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EFFECT OF L-DIHYROXYPHENYLALANINE IN BEHAVIOR OF RATS AND ON BRAIN CATECHOLAMINE METABOLISM OF RATS DIFFERING IN THEIR LEVEL OF EMOTIONAL-BEHAVIORAL REACTIVITY

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Differences in the neurochemical characteristics of inbred animals determine individual variability of the effects of drugs [10]. Among the population of noninbred animals (rats) groups differing significantly in sensitivity to psychotropic drugs also are distinguished, and this is associated with individual variations in catecholamine (CA) metabolism [2]. This is essential when the neurochemical mechanisms responsible for behavioral disturbance following injection of L-dihydroxyphenylalanine (L-dopa) — a dopamine (DA) precursor are studied. The aim of this investigation was to study L-dopa-induced changes in the behavior of rats differing in their level of emotional-behavioral reactivity (EBR) and concentrations of L-dopa, CA, and dihydroxyphenylacetic acid (DOPAA) in their brain structures.

EXPERIMENTAL METHOD

Experiments were carried out on noninbred male albino rats weighing 180-220 g. The animals were selected according to their EBR level on the basis of qualitative and quantitative parameters of behavior in the open field test (OF), in freely revolving drums (FRD), and in the extrapolation avoidance test (EAT) [1]. In OF the number of rearings in the central zone, the number of crosses from one square to another, the number of holes sniffed, and the number of acts of grooming were estimated; in FRD locomotor activity was assessed (in meters during 10-min intervals for 30 min after placement), and in EAT the latent period (LP) of motor activity, the number of attempts to run away, and LP of avoidance. Animals of two batches of identical composition (each consisting of groups of rats A and B differing in their EBR level) were given an interperitoneal injection, 3 days after testing, of 0.9% NaCl (control batch) or of the preparation Madopar-125 (from Galenika, Yugoslavia) in a dose of 150 mg/kg (which contains 100 mg of L-dopa and 25 mg of benserazide, an inhibitor of peripheral aromatic amino-acid decarboxylase), and 1 h later, half of the animals from the A and B groups (eight rats in each case) of each batch were again tested and the remaining half of the animals killed by decapitation.

Concentration of L-dopa, DA, noradrenalin (NA), and DOPAA in the brain structures of the rats were determined by high-pressure liquid chromatography with electrochemical detection [6]. The results were subjected to statistical analysis by the Wilcoxon—Mann—Whitney test.

EXPERIMENTAL RESULTS

Animals of group B differ from rats of group A in a number of behavior parameters (Table 1). The intensity of biased activity (the number of acts of grooming), reflecting conflict

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TABLE 1. Effect of Madopar on Behavior of Rats of Groups A and B (M ± m)

Test	Parameter	Experimental conditions	A	B
OF	Number of rearings in central zones	K	1,8±0,04	2,8±0,1*
		M	0,2±0,001**	0,1±0,003**
	Number of holes sniffed	K	9,3±1,1	13,2±1,2*
		M	0,7±0,2**	0,9±0,2**
	Number of acts of grooming	K	0,6±0,15	2,8±0,3*
		M	4,7±0,9**	5,1±1,4**
FRD	Number of squares crossed	K	61,4±2,4	54,4±1,8*
		M	33,2±1,5**	31,0±2,1**
	Locomotor activity, % of total, during 30 min			
	1—10 min	K	37,5±2,1	70,9±3,4*
		M	31,3±5,2	31,0±6,6**
	11—20 min	K	21,8±1,7	19,1±2,1
		M	30,8±1,2**	39,3±4,1**
	21—30 min	K	39,5±2,8	6,9±0,5**
		M	35,0±3,6	30,0±1,1**
EAT	Latent period of motor, activity, sec	K	4,9±0,6	5,7±0,4
		M	3,5±0,8**	3,6±0,3**
	Number of unsuccessful attempts at running away	K	2,3±0,8	28,3±3,1*
		M	120,0±5,4**	117,0±9,2**
	Latent period of avoidance, sec	K	8,8±0,5	43,2±2,4*
		M	none **	none **

Legend. K) Control; M) Madopar. *$p < 0.01$ for comparison of groups A and B in control test; **$p < 0.01$ for comparison of K and M.

between fear and investigation motivation, was greater in the rats of group B, which were characterized by more frequent ambulations and rearings in the central zones of OF, and also by an increased number of holes sniffed. Intensive investigative activity leads to a decrease in the number of squares crossed by the animals of this group, and may be the cause of the rapid habituation to new conditions in FRD. However, predominance of intensive locomotor activity in the initial period of the stay of the group B rats in the drums, and also the large number of unsuccessful attempts at running away at EAT are evidence of increased reactivity of the animals of this group to novelty.

