Differences in the locomotor activity of mice as measured by an Animex and photoresistor actometer

Recently, a new apparatus for measuring the locomotor activity of small animals has been introduced. The principle is the recording of changes in electromagnetic field (Svensson & Thieme, 1969). The apparatus (Animex) has been used by Svensson & Waldeck (1970), Strömberg (1970) and Corrodi, Fuxe & others (1970). We have noted that the apparatus gave qualitatively different results from those of the photoresistor actometer and we now report some significant differences between the results from the two instruments.

The experiments were made on male Albino Swiss mice, 18–23 g. The motor activity was measured using an Animex apparatus (Farad Electronics—Stockholm) and a photoresistor actometer (with one photoresistor and two light beams). The animals were placed individually in the instruments and their locomotor activity measured for the following 15 min. The experimental schedules and the environmental conditions were identical for both methods.

In reserpinized mice (5 mg/kg, s.c., 16 h before the test), L-dopa (500 mg/kg, i.p., 15 min before the test) increases the number of counts measured on the Animex apparatus at a sensitivity of 40 μA (as well as at 20 μA), but does not change the number of counts measured in photoresistor actometer (Table 1). Similar differences were noted in mice pretreated with α-methyltyrosine (250 mg/kg, i.p., 16 h before the test) and L-dopa (500 mg/kg, i.p., 15 min before test) (Table 1).

In reserpinized mice (5 mg/kg) amantadine (hydrochloride, 40, 80, or 160 mg/kg, s.c. 20 min before the test) markedly increased the number of counts in the Animex apparatus (sensitivity 40 μA) but did not change or only slightly increased (at 160 mg/kg) the number of movements recorded with the photoresistor actometer (Table 2). Similar differences between the results from these two instruments were found in reserpinized animals treated with phenoxybenzamine (20 mg/kg, i.p., 2 h before test) and amantadine (Table 2).

Strömberg, Svensson & Waldeck (1970) reported that the locomotor activity of reserpinized mice in groups of three measured by Animex was markedly increased by amantadine. We repeated this experiment using similar conditions and recording the locomotor activity with both methods. The only difference was that we used a dose of 5 mg/kg of reserpine instead of 10 mg/kg. Reserpine was given intraperi-

Table 1. *Effect of* L-*dopa on locomotor activity in mice pretreated with reserpine or* α-*methyltyrosine.* Drugs were given as follows: reserpine (s.c.) and α-methyltyrosine (i.p.) 16 h, L-dopa (i.p.) 15 min before the test. Locomotor activity was measured in individual animals for 15 min. Statistical significance was calculated with Student's *t*-test as follows: II/I; III/II; V/IV; VI/V; *—$P < 0.05$.

Group	Drug mg/kg	Activity counts Animex Sensitivity 40 μA	Activity counts Animex Sensitivity 20 μA	Actometer
I	—	419·0 ± 37·1	100·2 ± 18·3	148·2 ± 20·0
II	Reserpine 5	14·0 ± 3·6*	1·2 ± 0·37*	1·6 ± 0·18*
III	Reserpine 5 + L-dopa 500	204·8 ± 82·3*	77·5 ± 30·6*	3·4 ± 2·18
IV	—	549·6 ± 42·8	139·0 ± 14·3	175·5 ± 9·6
V	α-Methyltyrosine 250	20·0 ± 9·60*	1·0 ± 0·21*	4·2 ± 1·32*
VI	α-Methyltyrosine 250 + L-dopa 500	204·1 ± 50·3*	90·1 ± 34·9*	18·0 ± 8·5

Table 2. *Effect of amantadine on locomotor activity in mice pretreated with reserpine or reserpine and phenoxybenzamine.* Drugs were given at following time intervals: reserpine (s.c.) 16 h, phenoxybenzamine (i.p.) 2 h, amantadine hydrochloride (s.c.) 20 min before the test. Locomotor activity was measured in individual animals for 15 min. Statistical significance was calculated with Student's *t*-test as follows: II/I; III, IV, V/II; VI/II; VII, VIII/VI; *—$P < 0.05$.

Group	Drug mg/kg	Activity counts Animex	Actometer
I	—	507·7 ± 7·5	175·5 ± 9·5
II	Reserpine 5	9·75 ± 3·9*	1·4 ± 0·36*
III	Reserpine 5 + amantadine 40	30·1 ± 7·1*	4·4 ± 1·6
IV	Reserpine 5 + amantadine 80	116·8 ± 44·8*	5·5 ± 1·9
V	Reserpine 5 + amantadine 160	240·7 ± 65·7*	19·3 ± 7·6*
VI	Reserpine 5 + phenoxybenzamine 20	6·5 ± 1·7	0·7 ± 0·3
VII	Reserpine 5 + phenoxybenzamine 20 + amantadine 40	89·0 ± 35·5*	5·3 ± 2·9
VIII	Reserpine 5 + phenoxybenzamine 20 + amantadine 80	190·7 ± 86·3*	7·1 ± 4·3

toneally 6 h 40 min before the test and amantadine (hydrochloride, 150 mg/kg, i.p.) 1 h 40 min before the test. The mice were put into the Animex or photoresistor actometer in groups of three, 1 h 30 min after amantadine. The locomotor was counted for 30 min beginning 10 min after the mice were put into the cages.

