± 7†
3 mg/kg	34 ± 11	42 ± 15†	51 ± 12†	50 ± 8†	38 ± 9†
10 mg/kg	30 ± 9	27 ± 9	27 ± 4	26 ± 4†	27 ± 3†
Activity in the exploratory box					
Control	49 ± 18	34 ± 14	15 ± 13§	15 ± 14§	27 ± 17
0.03 mg/kg	9 ± 6†	1 ± 1†	0 ± 0	2 ± 2	1 ± 1*
0.1 mg/kg	38 ± 13	31 ± 11	34 ± 13	20 ± 9	12 ± 12
0.3 mg/kg	56 ± 11	55 ± 4	57 ± 12†	40 ± 9*	42 ± 29
1 mg/kg	81 ± 8†	70 ± 21†	65 ± 17†	62 ± 19†	49 ± 15
3 mg/kg	67 ± 21	77 ± 18†	67 ± 16†	60 ± 13†	54 ± 13*
10 mg/kg	83 ± 32†	75 ± 15†	73 ± 13†	64 ± 15†	49 ± 11

Effect of apomorphine on activity of the rat under different experimental conditions. (A) Home-cage activity following a period of acclimatization to the test conditions; (B) Home-cage activity in novel test conditions; and (C) Activity in the exploratory box. Animals were challenged with either vehicle or apomorphine (0.03–10 mg/kg SC) 5 min later the animals were exposed to the test condition of interest and activity monitored over five periods of 5-min activity. Data is expressed as mean count with standard error of the mean. There were six animals per group. *$p < 0.05$; †$p < 0.01$ vs. control; §$p < 0.01$ vs. vehicle control intervals 1–5.

for 3 h postchallenge) following apomorphine administration showed an effect of drug challenge, $F(3, 24) = 3.70, p = 0.025$. Post hoc comparisons revealed that apomorphine (3 and 10 mg/kg) increased the activity count detected with rats but not with mice ($p < 0.01$). Following the administration of apomorphine (10 mg/kg) there was a higher cumulative count in rats when compared to mice ($p < 0.01$). ANOVA of stereotypy scores prior to apomorphine administration showed an effect of species, $F(1, 24) = 114.15, p < 0.001$). Post hoc comparisons revealed that mice had higher scores (cumulative score over 60 min) than rats during the acclimatization stage prior to drug challenge ($p < 0.01$). ANOVA of stereotypy scores (cumulative score for 3 h postchallenge) following apomorphine administration showed an effect of species, $F(1, 24) = 17.99, p < 0.001$, drug challenge, $F(3, 24) = 3.70, p = 0.025$, and a species × challenge interaction, $F(3, 24) = 6.05, p = 0.003$. Post hoc comparisons revealed that apomorphine (1, 3, and 10 mg/kg) increased the stereotypy score with rats and mice ($p < 0.01$). Following the administration of apomorphine (1, 3, and 10 mg/kg) there was a higher cumulative score in rats when compared to mice ($p < 0.05$). ANOVA of locomotor scores prior to apomorphine administration showed an effect of species, $F(1, 24) = 114.15, p < 0.001$. Post hoc comparisons revealed that mice had higher scores (cumulative score over 60 min) than rats during the acclimatization stage prior to drug challenge ($p < 0.01$). ANOVA of locomotor scores (cumulative score for 3 h postchallenge) following apomorphine administration showed an effect of drug challenge, $F(3, 24) = 3.70, p = 0.025$. Post hoc comparisons revealed that apomorphine (1, 3, and 10 mg/kg) increased the locomotor score with rats ($p < 0.01$) and that apomorphine (3 and 10 mg/kg) increased the locomotor score with mice ($p < 0.01$) (Fig. 2).

Study 5: Effect of Apomorphine on Activity in Mice in Two Contrasting Test Environments—the Home Cage and Exploratory Box (Fig. 3)

There was no difference in activity (automated counts), stereotypy or locomotion scores in either the home cage or exploratory box between groups prior to apomorphine administration. ANOVA of home-cage activity (cumulative score for 3 h post challenge) following apomorphine administration showed no effect of drug challenge, $F(4, 15) = 0.25, p = 0.903$. ANOVA of the cumulative stereotypy score over 3 h post challenge showed an effect of challenge, $F(4, 15) = 93.73, p < 0.001$. Post hoc comparisons revealed that apomorphine (1, 3, 10, and 30 mg/kg) increased the stereotypy score ($p < 0.01$). ANOVA of the cumulative locomotor score over 3 h postchallenge showed an effect of challenge, $F(4, 15) = 9.12, p < 0.001$. Post hoc comparisons revealed that apomorphine (3 mg/kg) ($p < 0.05$) and

