Locomotor Activity and Regional Brain Noradrenaline Levels in Rats Treated with Prenylamine

Brain monoamines have been related to control of spontaneous locomotor activity. According to much evidence obtained by different approaches, catecholamines seem to exert a positive effect on motility: a) Intracerebral injection of noradrenaline induces hyperactivity in rats[1]; b) Adrenergic drugs are able to increase openfield locomotion[2,3]; c) Administration of catecholamine precursors (i.e. L-DOPA) cause a striking increase of motility and other complex behavioral signs[4]. On the other hand, reserpin- or prenylamine-induced depletion of brain catecholamines (and serotonin) reduces motor activity[5,6].

Our present report is concerned with simultaneous studies of locomotion and regional brain noradrenaline determinations in rats treated with different doses of prenylamine. In order to exclude a possible role of serotonin, low doses of prenylamine known to exhibit in rats a more marked effect on catecholamines[7,8] were employed.

Methods. Holtzman adult male rats weighing 180–200 g were employed. Animals were maintained in a room at 22–25 °C and controlled light. They were fed with Forramez laboratory rat chow and water ad libitum. Prenylamine (N-3'phenyl-propyl-(2')-diphenyl-propyl-(3)-amine gluconate) was injected s.c. at single doses of 10, 25 and 50 mg per kg. Controls were injected with isotonic glucose solution. Spontaneous locomotor activity and brain noradrenaline (NA) were determined at different times after prenylamine injection. Homogenates of 4 different zones of brain, hypothalamus, thalamus, mesencephalic tegmentum and cerebral cortex[9] were prepared in 10 ml of 0.4 *N* perchloric acid immediately after killing the animals by

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Table I. Effect of different doses of prenylamine on spontaneous motor activity

Doses (mg/kg)	Time after injection (h)							
	0		1		4		6	
0	33.5 ± 0.98[a]		—		26.0 ± 0.31		—	
10	34.5 ± 0.43	(8)[b]	39.7 ± 0.76	(4)	14.7 ± 0.36[d]	(8)	—	
0	50.5 ± 5.48		52.8 ± 3.12		54.6 ± 7.97		34.0 ± 3.08	
25	45.1 ± 4.28	(17)	29.8 ± 5.98[c]	(9)	25.5 ± 2.06[c]	(9)	6.7 ± 2.59[d]	(8)
0	31.1 ± 3.19		15.0 ± 1.70		10.0 ± 2.18		15.7 ± 3.18	
50	38.8 ± 3.75	(31)	9.1 ± 2.80[d]	(29)	2.7 ± 0.90[d]	(34)	3.8 ± 1.16[d]	(30)

[a] Mean ± standard error of the Mean. [b] Number of animals employed. Significant difference vs. controls 0 h: [c] $P < 0.05$; [d] $P < 0.01$. —, not determined.

Table II. Levels of noradrenaline in brain regions of rats treated with prenylamine

Dose mg/kg		Time after injection (h)		
		1	4	6
		Hypothalamus (1.84 ± 0.13)		
10	(12)[b]	2.06 ± 0.15	1.25 ± 0.10[d]	1.38 ± 0.17[c]
25	(12)	0.96 ± 0.33[c]	1.14 ± 0.18[d]	0.58 ± 0.22[d]
50	(18)	0.99 ± 0.23[d]	0.73 ± 0.21[d]	1.04 ± 0.27[c]
		Cerebral cortex (0.24 ± 0.03)		
10	(12)	0.16 ± 0.09	0.26 ± 0.08	0.14 ± 0.009[d]
25	(12)	0.19 ± 0.04	0.20 ± 0.04	0.20 ± 0.02
50	(21)	0.23 ± 0.03	0.12 ± 0.03[c]	0.12 ± 0.01[c]
		Thalamus (0.74 ± 0.11)		
10	(12)	0.98 ± 0.08	0.75 ± 0.05	0.51 ± 0.04
25	(12)	0.40 ± 0.10	0.57 ± 0.10	0.31 ± 0.09[c]
50	(18)	0.46 ± 0.01[c]	0.27 ± 0.07[d]	0.32 ± 0.06[c]
		Mesencephalon (0.52 ± 0.04)		
10	(12)	0.92 ± 0.10	0.63 ± 0.20	0.41 ± 0.03
25	(12)	0.41 ± 0.05	0.79 ± 0.14	0.41 ± 0.13
50	(21)	0.41 ± 0.09	0.39 ± 0.09	0.38 ± 0.06

[a] Noradrenaline, control level, in micrograms per g of wet tissue, Mean ± Standard error of the mean. [b] Number of animals employed at each dose. Significant difference vs control: [c] $P < 0.05$; [d] $P < 0.01$.

decapitation. NA was measured by a fluorimetric method[10] in eluates obtained from alumina columns. An open-field test was used to evaluate the running activity of rats. The apparatus briefly consisted of a black square, 1 m in diameter divided into 16 compartments. The activity was represented by the total number of squares (25 cm² each) traversed during periods of 3 min. Prenylamine-treated and control animals were tested before and at different times after injection of the drug or the vehicle. Differences between groups were considered significant when $P < 0.05$ employing the Student's t-test.