The results obtained with the Animex were similar to those described by Strömberg & others (1970). The number of counts after reserpine alone was 6·5 ± 1·5, after reserpine and amantadine—105·1 ± 39·9 ($P < 0.05$). The analogous values obtained in the photoresistor actometer were 0·7 ± 0·25 and 12·1 ± 3·6 ($P < 0.01$).

Visual observations revealed that in mice treated with L-dopa or amantadine there was some stimulation expressed as hyperexcitability, small movements, usually with single, uncoordinated jumps. In a light-beam actometer the animal usually does not cross the beam during such movements, and therefore they are not recorded. But in some experiments after amantadine we observed a not very large but statistically significant increase of counts by 30–40% compared with control values. This was probably caused by the fact that the mouse sat close to the light beam. Such an increase has been never observed after treatment with phenoxybenzamine.

With the use of the Animex, various types of movements are measured, including those which are not coordinated. The photoresistor actometer records primarily large, coordinated locomotive movements. The different results obtained with those two instruments may lead to different interpretations, which are exemplified by the experiments with amantadine.

It seems that different results are obtained if the balance between dopaminergic and noradrenargic neuron activity is shifted towards the former, particularly if dopaminergic neurons are stimulated with concomitant partial or total block of noradrenergic neurons.

The differences in the locomotor activity measured with two different automated methods indicate once again the importance of visual observation or animals tested.

Institute of Pharmacology,
Polish Academy of Sciences,
Cracov, Poland.

J. MAJ
B. DUREK
W. PALIDER

July 3, 1971

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On the action of bombesin on the kidney of the rat and the dog

Bombesin is the tetradecapeptide isolated from extracts of the skin of the European discoglossid frogs *Bombina bombina* and *Bombina variegata variegata* (Anastasi, Erspamer & Bucci, 1971).

We now report the action of bombesin on the kidney of the rat and the dog, and also on the renin-angiotensin system.

Bombesin produced a reduction of urine flow in the rat. In animals anaesthetized with ethanol (5 ml of 15% ethanol per 100 g weight) and then given an intravenous infusion of 2% ethanol (50 μl/min) the threshold dose of bombesin capable of reducing diuresis by intravenous infusion was 50 ng/kg per min; in conscious rats given a water load by oral route the threshold subcutaneous dose was 20–50 μg/kg.

Reduction in urine flow was accompanied by a reduction in glomerular filtration rats (creatinine clearance) and in renal plasma flow (*p*-aminohippurate clearance). In a typical experiment in which bombesin was infused for 30 min at a rate of 100 ng/kg per min reduction of urine flow was 80%, of glomerular filtration rate 75% and of renal plasma flow 68%. Fractional sodium reabsorption (C_{Na}/C_{Cr} %) decreased during antidiuresis, which was apparently counteracted by high sodium intake.

In conscious rats, both hydrated and non-hydrated, 100 μg/kg of bombesin given subcutaneously produced an increase of blood pressure (10–30 mm Hg) lasting more than 2 h. In rats anaesthetized with ethanol, the intravenous infusion of 100 ng/kg per min of bombesin for 30 min initially caused a rise in blood pressure (20–30 mm Hg) which was followed by slow decline and then, after the infusion was discontinued, by a return to normal levels or slight hypotension (10–20 mm Hg). Thus, changes of blood pressure could counteract, but not favour, bombesin antidiuresis.

The effect of bombesin on the dog kidney was intense. In animals anaesthetized with sodium pentobarbitone and given a 5% water load by stomach tube, the threshold dose of the polypeptide producing antidiuresis by rapid intravenous injection was about 0·1 μg/kg, by intravenous infusion 0·5 ng/kg per min, and by subcutaneous route 0·5 μg/kg. The effect was of rapid onset and its duration depended on the dose given. Tachyphylaxis readily occurred, with conspicuous differences from one animal to another. With low doses a fair dose response relation could sometimes be observed; with large doses tachyphylaxis was more prompt and intense. Generally, antidiuresis paralleled the rise in blood pressure produced by bombesin, especially in its duration.

As in the rat, antidiuresis was accompanied by reduction in glomerular filtration rate (creatinine clearance) and in renal blood or plasma flow (washout of ^{85}Kr and ^{3}H-*p*-aminohippurate clearance). The urine eliminated during moderate bombesin antidiuresis (20–50% reduction of urine flow) had a reduced concentration of sodium.

Results obtained in some typical experiments were as follows. 1 ng/kg per min of bombesin infused for 30 min elicited a 50% reduction of urine volume accompanied by a 50% reduction of creatinine clearance and a 48% reduction of *p*-aminohippurate clearance. Component I of the ^{85}Kr washout curve (outer cortical flow) was reduced