TABLE 2

EFFECT OF APOMORPHINE ON CORE BODY TEMPERATURE OF THE RAT IN DIFFERENT TEST CONDITIONS

Group	T 0	T 30	T Change
Home cage			
Vehicle	37.30 ± 0.25	37.95 ± 0.30	0.65 ± 0.53
0.01 mg/kg	37.16 ± 0.08	37.53 ± 0.26	0.37 ± 0.33
0.03 mg/kg	36.87 ± 0.18	37.09 ± 0.43	0.22 ± 0.30
0.01 mg/kg	37.05 ± 0.15	37.18 ± 0.22	0.12 ± 0.15
0.3 mg/kg	37.36 ± 0.19	36.96 ± 0.49	−0.40 ± 0.47*
1 mg/kg	37.38 ± 0.22	36.68 ± 0.29*	−0.71 ± 0.26*
3 mg/kg	37.29 ± 0.18	36.38 ± 0.45†	−0.92 ± 0.36†
10 mg/kg	37.14 ± 0.10	36.54 ± 0.30†	−0.60 ± 0.28*
30 mg/kg	37.27 ± 0.34	36.03 ± 0.33†	−1.24 ± 0.45†
Exposure to novel test cage			
Vehicle	36.60 ± 0.14	37.83 ± 0.20	1.23 ± 0.27
0.03 mg/kg	37.02 ± 0.19	37.37 ± 0.17	0.35 ± 0.28
0.1 mg/kg	36.98 ± 0.28	37.43 ± 0.13	0.46 ± 0.30
0.3 mg/kg	37.06 ± 0.24	37.58 ± 0.24	0.53 ± 0.42
1 mg/kg	37.22 ± 0.24	37.45 ± 0.42	0.23 ± 0.45
3 mg/kg	37.94 ± 0.21	37.58 ± 0.55	0.64 ± 0.46
10 mg/kg	36.80 ± 0.22	36.95 ± 0.30	0.16 ± 0.46
Exposure to exploratory box			
Vehicle	37.33 ± 0.26	37.86 ± 0.18	0.53 ± 0.20
0.03 mg/kg	37.05 ± 0.11	37.72 ± 0.27	0.67 ± 0.25
0.1 mg/kg	37.06 ± 0.16	36.73 ± 0.20†	−0.33 ± 0.30*
0.3 mg/kg	37.20 ± 0.22	36.75 ± 0.15†	−0.45 ± 0.27*
1 mg/kg	37.24 ± 0.16	37.09 ± 0.15†	−0.16 ± 0.26
3 mg/kg	37.25 ± 0.15	36.83 ± 0.39†	−0.41 ± 0.39*
10 mg/kg	37.13 ± 0.20	36.75 ± 0.21†	−0.38 ± 0.17*

Effect of apomorphine (0.01–30 mg/kg SC) on core body temperature of the rat in (A) animals housed in their home cage, (B) animals exposed to a novel test cage (fresh bedding), and (C) animals exposed to the exploratory box. Colonic temperatures were taken immediately prior to [T(0)] administration of either vehicle or apomorphine. Five minutes later the animals were exposed to the test condition of interest for 25 min. The temperatures were taken again 30 min post challenge [T(30)]. Data is expressed as mean temperature and temperature change with standard error of the mean. There were five to six animals per group. *$p < 0.05$; †$p < 0.01$ vs. vehicle control.

apomorphine (10 and 30 mg/kg) ($p < 0.01$) increased the locomotor score.

ANOVA of activity in the exploratory box (cumulative score for 3 h postchallenge) following apomorphine administration showed only a modest effect of drug challenge, $F(4, 15) = 1.98$, $p = 0.149$. ANOVA of the cumulative stereotypy score over 3 h postchallenge showed an effect of challenge, $F(4, 15) = 65.71$, $p < 0.001$. Post hoc comparisons revealed that apomorphine (3, 10, and 30 mg/kg) increased the stereotypy score ($p < 0.01$). ANOVA of the cumulative locomotor score over 3 h postchallenge showed an effect of challenge, $F(4, 15) = 12.85$, $p < 0.001$. Post hoc comparisons revealed that apomorphine (10 and 30 mg/kg) increased the locomotor score ($p < 0.01$).

