# ANTICATALEPTOGENIC EFFECT OF DELTA-SLEEP INDUCING PEPTIDE AND ITS EFFECT ON BRAIN MONOAMINE OXIDASE OF RATS GENETICALLY PREDISPOSED TO CATALEPSY

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Regulation of activity of monoamine oxidase (MAO), an enzyme involved in biogenic amine metabolism, still remains relatively unstudied. Particular attention is drawn to the regulation of brain MAO activity during pathological forms of behavior [9], especially in schizophrenia, which is associated with a disturbance of MAO activity and biogenic monoamine metabolism [1, 7, 12]. There is reason to suppose that rats genetically predisposed toward catalepsy [13] can be used as a model of schizophrenia and to study the characteristics of neurochemical systems in this disease [4]. The writers showed previously [8] that MAO activity, relative to deamination of serotonin and dopamine, in the brain of cataleptic rats differs from the corresponding activity in normal Wistar rats. It is likely that peptides and, in particular, delta sleep inducing peptide (DSIP), which depresses type B MAO activity and enhances type A MAO activity in the brain [2], may be endogenous regulators of MAO activity. The effect of DSIP on brain MAO of rats predisposed to catalepsy has not been studied.

The aim of this investigation was to study the effect of DSIP on the development of catalepsy and on the brain MAO activity of these animals.

## EXPERIMENTAL METHOD

Experiments were carried out on rats predisposed toward catalepsy [13] and selected from an inbred Wistar line. All the animals were aged 4 months and weighed 220-250 g. The rats were put into individual cages 2 days before the experiment, and allowed free access to water and food. The period of daylight and darkness corresponded to the season (November). The experiments were carried out at 9 a.m. Rats which, from the first attempt adopted the vertical posture, forced upon them, and maintained it for at least 10 sec in their own living cages, were considered to be predisposed to catalepsy. The length of stay in the imposed posture was measured in seconds. The motor activity of the cataleptic rats was studied in a separate series of experiments in the open field test [11, 18]. For this purpose the rats were placed on an area marked out in squares, measuring 120 × 80 cm, with transparent edges 20 cm high. The rats' movements were recorded for 5 min as the number of times they crossed from one square into another. Experimental rats were given an intraperitoneal injection of DSIP (synthesized at the M. M. Shemyakin Institute of Bioorganic Chemistry, Academy of Sciences of the USSR and generously provided by I. I. Mikhaleva), 60 min before testing, in a small dose of 12 and a larger dose of 120 $\mu$g/100 g body weight [5]. Control rats received an injection of physiological saline. The effect of DSIP on MAO activity was determined 60 min after injection of DSIP in a dose of 120 $\mu$g/100 g body weight in the experimental rats and 60 min after the injection of physiological saline in the control rats. Together with cataleptic rats, the effect of DSIP on brain MAO also was determined in noncataleptic Wistar rats. After decapitation, the brain excluding the cerebellum was homogenized in 0.32 M sucrose. The unpurified mitochondrial fraction ($P_2$) was isolated by differential centrifugation, as in our previous investigation [8] and MAO activity was determined as in [1]. Serotonin Creatinine-sulfate (Reanal, Hungary) was used as the specific substrate for type A MAO, and benzylamine hydrochloride (USSR) in

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TABLE 1. Some Behavioral Characteristics of Rats Predisposed and Not Predisposed toward Catalepsy in the Open Field Test (M ± m)

| Series of experiments | Number of animals | Time of maintaining enforced posture, sec | Motor activity, number of squares crossed in 5 min | Rearing | Grooming | Hyperkinesias |
|---|---|---|---|---|---|---|
| Cataleptics | 26 | 30,3±8,8* | 42,0±4,0* | 8,4±1,5* | 0* | 0,6±0,2* |
| Noncataleptics | 39 | 8,5±2,8 | 59,0±3,4 | 16,2±1,4 | 4,5±0,8 | 4,1±0,8 |