Differences in the behavior of intact animals of groups A and B correlate with differences in concentrations of L-dopa, DA, and NA, and also with the rate of DA turnover, determined by the DOPAA/DA ratio (Table 2). An increased L-dopa concentration in all the brain structures of the group A rats investigated may not necessarily reflect differences in activity of the synthesizing (tyrosine hydroxylase) and metabolising (aromatic amino-acid decarboxylase) enzymes [4]. The basal level of L-dopa is mainly determined by the rates of its arrival in two pools — slowly and rapidly metabolized (connected with DA formation [11]. The intensity of metabolism of L-dopa (DA/L-dopa) was significantly lower in all brain structures investigated in the animals of group A (by 4.2 times for the striatum, by 6.6 times for the nucleus accumbens, by 1.4 times for the hypothalamus), which may be connected with the fact that L-dopa is located predominantly in the slowly metabolized pool. Conversely, for rats of group B L-dopa may perhaps enter the rapidly metabolized pool, leading to an increased DA concentration in the striatum and NA concentration in the nucleus accumbens — structures connected with locomotor and emotional expression [5]. This assumption is confirmed by the absence of differences between the groups in the DOPAA level, reflecting inactivation of DA in intact animals with the aid of MAO in the process of DA-mediation [9].

Madopar in a dose of 125 mg/kg disturbed the behavior of the animals of both groups equally, sharply increased the number of attempts to run away in EAT, and abolished ability to escape from an acute stress situation. In OF, investigative activity was inhibited, and

TABLE 2. Differences between Groups in CA, DOPAA, and Dopa Concentrations in Intact Animals and Animals Receiving Madopar (M ± m)

Experimental conditions	Brain structure	Group of rats	Concentration, μmoles/g tissue				DOPAA/DA
			dopa	NA	DA	DOPAA	
Control	Striatum	A	0,32±0,04*	1,2±0,1	37,0±4,2*	5,4±1,2	0,120±0,65*
		B	0,21±0,04	1,2±0,1	51,6±8,8	4,6±1,6	0,058±0,001
	Nucleus accumbens	A	1,18±0,13*	2,2±0,6*	41,4±3,8	6,2±1,4	0,164±0,09**
		B	0,43±0,04	2,9±0,2	49,4±9,0	6,3±0,8	0,128±0,01
	Hypothalamus	A	1,45±0,7**	7,6±1,3	4,7±1,8	1,7±0,5	0,243±0,01*
		B	0,69±0,3	7,7±0,5	3,1±0,6	2,3±0,2	0,493±0,027
Madopar	Striatum	A	44,3±5,9*'***	1,2±0,3	130,8±27,6***	72,8±21,6*,***	0,450±0,02
		B	17,8±4,4***	1,2±0,3	122,4±17,4***	26,8±2,6***	0,216±0,019***
	Nucleus accumbens	A	58,2±5,9*'***	2,0±0,4	86,0±26,2***	79,0±18,4*,***	0,940±0,37*,***
		B	29,6±6,1***	3,0±0,5	84,4±14,8***	32,0±6,6***	0,368±0,072***
	Hypothalamus	A	68,6±11,4*'***	4,5±0,6***	14,7±4,6***	37,5±11,4*,***	2,60±0,18*,***
		B	30,7±6,3***	6,2±0,6	12,0±1,4***	9,4±0,9***	0,920±0,11***

Legend. *p < 0.01, **p < 0.05 for comparison of groups A and B; ***p < 0.01 compared with control of the same group.

stereotyped running in FRD (Table 1). This was accompanied by a considerable rise of the DA level in all brain structures of animals of both groups investigated and by disappearance of differences between the groups for this parameter. The absence of proportionality of the DA concentration (most of which belongs to the vesicular pool) with the L-dopa concentration following administration of Madopar may be connected with the development of saturation of the vesicular DA pool in animals of both groups and with reduction of utilization of this pool as the result of inhibition of spontaneous electrical activity of DA-neurons during L-dopa loading [3]. Under these conditions the main contribution to the DOPAA concentration may be made by cytoplasmic DA which, during L-dopa loading, is synthesized mainly from exogenous L-dopa. Direct dependence between the L-dopa concentration and the DOPAA level (the DOPAA/dopa ratio was 1.2 ± 0.03 from the striatum and nucleus accumbens and 0.6 ± 0.01 for the hypothalamus in the animals of both groups), confirms this hypothesis. The significant fall in the NA level in the hypothalamus of the animals of group A, characterized by a raised DOPAA concentration, may also be the result of an increase in the cytoplasmic DA pool, for exhaustion of NA during L-dopa loading is caused by its displacement by cytoplasmic DA from deposition sites [7]. Madopar in a small dose (50 mg/kg) significantly disturbed behavior of the animals in EAT, mainly in the case of intact group A rats with a lowered EBR and a relatively large volume of rapidly metabolized cytoplasmic DA (determined from the DOPAA/DA ratio). Individual differences in dopa and DA compartmentation evidently have a significant influence on DA metabolism in the animals of the groups distinguished in these experiments, manifested as different levels of emotional-behavioral expression in stress situations and after administration of small doses of Madopar. Excessive saturation of brain structures with DA cancels out differences in the behavior of the groups of animals distinguished above to stress, possibly due to a nonlinear relationship between the subsequent rise in mediator concentration, "occupation" of the receptor, and physiological effect with high DA concentrations [8].

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