When compared together, ANOVA of the activity counts in the home cage and exploratory box showed effects of test environment, $F(1, 30) = 12.63$, $p = 0.001$. Post hoc comparisons revealed higher counts in the exploratory box in response to 3 and 10 mg/kg apomorphine when compared to their home-cage counterparts. ANOVA of the stereotypy

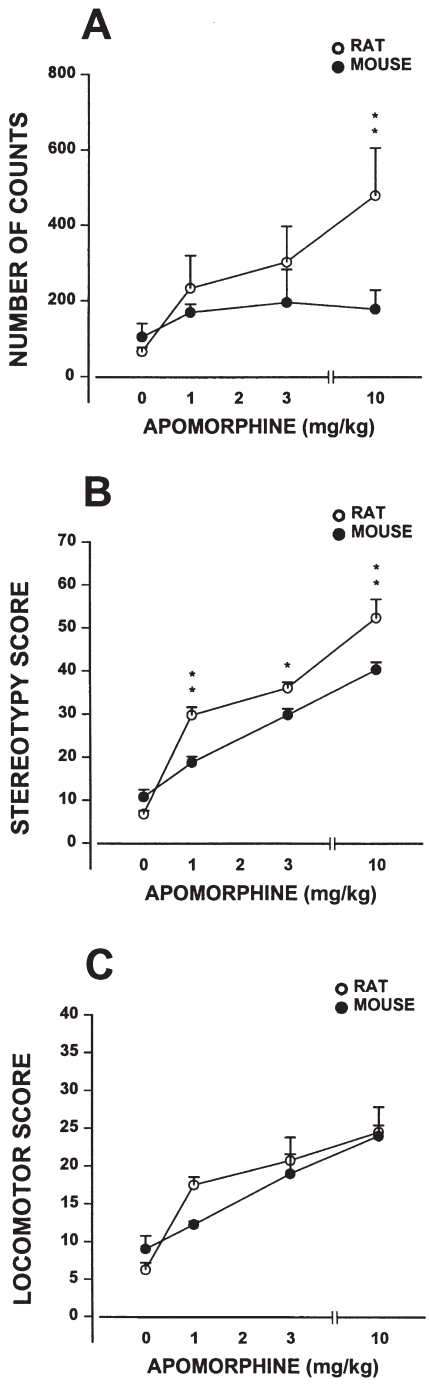

FIG. 2. Effect of apomorphine on activity of rats and mice in the home cage. Animals were placed singly in a standard plastic bottomed cage with fresh bedding. Behavior was measured automatically in the home cage activity monitor with passive infrared sensors mounted above the floor of the test cage in combination with observational recording for 1 h pre- and 3 h post-challenge with 1, 3, and 10 mg/kg apomorphine SC. Data is represented as mean cumulative score with standard error of the mean of four animals for 3 h post-challenge. *$p < 0.05$; **$p < 0.01$ vs. mouse counterpart, Fishers LSD.