**Legend.** *$p < 0.05$.

TABLE 2. Changes in Brain MAO (in units of activity) of Rats Genetically Predisposed toward Catalepsy, under the Influence of DSIP (120 $\mu$g/100 g body weight) (M ± m)

| Animals | Series of experiments | MAO A | MAO B |
|---|---|---|---|
| Noncataleptics | Physiological saline | 0,83±0,09 (5) | 2,46±0,17 (6) |
| | DSIP | 1,48±0,05* (5) | 1,72±0,16* (6) |
| Cataleptics | Physiological saline | 1,98±0,06 (4) | 1,84±0,03 (4) |
| | DSIP | 1,95±0,12 (4) | 1,25±0,08* (4) |

**Legend.** *$p < 0.05$ compared with physiological saline. Number of animals given in parentheses.

concentrations of 1 mM was used as the specific substrate for type B MAO. MAO activity was expressed in nanomoles of ammonia formed in 30 min during deamination of the substrate at 37°C per milligram protein per minute. Protein was determined by Lowry's method.

## EXPERIMENTAL RESULTS

The results show that rats predisposed toward catalepsy maintained the vertical posture imposed upon them four times longer than the noncataleptic rats (Table 1). The cataleptics also were characterized by depressed motor activity, by a reduced number of unmotivated stereotyped behavioral acts (rearing, grooming) in the open field test, as well as by a high emotional reaction to handling (the rats struggled to get free, bit, and vocalized).

Under the influence of DSIP in a dose of 120 $\mu$g/100 g the behavior of the cataleptic rats was found to closely resemble that of the noncataleptics. The duration of stay of the cataleptic rats in the enforced posture was reduced by half (Fig. 1), and their emotional excitation during handling disappeared (the rats were calm, did not bite, and did not vocalize). Motor activity of the rats was unchanged in the open field test. The control cataleptic rats, receiving physiological saline, were indistinguishable in all respects from intact cataleptic rats not receiving injections of any kind. A small dose of DSIP (12 $\mu$g/100 g) had no significant effect on behavior, although the tendency for the duration of stay in the enforced vertical posture was maintained.

The action of DSIP on brain MAO of the cataleptic rats differed from its action on brain MAO of the noncataleptic rats. In the cataleptic rats type A MAO was resistant to the action of a high dose of DSIP (120 $\mu$g/100 g) unlike the control noncataleptic rats, in which type A MAO activity was enhanced by DSIP (Table 2). It can be tentatively suggested that in cataleptics type A MAO is insensitive to the action of DSIP because of structural changes in this mitochondrial brain enzyme. This hypothesis is based on our data [3, 8] showing changes in the catalytic properties of the enzyme and a redistribution of its activity in subcellular fractions in cataleptic rats, and also on data [7] which revealed differences in the catalytic properties of an electrophoretically homogeneous preparation of MAO II-$\beta$, isolated from solubilized brain mitochondrial membranes from patients with schizophrellia, but not from healthy subjects.

One functional feature distinguishing type A MAO is that it regulates the level and reuptake of serotonin [14], and also the level, release, and reuptake of dopamine [15], both of which are brain neurotransmitters involved in the genesis of catalepsy [6, 10, 16]. A change in functional activity of the dopaminergic system is regarded as one cause of the genesis of schizophrenia in man [16]. The level, release, and reuptake of dopamine in the brain are also regulated by type B MAO [14, 15]. Our data show that both in cataleptic and in noncataleptic rats DSIP induces a decrease in type B MAO activity in the brain (Table 2).

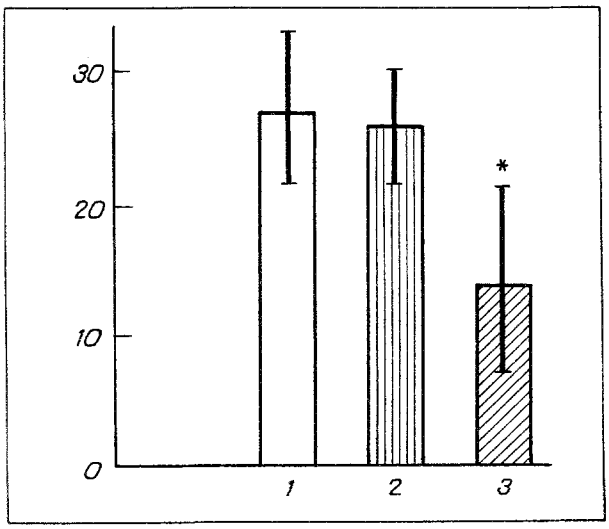


Fig. 1. Duration of stay (in sec) of rats predisposed toward catalepsy in an enforced vertical posture. 1) Intact; 2) physiological saline; 3) DSIP. *$p < 0.01$ by Wilcoxon's test.

