ANALYSIS OF MONOAMINERGIC MECHANISMS OF THE DORSAL HIPPOCAMPUS PRODUCING THE CONDITIONED AVOIDANCE REACTION IN RATS

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The local microinjection of dopamine (DA), noradrenalin (NA), and serotonin (5-HT) into the dorsal hippocampus of rats in doses of 5 μg did not affect muscle tone or spontaneous motor activity but lengthened the latent period of the conditioned avoidance reflex. The inhibition of the reflex by DA took place through receptors of neurons sensitive to haloperiodol only. The effect of NA was abolished by α-adrenolytics. Perphenazine did not change the inhibitory effect of all the bioamines on the conditioned defensive reflex, but if given before NA the latter significantly stimulated the motor activity of the rats. After melipramine, DA inhibited the avoidance reaction more strongly and stimulated the spontaneous motor activity, whereas microinjection of NA under the same conditions was followed by inhibition of the animals' motor activity.

KEY WORDS: hippocampus; avoidance reaction; biogenic amines.

The hippocampus exerts inhibitory control over the integrative mechanisms forming conditioned defensive reflexes [3, 14, 17]. However, the neurochemical mechanisms of this effect have not been explained. The high level of noradrenalin (NA) and serotonin (5-HT) in the neuronal structures of the hippocampus [16], decreasing in response to electrical stimulation of the ascending monoaminergic pathways [2], and also the marked sensitivity of single hippocampal neurons to dopamine (DA), NA, and 5-HT [12] could indicate the functional significance of the hippocampal bioamines in the mechanisms of conditioning.

The neurochemical mechanisms of the effects of bioamines (DA, NA, 5-HT) injected into the dorsal hippocampus on the conditioned avoidance reaction of rats were studied.

EXPERIMENTAL METHOD

Experiments were carried out on 24 male rats weighing 210-330 g. A conditioned avoidance reflex was produced in a special chamber by the method described previously [5, 7]. When the conditioned reflex had become stabilized, a cannula through which the microinjections of the solutions of the test substances were given was inserted into the animals under ether anesthesia, in accordance with an atlas of the rat's brain [2], by means of the SÉZh-2 stereotaxia apparatus into the right dorsal hippocampus (area CA_1). The effect of the monoamines on the conditioned avoidance reflex was assessed from the duration of its latent period, changes in which were estimated 5 and 30 min after injection of the monoamines into the hippocampus. To analyze the receptor structures through which the catecholamines exerted their effects, the α-adrenergic blocking agents chlorpromazine, phentolamine, and dihydroergotamine (DET) were injected 15 min before them into the hippocampus. Deseryl (IMα-491), chlorpromazine, DET, and haloperidol were used as effective antagonists of 5-HT and DA. The effect of perphenazine (5 μg) and melipramine (15 μg) on the effects of the biogenic amines also was studied. The NA, DA, 5-HT (5 μg of the base) and their antagonists (in doses of 1.25-5 μg of the base) were given in a volume of 2-5 μl by means of the mi-

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TABLE 1. Effect of Bioamines and Their Antagonists, Injected into the Dorsal Hippocampus, on Conditioned Avoidance Reaction, Motor Activity, and Muscle Tone of Rats (M ±m)

Drug	Dose (in μg)	No. of expts.	Latent period of conditioned avoidance reflex (in sec)		Motor activity (pulses/25 min)	Muscle-relaxing action (% of animals falling off the shaft)
			5 min after injection	30 min after injection		
Bidistilled water	5 μl	5	1,23±0,12	0,98±0,09	51±7 4	0
DA	5	5	2,12±0,09*	2,55±0,12*	51,4±5,9	0
NA	5	5	2,1±0,09*	2,39±0,18*	51,8±5,4	0
5-HT	5	5	2,12±0,08*	2,32±0,1*	49,8±6,1	0
Phentolamine	5	4	1,49±0,12	1,13±0,08	49,5±8,9	0
Chlorpromazine	5	4	1,61±0,15	1,11±0,09	47,7±7,5	0
DET	5	4	1,12±0,15	1,12±0,13	58,2±7,9	0
Deseryl	1,25	4	1,06±0,06	1,1±0,03	54,2±7,7	0
Haloperidol	5	4	1,16±0,09	1,06±0,09	52,5±6,4	0
Perphenazine	5	5	1,3±0,2	1,34±0,2	48,6±5,7	0
Melipramine	15	4	2,2±0,15	1,15±0,06	49,0±2,9	0