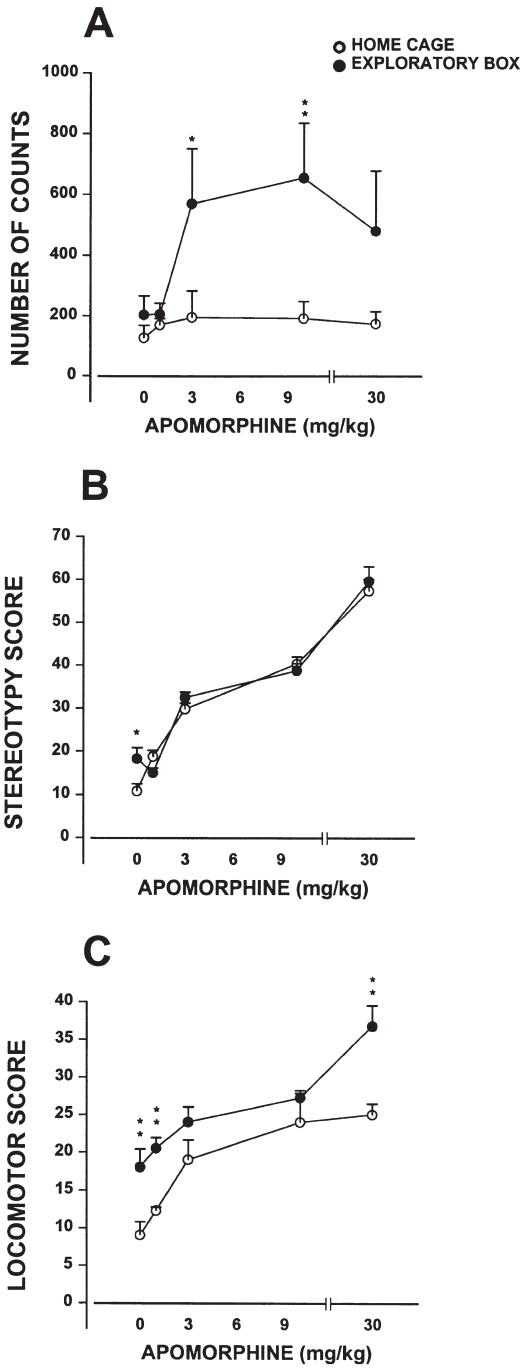

FIG. 3. Effect of apomorphine on activity of mice in two contrasting test environments, the home cage, and the exploratory box. Animals were placed singly in a standard plastic-bottomed cage with fresh bedding or in the exploratory box. Behavior was measured automatically in the home-cage activity monitor with passive infrared sensors mounted above the floor of the test cage and similarly by infrared transmitters and receivers mounted symmetrically around the arena in combination with observational recording for 1 h pre- and 3 h postchallenge with 1, 3, 10, and 30 mg/kg apomorphine SC. Data is represented as mean cumulative score with standard error of the mean of four animals for 3 h postchallenge. *$p < 0.05$; **$p < 0.01$ vs. home-cage counterpart, Fishers LSD.

scores demonstrated the effects of the drug, $F(4, 30) = 152.91$, $p < 0.001$. Post hoc comparisons revealed higher stereotypy scores in vehicle-treated animals exposed to the exploratory box when compared to their home-cage counterparts ($p < 0.05$). ANOVA of the locomotion scores showed effects of drug, $F(4, 30) = 20.28$, $p < 0.001$, and test condition, $F(1, 30) = 28.84$, $p < 0.001$. Post hoc comparisons revealed higher locomotion scores in vehicle-treated and animals treated with 1 and 30 mg/kg apomorphine in the exploratory box when compared to their home-cage counterparts ($p < 0.01$) (Fig. 3).

Study 6: Effect of Test Conditions on Apomorphine-Induced Changes to Dopamine Turnover in Mouse Striatum (Table 3)

When compared together, ANOVA of striatal DOPAC concentrations in animals exposed to the home cage and exploratory box showed effects of test environment, $F(1, 51) = 5.69$, $p = 0.021$, and drug challenge, $F(4, 51) = 14.97$, $p < 0.001$. Post hoc comparisons revealed that DOPAC concentrations in response to exposure to the exploratory box were reduced when compared to the home-cage counterparts ($p < 0.05$). Apomorphine (1, 3, 10, and 30 mg/kg) reduced DOPAC concentrations in the striatum in animals exposed to both test environments ($p < 0.01$).

ANOVA of HVA concentrations showed effects of test environment, $F(1, 51) = 7.62$, $p = 0.008$, and drug challenge, $F(4, 51) = 22.21$, $p < 0.001$. Post hoc comparisons revealed that apomorphine (1, 3, 10, and 30 mg/kg) reduced HVA concentrations in the striatum in animals exposed both to the home test cage and exploratory box ($p < 0.01$).

ANOVA of dopamine concentrations showed effects of test environment, $F(1, 51) = 4.52$, $p = 0.038$, drug challenge, $F(4, 51) = 2.85$, $p = 0.033$, and a test × drug interaction, $F(4, 51) = 2.66$, $p = 0.043$. Post hoc comparisons revealed that apomorphine (10 mg/kg) increased dopamine concentrations in animals exposed to the home test cage environment only ($p < 0.05$).

ANOVA of the dopamine turnover ratio DOPAC:DA showed effects of drug challenge, $F(4, 51) = 14.08$, $p < 0.001$, and test environment, $F(1, 51) = 13.82$, $p < 0.001$. Post hoc comparisons revealed that the turnover ratio in those animals exposed to the exploratory box was lower when compared to their home cage counterparts ($P < 0.01$). Apomorphine (1, 3, 10, and 30 mg/kg) reduced the turnover ratio in animals exposed to both test conditions ($p < 0.01$).

ANOVA of the dopamine turnover ratio HVA:DA showed effects of drug challenge, $F(4, 51) = 7.50$, $p < 0.001$, and test environment, $F(1, 51) = 13.5$, $p < 0.001$. Post hoc comparisons revealed that the turnover ratio in those animals exposed to the exploratory box was lower when compared to their home-cage counterparts ($p < 0.01$). Post hoc comparisons revealed that apomorphine (3, 10, and 30 mg/kg) reduced the turnover ratio in those animals exposed to the home-cage test environment ($p < 0.01$). Similarly, apomorphine (1 and 10 mg/kg) reduced the turnover ratio in those animals exposed to the exploratory box ($p < 0.05$) (Table 3).