The action of DSIP on type B MAO can evidently be realized at the level of degradation of the enzyme, for DSIP is known to regulate proteolysis [5].

The results (Table 2) are evidence that functioning of types A and B MAO in cataleptic rats is uncoordinated, and the type A MAO of cataleptics is evidently unable to create an adequate level, release, and reuptake of serotonin and dopamine, and this may be the true cause of the catalepsy.

However, an adequate reaction of the brain type B MAO of cataleptic rats to DSIP, it can be tentatively suggested, is able to some degree to compensate for function of type A MAO in the case of dopamine deamination, for dopamine is a common substrate for types A and B MAO in the brain.

A particular feature of type B MAO is that it is localized in the rat brain outside the dopaminergic neuron [17] and it can deaminate dopamine only in the case of a high dopamine concentration in the synaptic space, when dopamine reuptake is impeded [15]. The possibility cannot be ruled out that dopamine reuptake in cataleptic rats is impeded, and its deamination by type B MAO takes place outside the dopaminergic neuron. It can be postulated that DSIP, by inhibiting type B MAO, can correct the dopamine level on its receptors, leading to weakening of the catalepsy in rats genetically predisposed to its development. The data given above suggest that the anticataleptogenic effect of DSIP is mediated, perhaps only partly, by inhibition of type B MAO and compensation of the function of type A MAO.

## LITERATURE CITED

1. V. Z. Gorkin and T. A. Moskvitina, Zh. Nevropatol. Psikhiat., **85**, No. 7, 1062 (1985).
2. E. L. Dovedova, N. S. Popova, and L. M. Kachalova, Neirokhimiya, **2**, No. 2, 138 (1983).
3. N. S. Kamyshanskaya, V. Z. Gorkin, and N. N. Voitenko, Vopr. Med. Khim. (1990).
4. V. G. Kolpakov, Essays on the Genetics of Behavior [in Russian], Novosibirsk (1987), p. 186.
5. A. A. Krichevskaya, T. I. Bondazenko, and E. S. Grishchenko, Fundamental Progress in Neurochemistry — Medicine [in Russian], Gor'kii (1987), p. 78.
6. A. V. Kulikov, E. Yu. Kozlachkova, and N. K. Popova, Mediators and Behavior [in Russian], Novosibirsk (1988), p. 56.
7. T. A. Moskvitina, N. S. Kamyshanskaya, and V. Z, Gozkin, Vopr. Med. Khim., No. 1, 98 (1986).
8. N. K. Popova, E. V. Naumenko, and V. G. Kolpakov, Serotonin and Behavior [in Russian], Novosibirsk (1978), p. 197.
9. N. K. Popova, A. V. Kulikov, V. G. Kolpakov, et al., Zh. Vyssh. Nerv. Deyat., **85**, No. 4, 742 (1985).
10. N. K. Popova, N. N. Voitenko, V. G. Kolpakov, and T. A. Alekhina, Neirokhimiya, **7**, No. 4, 535 (1988).
11. R. J. Blanchard and D. C. Blanchard, J. Comp. Physiol. Psychol., **67**, 370 (1969).
12. C. J. Fowler, A. Carlsson, and B. Winblad, J. Neural Transmiss., **52**, 23 (1981).

13. V. Kolpakov, N. Barykina, and I. Chepkasov, Behav. Proc., **6**, 269 (1981).
14. J. C. K. Lai, T. K. C. Leung, J. F. Guest, et al., Biochem. Pharmacol., **29**, 2763 (1980).
15. J. Liccione and A. J. Azzaro, Naunyn-Schmiederberg, Arch. Phazmacol., **337**, 156 (1988).
16. A. V. P. MacKay, Brit. J. Psychiat., **137**, 379 (1980).
17. D. D. Schoepp and A. J. Azzaro, Biochem. Pharmacol., **31**, 2961 (1982).
18. J. M. Weiss, E. E. Krieckhaus, and R. Conte, J. Comp. Physiol. Psychol., **65**, 415 (1968).