*Differences from control statistically significant ($P < 0.05$).

croinjection system. To study the selectivity of action of the monoamines on the conditioned avoidance reaction, their muscle-relaxing action and their ability to modify the spontaneous motor activity of the rats were investigated simultaneously [5, 7]. A control group of animals received an injection of 5 μl bidistilled water. The site of injection of the preparations into the hippocampus was verified histologically. A photomicrograph of a frontal section through the brain of one of the experimental rats is illustrated in Fig. 1.

EXPERIMENTAL RESULTS AND DISCUSSION

Local microinjection of 5 μg DA, NA, or 5-HT into the dorsal hippocampus did not affect the muscle tone or the spontaneous motor activity but it inhibited the conditioned avoidance reflex. This was shown by the fact that the latent period of the reflex recorded 5 and 30 min after microinjection of the amines into the hippocampus (Table 1) was substantially increased. After injection of perphenazine (5 μg), melipramine (15 μg), phentolamine, chlorpromazine, DET, haloperidol (5 μg), and deseryl (1.25 μg) into the hippocampus no appreciable changes in the behavioral activity of the animals could be seen, although in larger doses they induced changes in the parameters recorded.

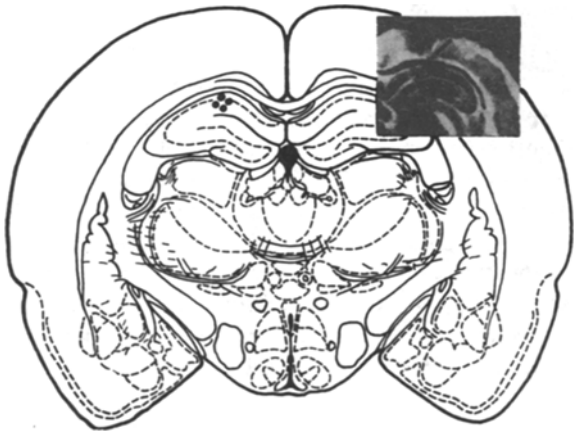

Fig. 1. Scheme of frontal section through rat brain at the level of the hippocampus. Black circles mark sites of microinjections of biogenic amines and their antagonists. Photomicrograph of frontal section through the same zone shown in top right hand corner. Localization of tip of injection cannula visible in the region of the dorsal hippocampus (marked by arrow).

Changes in the conditioned-reflex activity of the rats under the influence of the catecholamines and 5-HT were due to their effects on neurons of the dorsal hippocampus. This was confirmed by control experiments in which thionine solution was injected into the hippocampus (it does not spread outside the hippocampus for 30 min after microinjection) and by the specificity of the neurophysiological effects produced by the bioamines when injected in equal doses into different formations of the limbic system. In fact, microinjection of DA and 5-HT (5 μg) into the amygdala also inhibited the conditioned avoidance reflex, but at the same time it significantly stimulated the spontaneous motor activity of the rats [5].

Analysis of the morphological material showed that the DA, NA, and 5-HT injected into the dorsal hippocampus acted mainly on neurons of the str. oriens and str. pyramidale, and in some experiments, str. lacunosum and str. moleculare. Since the iontophoretic application of NA, DA, and 5-HT to single hippocampal neurons was followed by inhibition of