DISCUSSION

Behavioral effects of apomorphine in the present study are similar to those reported previously where it has also been shown than dopamine antagonists such as haloperidol and sulpiride can block these locomotor effects and inhibit the stereotypy when apomorphine activates postsynaptic dopamine receptors (15,29). Although changes in activity were ob-

TABLE 3

EFFECT OF TEST CONDITIONS ON APOMORPHINE-INDUCED CHANGES TO DOPAC, HVA, AND DA CONCENTRATIONS IN MOUSE STRIATUM

Group	DOPAC	HVA	DA	DOPAC/DA	HVA/DA
Exposure to home test cage					
Vehicle	1291 ± 248	1332 ± 114	6343 ± 765	0.206 ± 0.036	0.222 ± 0.027
1 mg/kg	466 ± 165†	568 ± 90†	5195 ± 1148	0.117 ± 0.034†	0.166 ± 0.060
3 mg/kg	529 ± 133†	867 ± 67†	7669 ± 975	0.081 ± 0.028†	0.118 ± 0.008†
10 mg/kg	476 ± 110†	858 ± 66†	9521 ± 719	0.049 ± 0.010†	0.091 ± 0.004†
30 mg/kg	351 ± 66†	813 ± 34†	8072 ± 817	0.042 ± 0.004†	0.105 ± 0.010†
Exposure to exploratory box					
Vehicle	924 ± 84	1177 ± 69	8021 ± 430	0.114 ± 0.006*	0.147 ± 0.003†
1 mg/kg	389 ± 26‡	665 ± 60§	9143 ± 783	0.042 ± 0.003§	0.075 ± 0.008‡
3 mg/kg	293 ± 42§	501 ± 79§	6963 ± 1064	0.045 ± 0.007§	0.076 ± 0.010‡
10 mg/kg	256 ± 23§	646 ± 74§	9065 ± 553	0.028 ± 0.003§	0.071 ± 0.006§
30 mg/kg	364 ± 69§	788 ± 71§	9085 ± 750	0.041 ± 0.009§	0.088 ± 0.010

Animals were placed singly in a standard plastic bottomed cage with fresh bedding or in the exploratory box and left to acclimatize to the test condition for 2 h. Following this period animals were challenged with 1, 3, 10, or 30 mg/kg apomorphine or vehicle SC and sacrificed 40 min postchallenge. Neurotransmitter concentrations are expressed as ng/g wet weight of tissue. Data is represented as mean with standard error of the mean of 5-6 animals *$p < 0.05$; †$p < 0.01$ vs. home-cage vehicle control; ‡$p < 0.05$; §$p < 0.01$ vs. exploratory box vehicle control. Fishers LSD. DOPAC: Dihydroxyphenylacetic acid; HVA: Homovanillic acid; DA: Dopamine.

served in all test environments, the present study indicates that when assessing apomorphine-induced changes in activity it is essential to consider the environmental conditions. The degree of familiarity with the test environment has an impact on the observed effect of the drug and detection of drug-induced effect on behavior. A previous study by Adams and Geyer (1) demonstrated that reductions in locomotion and investigatory holepoking produced by low doses of hallucinogens in rats tested in a novel test chamber are not seen if the rats are already familiar with the test chamber. Beninger (3) also found that two drugs, metergoline and quipazine, that interact with the serotonergic system, produced different activity profiles depending on the familiarity of the testing environment. Other studies have shown that habituation to the test conditions can lead to failure in detecting apomorphine-induced hypoactivity (31). From the present results, it is clear that when assessing apomorphine-induced hypoactivity in a nonstressful environment such as the home cage, subjects should not be acclimatized to the test conditions. Moreover, trials must be short in duration, as drug-induced hypoactivity is only detected during the brief period that vehicle-treated subjects acclimatize to the test environment. Tests conducted in novel environments such as the exploratory box or the widely employed "open field" are particularly suited to this end. The use of paradigms having limited ability to detect drug-induced decreases in activity is a common problem in behavioral studies on the effects of psychoactive drugs.

In contrast to hypoactivity, hyperactive effects are best detected following a period of acclimatization to the test conditions. Subjects should be acclimatized prior to drug treatment or during the time it takes for the drug to take effect. Alternatively, activity during preexposure sessions shows that such activity is subject to habituation, and thus, stimulant drug effects can be more easily detected (17). The present study demonstrated in rats that apomorphine (0.3–10 mg/kg) disturbs the habituation-related process of a decrease in rat locomotor activity in the home cage, and that drug-induced activity is best detected following a period of acclimatization to the monitor. Placement of an animal in a novel environment resulted in a behavioral response by the animal that interacted directly with the drug effect to mask drug-induced activity. In the activity box, drug-related increases in activity were best seen towards the end of the observation period. As vehicle-treated control animals acclimatized to the arena, the hyperkinetic effects of apomorphine on locomotor activity became more apparent. As has been reported previously, a change of environment can influence the manner in which drug-induced alteration of behavior is detected (9,17,21). The primary influences of novelty are to delay the detection of drug-induced activity, which may shorten the apparent duration of activity and mask the activity induced by the drug.

