CHANGES IN BRAIN MONOAMINE OXIDASE ACTIVITY DURING CONDITIONED PASSIVE AVOIDANCE RECALL IN RATS

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Involvement of the serotoninergic and dopaminergic systems of the brain in the mechanisms of memory regulation is now not in dispute. Much evidence has been obtained, both of changes in conditioned-reflex activity during an experimental decrease or increase in their activity, and also of changes in serotonin and dopamine levels in brain structures during learning [2, 6, 10-12]. However, the biochemical mechanism of regulation of activity of the brain serotoninergic and dopaminergic systems in the process of skill preservation has not yet received adequate study. The author showed previously [5] that during conditioned passive avoidance (CPA) recall the functional activity of postsynaptic type 1 serotonin receptors is reduced in the amygdaloid complex (AC) and central gray matter of the midbrain, structures responsible for one-stage learning [4], and caused by predominance of low-affinity binding in trained animals.

In the investigation described below the presynaptic component of function of the brain serotoninergic and dopaminergic systems was studied: activity and the catalytic properties of monoamine oxidase (MAO), an enzyme participating in serotonin and dopamine metabolism and regulating their level and reuptake in presynaptic endings [8, 13], in conditioned rats, were determined. According to existing data [3, 9], the use of an MAO inhibitor disturbs conditioned-reflex activity, but it is not known how the function of the MAO itself changes during learning in different brain structures involved in cognitive activity. Elucidation of this problem was also an aim of the present investigation.

METHODS

Experiments were carried out on male Wistar rats weighing 180-200 g. PCA was formed by the usual method [7] in an apparatus consisting of two compartments — lit (safe) and dark (punished), where the rats received a single electrodermal stimulus (0.75 mA, 2 sec). The latent period (LP) of moving from the lit compartment into the dark compartment was recorded (time of observation 180 sec). The criterion of learning was an LP of the move during animals of not less than 180 sec after 24 h. Rats with LP of the move of between 2 and 30 sec were classed as untrainable. Rats placed in the apparatus but not subjected to electrodermal stimulation served as the control. Immediately after testing the animals were decapitated, the brain was removed, and in the cold, the frontal cortex, striatum, hippocampus, and amygdaloid complex were isolated. The tissue was homogenized in 0.32 M sucrose. Unpurified synaptosomes (P_2) were isolated by differential centrifugation from the homogenates, and their MAO activity was determined by the method described previously [1]. The protein concentration in the homogenates was determined by Lowry's method. Serotonin creatinine-sulfate ("Reanal") and dopamine hydrochloride ("Serva") were used as substrates. To calculate the kinetic parameters of the reaction of oxidative deamination of serotonin and dopamine, namely the Michaelis constant (K_m) and the maximal reaction velocity (V_{max}), initial reaction velocities were measured in 6 or 7 different substrate concentrations. Apparent K_m and V_{max} were found by the Cornish—Bowden method [2]. The results were evaluated by Student's t test.

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TABLE 1. MAO Activity (in nmoles ammonia/mg·min) during Deamination of Serotonin (3.0 mM) and Dopamine (3.0 mM) in Different Brain Structures in Control Rats and Rats Trained in CPA (M ± m)

Brain structures	Control		Conditioned	
	serotonin	dophami	serotonin	dopamine
Amygdaloid complex	2,6±0,1 (5)	1,6±0,3 (3)	3,4±0,3* (6)	1,6±0,3 (3)
Striatum	2,5±0,3 (8)	1,7±0,1 (7)	1,8±0,1* (6)	1,1±0,1* (8)
Frontal cortex	1,9±0,2 (8)	1,7±0,7 (7)	1,9±0,2 (7)	0,6±0,1* (8)
Hippocampus	2,4±0,2 (6)	1,8±0,2 (7)	1,8±0,2 (5)	1,5±0,2 (4)

Note. Here and in Table 2, significant differences from control indicated by asterisk. Numbers of experiments given in parentheses.