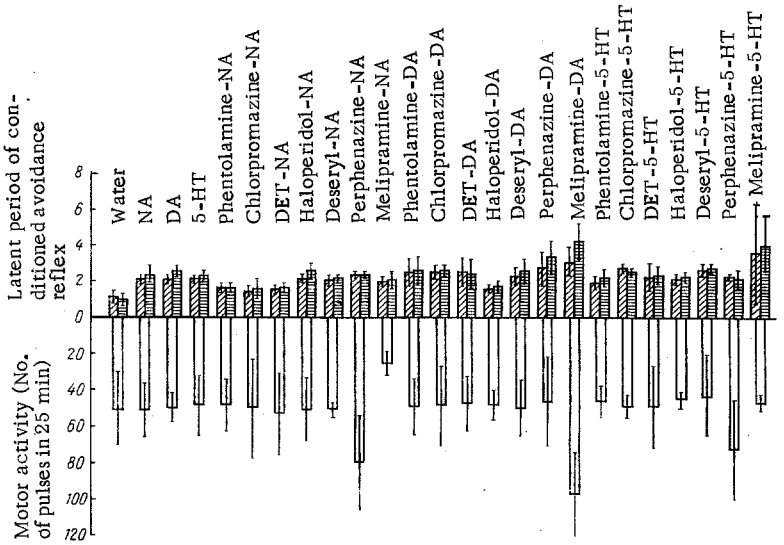

Fig. 2. Effect of phentolamine, chlorpromazine, dihydroergotamine (DET), deseryl, haloperidol, perphenazine, and melipramine on effects of noradrenalin (NA), dopamine (DA), and serotonin (5-HT) when given by microinjections into the rat hippocampus. Obliquely shaded columns denote latent period of conditioned avoidance reflex 5 min, and horizontally shaded columns the same 30 min after injection of bioamines; unshaded columns represent motor activity of rats.

their bioelectrical activity [12], presumably the difficulty in reproducing the conditioned defensive reflex evoked by local microinjection of bioamines into the hippocampus was the result of inhibition of the inhibitory neurons detected in str. oriens or of inhibitory synapses on the hippocampal pyramidal neurons [11], on account of which this limbic structure is activated. The latter is known to interfere with the conduction of impulses in the ascending reticular formation [9] or to lead to excitation of the anterior and intralaminar thalamic nuclei which inhibit cortical neurons [6]. The presence of direct nerve pathways from the hippocampus converging on neurons of the mesencephalic reticular formation or of the anterior and intralaminar thalamic nuclei [3, 7] confirms this conclusion.

Inhibition of the conditioned avoidance reflex by DA when injected into the hippocampus was brought about through receptors of neurons of this structure belonging to the limbic system that are sensitive to haloperidol only. This is shown by the fact that the inhibitory action of this amine on the conditioned defensive reflex was unchanged by phentolamine, chlorpromazine, deseryl, or DET (Fig. 2). Meanwhile the effect of DA was specific, for haloperidol did not affect inhibition of the conditioned defensive reflex evoked by microinjection of NA; the effect of the latter was prevented only by α-adrenolytics. On the other hand, the inhibitory effect of 5-HT on the conditioned avoidance reflex was not connected with the action of the amine on α-adreno-D-serotoninergic or dopaminergic receptors of the dorsal hippocampus, for the inhibition of the conditioned defensive reflex caused by 5-HT was not weakened by chlorpromazine, DET, deseryl, or haloperidol, but was effected through the muscarinic serotoninergic structures of this formation belonging to the limbic system [7].

Perphenazine (5 μg) did not change the inhibitory effect of all the bioamines on the conditioned avoidance reflex, but after the preliminary injection of this neuroleptic into the hippocampus NA essentially stimulated the spontaneous motor activity of the rats.

After the preliminary injection of melipramine into the hippocampus, DA inhibited the conditioned avoidance reflex and stimulated the animals' spontaneous motor activity more strongly. The effect of 5-HT on both these parameters was unchanged. After melipramine, NA inhibited the spontaneous motor activity, although its effect on the conditioned avoidance reflex was not changed by melipramine. It can be concluded from these results that the facilitatory and inhibitory effects of the hippocampus on the evoked and spontaneous activity of the spinal neurons established by several investigators [4] and brought about through the intermediary of the hypothalamic structures and the mesencephalic reticular formation [8] are

monoaminergic in nature. It can be postulated that the changes in motor activity evoked by NA after the preliminary injection of perphenazine or melipramine, and also the changes in behavioral activity evoked by DA after preliminary administration of melipramine, may be attributable to differences in the degree of the effect of these psychotropic drugs on the recapture of DA and NA by the axons of dopaminergic and noradrenergic neurons [1, 13] converging on the hippocampal neurons.

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