Given the complex profile of apomorphine on stereotypic behavior it is essential to consider these effects in conjunction with any locomotor changes. In mice, apomorphine-induced stereotyped behavior is similar to that previously reported following apomorphine administration to rats (12). In the home cage and exploratory box at the higher doses of apomorphine, mice show intense stereotypic behavior and remain in a relatively restricted area of the cage. Consequently, locomotor activity of these mice was depressed when compared with mice treated with lower apomorphine doses, but is still higher than that of the vehicle-treated mice. Although it was anticipated that the nature of the test chamber would have important consequences with regard to drug-induced changes to both locomotor and stereotypical movement, the intensity of apomorphine-induced stereotyped behaviors did not differ across test conditions in the present study. Previous studies have reported that stereotypy induced by amphetamine is lessened in a novel environment (22). This is suggested to be due to an increase in exploration in the novel environment that interferes with stereotypy. Apomorphine has been reported to fail to produce significant stereotypy in an open-field test (18). Such differences imply that apomorphine induces a greater locomotor effect with diminished stereotypy in the open field when compared to the home environment. As previously suggested by Havemann and co-workers (12),

patterns of behavior in response to increasing doses of apomorphine change in response to environmental factors, thereby demonstrating behavioral competition among motor responses. Joyce and Iversen (13) interpreted similar results obtained after amphetamine administration as competition between two systems, namely mesocorticolimbic and nigrostriatal, for motor output pathways. Although apomorphine-induced stereotypy scores did not differ across test conditions in the present study, stereotypy and locomotion scores of vehicle-treated animals in the exploratory box were greater when compared to animals observed in a home test cage. Moreover, locomotion scores in response to apomorphine in the exploratory box were higher when compared to apomorphine treated animals in the home test cage. It is suggested that stereotypic behavior induced by apomorphine may be regarded as a more compulsive behavior, as it is not altered by environmental factors such as the construction and the area of the test environment, or by psychological factors including the degree of novelty or the familiarity of the animals with the test box. By contrast, locomotion is more affected by changes in the experimental conditions. Apomorphine-induced locomotion seems to be of a less compulsive nature, as it is clearly influenced by the environment and experimental conditions. Moreover, automated monitoring systems differ to a great extent in relation to their similarity or dissimilarity to the animals home environment, which may often lead to variation in the detection of particular drug-induced effects under prescribed experimental conditions.

A direct comparison of rat and mouse demonstrated that the gross activity count in response to apomorphine was detected in rats but not in mice in the automated home-cage activity monitor. This may be attributed to the smaller size of the mouse, possibly making it more difficult to detect its activity in the home cage with passive infrared sensors. The fact that automated activity scores do not correspond to observation recordings questions the usefulness of this type of automated activity system for detecting the psychostimulant effects of apomorphine in mice. We have, however, been able to detect the psychomotor stimulant effects of other drugs including *N*-methyl-D-aspartate receptor antagonists and amphetamine in mice with this system. Scoring the behavior using concurrent stereotypy and locomotor scales refines the monitoring system approach and allowed for specification of behaviors that are modified following drug treatment or exposure to novel environments. In this regard, the measured stereotypic and locomotor response to apomorphine in both rats and mice is comparable, although rats exhibited a more intense stereotypic response to apomorphine than mice over the dose range used.

In all the tests studied here, apomorphine reduced core body temperature. However, when measuring the hypothermic response to apomorphine, the test conditions impacted upon the response to the drug. Fresh bedding in home cage tests and exposure to novel environments blunted the hypothermic effects of apomorphine. The response following exposure to these test conditions no longer followed a dose-related reduction in temperature following acute challenge with apomorphine. The blunted hypothermia in response to apomorphine is likely to be attributed to exposure to a novel environment. Arousal following exposure to a novel test environment may be evaluated by measuring corticosterone concentration in the blood. Animals are generally aroused by their surroundings, and exposure to the novel test cage and exploratory box induce increases in serum corticosterone concentrations in rodents. Moreover, exposure to such conditions can lead to modest increases in body temperature. This is particularly apparent following exposure to a novel test cage. The exploratory box, although deemed a more stressful environment (bright lighting, perspex walls and white floor), was less potent at producing this temperature increase. It is apparent, therefore, that the novel test cage had a greater impact on the response to drug challenge in this test. Such findings have implications in attempts to comeasure behavioral and physiological parameters simultaneously in pharmacological challenge studies.