TABLE 2. Kinetic Parameters (K_m, in mM, and V_{max}, in nmoles/mg·min) of Serotonin and Dopamine Deamination Reaction by MAO in Various Brain Structures of Control and Conditioned Rats (M ± m)

Brain structures	Control				Conditioned			
	serotonin		dopamine		serotonin		Dopamine	
	$K_м$	V_{max}	$K_м$	V_{max}	$K_м$	V_{max}	$K_м$	V_{max}
Amygdaloid complex	0,14±0,04	3,1±0,4	—	—	0,04±0,01	3,4±0,3	—	—
Striatum	0,05±0,01	2,8±0,2	0,09±0,01	2,1±0,1	0,10±0,03	2,0±0,2*	0,05±0,02	1,4±0,1*
Frontal cortex	—	—	0,01±0,00	1,8±0,1	—	—	0,12±0,02*	1,9±0,2

RESULTS

Data on deamination of serotonin and dopamine by MAO in different brain structures are given in Table 1. In rats not trained in PCA, activity of the enzyme did not differ from that in animals of the control group. In trained rats, MAO activity was changed in such brain structures as AC, the striatum, and frontal cortex. No changes in enzyme activity were found in the hippocampus. MAO activity in AC during serotonin deamination increased, indicating more rapid destruction of serotonin in this brain structure during conditioned passive avoidance recall in rats. Deamination of both serotonin and dopamine was changed in the striatum. By contrast with AC, a decrease in MAO activity was found in the striatum during deamination of serotonin. During deamination of dopamine, MAO activity was reduced in the striatum and frontal cortex.

The kinetic studies showed that rates of deamination of serotonin in AC were increased in the conditioned rats throughout the range of serotonin concentrations tested (from 0.025 to 0.5 mM). In the presence of low, near-physiological, serotonin concentrations MAO activity was doubled. The Michaelis constant for the serotonin deamination reaction in AC was reduced by two-thirds, but the maximal reaction velocity was unchanged (Table 2). The results are evidence that the increase in MAO activity in AC of the conditioned rats was connected with increased affinity of the enzyme for serotonin. Changes in the velocity of the serotonin deamination reaction in the striatum are shown in Fig. 1b. MAO activity in the region of low serotonin concentrations was lowered by 50-67%. The decrease in enzyme activity in the striatum of the conditioned rats was mainly connected with a decrease in the maximal velocity of the serotonin deamination reaction (Table 2). It is possible that the decrease in MAO activity also was influenced by a decrease in affinity of the enzyme for serotonin, since K_m was doubled, although the increase was not statistically significant ($P > 0.05$). A decrease in the velocity of dopamine deamination by MAO in the striatum of the conditioned rats also was due to a reduction of the parameter V_{max} by 1.5 times (Table 2). A change in the velocity of dopamine deamination also was demonstrated in the frontal cortex. With dopamine in a concentration of 3 mM, MAO activity was below V_{max}, evidence of substrate inhibition of the enzyme in the range of high saturating dopamine concentrations.

In the frontal cortex a marked (eightfold) increase in K_m was found without any change in the maximal reaction velocity (Table 2), evidence of a definite decrease in the affinity of MAO for dopamine in the conditioned animals.

The results indicate that MAO is involved in the process of memory trace recall, for no change in activity of the enzyme was discovered in the trained rats but not in the untrained rats which did not reproduce the conditioned response. Recall of the memory trace was accompanied by a change in the catalytic properties of MAO in the brain structures associated with cognitive activity. Its more intensive degradation in AC took place on account of an increase in affinity of the enzyme for serotonin, evidently because of the inhibitory action of serotonin on neuronal activity in the amygdala [14], which is involved in the evaluation of information reaching the brain and selection of signals for subsequent recall [4]. It can be tentatively suggested that weakening of postsynaptic receptor binding of serotonin in AC, which the author demonstrated previously [5], reduced the inhibitory effect of the serotoninergic system on neurons of the amygdala and is accompanied by activation of the emotiogenic regulatory system of memory [4]. As a result of a decrease in serotonin binding by the postsynaptic membrane, its level in the synaptic space evidently rises, with a corresponding increase in reuptake of the mediator and its further destruction by MAO. Deamination of dopamine in AC during recall of the conditioned response was unchanged. Unlike in the amygdala, the level of serotonin turnover was unchanged but degradation of dopamine by MAO was reduced on account of a decrease in affinity of the enzyme for the mediator. The lowered level of dopamine and serotonin metabolism discovered in the striatum is connected with a decrease in the turnover rate of the enzyme.

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