Results. Locomotor activity. It is obvious from Table 1 that the motor activity scores of controls displayed a wide variation from 1 experiment to another. This fluctuation may be attributed to the influence of ambient conditions and spontaneous fluctuations as was previously observed by other authors[8]. In consequence, to examine the effects of prenylamine, controls and drug-treated rats were simultaneously tested in each experiment.

Prenylamine gluconate injected s.c. at doses of 25 and 50 mg/kg produced signs of sedation and a significant decrease of scores (about a 200% decrease) in the open-field test. Sedation began within 1 h and remained during the 6 h of observation. Prenylamine injected at doses of 10 mg/kg caused a 50% decrease of locomotor activity 4 h after injection as shown in Table I but no obvious changes were observed within 1 h.

Noradrenaline content. Concentration of noradrenaline in hypothalamus was reduced at 1,4 and 6 h after injection of 25 and 50 mg/kg of prenylamine (Table II). No differences in hypothalamic noradrenaline between controls and treated rats with 10 mg/kg were detected at 1 h period. In the following hours, a slight decrease took place. NA levels in the cerebral cortex were reduced with the highest doses. Instead, in animals treated with 10 mg/kg NA concentration was normal 1 h before sacrifice; longer intervals (6 h) were required to show a decrease. Thalamic NA fell with doses of 25 and 50 mg/kg. No changes in NA of mesencephalic tegmentum were observed with the 3 different doses of prenylamine employed.

Comments. Our results give further information on relationships between brain NA levels and locomotor activity. The role of DA remains to be evaluated Prenylamine, a drug which depletes brain NA causes, at the same time a marked decrease of motility as shown by the open-field test. According to these data such correlation is more close when hypothalamic NA concentration is related to locomotor activity scores. Fall of hypothalamic NA after prenylamine administration has been previously reported[7]. In the other cerebral regions studied, a reduction in NA levels did not correspond to changes in motor activity. It is probable that modifications of hypothalamic NA have functional significance in the control of sponta neous activity. In this connection, it was recently reported that localization of minute amounts of NA into the hypothalamus induces hyperactivity in rats[11,12].

Resumen. Ratas inyectadas con diferentes dosis de prenylamina (gluconato) presentaron una marcada disminución de la actividad motora espontánea y de los niveles de noradrenalina en el hipotálamo, tálamo y corteza cerebral, pero no en el tegmento del mesencéfalo. Se detectó una estrecha relación entre la caída de la concentración de NA hipotalámica y la disminución de la actividad espontanea con las diferentes dosis de prenylamina empleadas.

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The Effect of Oxymetholone on Tissue Replacement in the Rabbit's Ear

Male sex hormones affect protein metabolism and promote the growth of muscle and bone. They can also stimulate the production of new tissue at sites of injury[1]. The risk of virilization limits the use of such substances and new anabolic drugs have been synthesized in an attempt to reduce androgenic effects while retaining anabolic properties. The clinical efficacy of these compounds as anabolic agents has been reviewed by MOLDAWER[2]. One such drug is oxymetholone (2-hydroxymethylene-17α-methyl-17β-hydroxy-5α-androstan-3-one). It has an advantage in that it can be administered orally. It is structurally related to testosterone but has a higher anabolic effect and an androgenicity of only 1/7th that of testosterone[3].

Material and methods. The effect of oxymetholone on tissue replacement has been tested using the rabbit's ear as the replacement site. If a hole, 1 cm square, is made through the full thickness of the external ear and the tissue completely removed, the tissue removed is replaced by skin and elastic cartilage. The growth of the new tissue can be measured throughout the replacement period without causing further trauma. The technique used involves the calculation of the surface area of the new tissue and has been described elsewhere[4]. All areas are recorded as percentages of the initial lesion to aid comparison.

Tissue was removed from the ears of adult rabbits under pentobarbitone anaesthesia. 3 groups of rabbits were given oxymetholone in distilled water (5 mg/ml) 5 times each week. The dose was 5 mg/3 kg initial body weight. In the first group (4 females) the steroid was administered by stomach tube. This procedure was very stressful and the other groups, 1 of 6 males and 1 of 6 females, were given the steroid into the buccopharynx. A group of control females was given 1 ml/3 kg body weight of the carrier by the same route. There were 2 untreated control groups, 1

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