Changes that occur in locomotor activity following the administration of apomorphine are related to changes in dopaminergic activity in mesolimbic and nigrostriatal dopamine pathways and their terminal regions, the nucleus accumbens and the striatum (2,20,26,28). In the present study, apomorphine reduced DOPAC and HVA concentrations in the mouse striatum. As steady-state dopamine concentrations remained unchanged, these reductions equated to a reduction in dopamine turnover. Short-term reduction of DOPAC in the striatum of rodent brain is thought to provide an accurate reflection of activity of dopamine neurons of the nigrostriatal pathway. Reduction in dopamine turnover by apomorphine is believed to reflect the activation of presynaptic autoreceptors, consequently attenuating dopamine synthesis and release (5). This ex vivo measure correlates significantly to other in vivo, behavioral, and electrophysiological measures of autoreceptor activation (16,19,32). The reduction in dopamine turnover as reported in the exploratory box when compared to the home cage possibly reflects a reduction in basal dopamine release, reuptake, and intraneuronal metabolism. Although animals exposed to the exploratory box had dopamine turnover ratios significantly lower than their home-cage counterparts, apomorphine produced reductions in turnover in both test environments. A dose–effect relationship was apparent with animals exposed to the home test cage but not to the exploratory box. Such a test of presynaptic activity was dissimilar in animals exposed to contrasting test environments, an important consideration when drawing correlates between drug-induced behavioral and neurochemical responses.

The contribution of pharmacological agents as CNS probes to elicit quantifiable behavioral, physiological, and neurochemical responses has been invaluable in increasing our knowledge of the underlying neural substrates of normal and abnormal brain function. The apomorphine challenge test has a long history in psycho- and behavioral pharmacology. Responses induced by apomorphine have been proposed as a model of acute psychosis, a way to screen compounds for potential antipsychotic activity, and a means to test the sensitivity of the central dopaminergic system in vivo. Given that the underlying factors contributing to a drug response include effects of the drug itself as well as behavior invoked by the particular environment, the importance of assessing the effects of psychostimulants in the appropriate experimental conditions is emphasized. Although this study examined the responses to apomorphine, there is no reason why the experimental conditions set up to detect the biphasic effects of apomorphine could not also extend to the detection of hyperkinesis or sedation following the administration of other central stimulant or depressant drugs.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the support of the Higher Education Authority of Ireland.

REFERENCES

1. Adams, L. H.; Geyer, M. A.: A proposed animal model for hallucinogens based on LSD's effects on patterns of exploration in rats. Behav. Neurosci. 5:881–900; 1985.
2. Arnt, J.: Behavioral studies of dopamine receptors: Evidence for regional selectivity and receptor multiplicity. In: Creese, I.; Fraser, C. M.; Liss, A. R., eds. Dopamine receptors. New York: Alan R. Liss; 1987:199.
3. Beninger, R. J.: Effects of metergoline and quipazine on locomotor activity of rats in novel and familiar environments. Pharmacol. Biochem. Behav. 20:701–705; 1984.
4. Constentin, J.; Duterte-Boucher, D.; Panissaud, C.; Turtus, A. M.: Dopamine D_1 and D_2 receptors mediate opposite effects of apomorphine on the body temperature of reserpinized mice. Neuropharmacology 29:31–35; 1990.
5. Costall, B.; Lim, S. K.; Naylor, R. J.: Characterization of the mechanisms by which purported dopamine agonists reduce spontaneous locomotor activity of mice. Eur. J. Pharmacol. 73:175–188; 1981.
6. Di Chiara, G.; Corsini, G. U.; Mereu, G. P.; Tissari, A.; Gessa, G. L.: Self inhibitory dopamine receptors: Their role in the biochemical and behavioral effects of low doses of apomorphine. In: Roberts, P. J., ed. Advances in biochemical psychopharmacology. New York: Raven Press; 1978:275–284.
7. Earley, B.; Burke, M.; Leonard, B. E.: Behavioral, biochemical and histological effects of trimethyltin (TMT) induced brain damage in the rat brain. Neurochem. Int. 21:352–366; 1992.
8. Ellinwood, E. H.; Balster, R. L.: Rating the behavioral effects of amphetamine. Eur. J. Pharmacol. 28:35–41; 1974.
9. Gayton, O.; Swann, A.; Dafny, N.: Diurnal differences in rats motor response to amphetamine. Eur. J. Pharmacol. 345:119–128; 1998.
10. Geyer, M. A.; Russo, P. V.; Masten, V. L.: Multivariate assessment of locomotor behavior: Pharmacological and behavioral analyses. Pharmacol. Biochem. Behav. 25:277–288; 1986.
11. Haemisch, A.; Voss, T.; Gartner, K.: Effects of environmental enrichment on aggressive behavior, dominance hierarchies and endocrine states of male DBA/2S mice. Physiol. Behav. 56:1041–1048; 1994.
12. Havemann, U.; Magnus, B.; Moller, H. G.; Kuschinsky, K.: Individual and morphological differences in the behavioral response to apomorphine in rats. Psychopharmacology (Berlin) 90:40–48; 1986.
13. Joyce, E.; Iversen, S. D.: Dissociable effects of 6-OH-dopamine-induced lesions of neostriatum on anorexia, locomotor activity and stereotypy: The role of behavioural competition. Psychopharmacology (Berlin) 83:363–366; 1984.
14. Lals, S.: Apomorphine in the evaluation of dopaminergic function in man. Prog. Neuropsychopharmacol. 12:117–124; 1988.
15. Leonard, B. E.: The pharmacology of the phenothiazines, butyrophenones, thioxanthines and diphenylbutyl piperidines. In: Barnes, T., ed. Antipsychotic drugs and their side effects. London: Academic Press; 1993:3–23.
16. Martin, G. E.; Bendesky, R. J.: Mouse locomotor activity: An in vivo test for dopamine autoreceptor activation. J. Pharmacol. Exp. Ther. 229:706–711; 1984.
17. Mazurski, E. J.; Beninger, R. J.: Stimulant effects of apomorphine and (+) amphetamine in rats with varied habituation to test environment. Pharmacol. Biochem. Behav. 29:249–255; 1988.
18. Mueller, K.; Peel, J. L.: Scopolamine produces locomotor stereotypy in an "open field" but apomorphine does not. Pharmacol. Biochem. Behav. 36:613–617; 1990.
19. Ozaki, N.; Nakahara, D.; Miura, H.; Kasahara, Y.; Nagatsu, T.: Effects of apomorphine on in vivo release of dopamine and its metabolites in the prefrontal cortex and the striatum studies by a microdialysis method. J. Neurochem. 53:1861–1864; 1989.
20. Plaznik, A.; Stefansli, R.; Kostowski, W.: Interaction between accumbens D_1 and D_2 receptors regulating rat locomotor activity. Psychopharmacology (Berlin) 99:558–562; 1989.
21. Riffee, W. H.; Wilcox, R. E.; Smith, R. V.: Modification of drug-induced behavioral arousal by pre-injection routines in mice. Psychopharmacology (Berlin) 63:1–5; 1979.
22. Russel, R. L.; Pihl, R. O.: The effect of dose, novelty and exploration on amphetamine-produced stereotyped behavior. Psychopharmacology (Berlin) 6:93–100; 1978.
23. Serra, G.; Argiolas, A.; Klimek, V.; Fadda, F.; Gessa, G. L.: Chronic treatment with antidepressants prevents the inhibitory effect of small doses of apomorphine on dopamine synthesis and motor activity. Life Sci. 25:41–424; 1979.
24. Setler, P. E.; Sarau H. M.; Zirkle, C. L.; Saunders, H. L.: The central effects of a novel dopamine agonist. Eur. J. Pharmacol. 50:419–430; 1978.
25. Seyfried, C. A.; Adams, G.; Greve, T.: An automated direct-injection HPLC method for the electrochemical/fluorimetric quantitation of monoamines and related compounds optimized for screening of large numbers of animals. Biomed. Chromotogr. 1:78–88; 1986.
26. Starr, M. S.: D_1/D_2 behavioral interactions in the rat involving striatal dopamine D_1 receptors. Eur. J. Pharmacol. 151:479–482; 1988.
27. Sturgeon, R. D.; Fessler, R. G.; Meltzer, M. Y.: Behavioral rating scales assessing phencyclidine-induced locomotor activity, stereotyped behavior and ataxia in rats. Eur. J. Pharmacol. 59:169–179; 1979.
28. Swerdlow, N.; Koob, G.: The neural substrates of apomorphine-stimulated locomotor activity following denervation of the nucleus accumbens. Life Sci. 35:2537–2544; 1984.
29. Vaccheri, A.; Dall'Olio, R.; Gandolfi, O.; Mantanaro, N.: Involvement of different dopamine receptors in rat diphasic motility response to apomorphine. Psychopharmacology (Berlin) 89:265–268; 1986.
30. Van De Weerd, H. A.; Van Loo, P. L. P.; Van Zutphen, L. F. M.; Koolhaas, J. M.; Baumans, V.: Nesting material as environmental enrichment has no adverse effects on behavior and physiology of laboratory mice. Physiol. Behav. 62:1019–1028; 1997.
31. Yarbrough, G. G.; McGuffin-Clineschmidt, J.; Singh, D. K.; Haubrich, D. R.; Bendesky, R. J.; Martin, G. E.: Electrophysiological, biochemical and behavioral assessment of dopamine autoreceptor activation by a series of dopamine agonists. Eur. J. Pharmacol. 99:73–78; 1984.
32. Vos, P. E.; Wolterink, G.; Van Ree, J. M.: The behavioral effects of habituation and challenge with apomorphine after 6-OH DA lesioning of the nucleus accumbens in rats. Eur. Neuropsychopharmacol. 5:471–476; 1995.