PATHOLOGICAL PHYSIOLOGY AND GENERAL PATHOLOGY

Alimentary Modification of Brain Catecholamine Level and Activity of the Thiol-Dependent Protective System under Conditions of Experimental Seizures

Sh. S. Tazhibaev, O. V. Ikkert, L. Zh. Zhumasheva, E. G. Tishchenko, B. U. Sydykov, B. T. Sarbaev, and N. R. Alekseeva

UDC 616.8-009.24-085-092.9:612.82.015]-07

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 115, № 5, pp. 459-460, May, 1993
Original article submitted May 28, 1992

Key Words: *catecholamines; thiol-dependent system; chronic seizure; specialized diet*

Epileptic activity in the CNS may be caused by disturbances in the metabolism of inhibitory neurotransmitters [1] and by free-radical pathology of neuronal membranes [3]. An important role is allocated to the disturbances in catecholamine (CA) metabolism [1,4,5], in particular, to a decrease in tissue levels. Several pathways of CA catabolism are known to be accompanied by the generation of oxygen anion-radical, hydrogen peroxide, and singlet oxygen, supplying the hydroxyl radicals for the Haber-Weiss cycle; the latter are the immediate initiators of free-radical lipid peroxidation (FRLP), which exerts a damaging influence on the membrane structures of nerve cells. On the other hand, CA might improve the antioxidant status of the organism through specific regulation of the protective thiol-dependent mechanisms by activating glutathione-S-transferase (GS-T), and selenium glutathione peroxidase (GSH-Px) and inhibiting γ-glutamyltransferase [2].

Laboratory of Molecular Bases of Protective Effect of Nutrients, Research Center of Regional Problems of Nutrition, National Academy of Kazakhstan, Alma-Ata
(Presented by T. Sh. Sharmanov, Member of the Russian Academy of Medical Sciences)

In the present study possible ways of alimentary modification of CA metabolism and the state of the thiol-dependent system were investigated.

MATERIALS AND METHODS

The experiments were carried out on 100 August male rats with initial weight 140-160 g, divided into 3 series and maintained for 30 days on the following rations: I) complete balanced ration (control); II) a taurine-enriched ration (26.8 ml/kg dry chow); III) a ration enriched with tryptophan (1.55 g/kg chow), copper sulfate (20 mg/kg), nicotinamide (4.8 g/kg), and pyridoxine (120 mg/kg). The enrichment of the diet with the inhibitory amino acid taurine (ration II), the serotonin precursor tryptophan, and with regulators of several steps in the synthesis of serotonin (nicotinamide, pyridoxine, copper), γ-butyrate (pyridoxine) and norepinephrin (pyridoxine, copper) (ration III) served to maintain the levels of the inhibitory transmitters in the brain. The animals of each series were divided into two groups: the 1st group comprised healthy rats, while the 2nd group consisted of rats with seizures receiving anticonvulsant therapy. A corazole kindling was used as an experimental model for chronic seizure, namely,

0007-4888/93/0005-0492$12.50

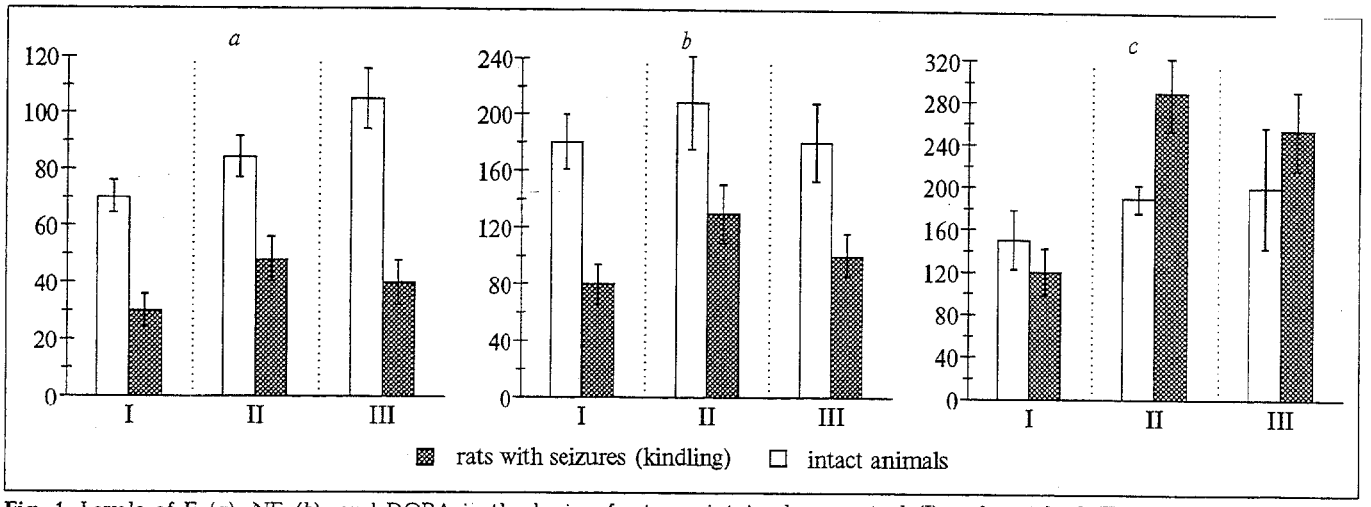

Fig. 1. Levels of E (*a*), NE (*b*), and DOPA in the brain of rats, maintained on control (I) and enriched (II, III) rations.

starting from the 20th day of the experiment the animals were injected intraperitoneally with the seizure inductor metrazole in a dose of 40 mg/kg body weight 6 times at 48-hour intervals. The content of CA in brain homogenate was determined fluorimetrically [6] with a Hitachi-650-60 fluorescent spectrophotometer, the level of SH-groups was measured as described previously [8], and GS-T activity in the microsomal fraction and in the brain cytoplasm was determined as described in [9] using 1-chloro-2,4-dinitrobenzene as substrate. GHS-Px activity in the brain cytoplasm was evaluated in a coupled glutathione reductase system using cumene hydroperoxide as substrate. The microsomal fraction and postmicrosomal cytoplasm were separated by differential ultracentrifugation.

Statistical processing of the results was performed using the Student *t* test; correlation analysis was carried out on an Iskra 1030.11 PC with STATGRAF statistical software.

RESULTS

Seizures caused a marked reduction in the brain epinephrine (E) and norepinephrin (NE) content in rats maintained on ration I (Fig.1). The enriched rations II and III exhibited a protective effect by decreasing reliably the degree of E and NE depletion and elevating the level of DOPA in the brain tissue under conditions of metrazole kindling and anticonvulsant therapy. The mechanism of the protective action of the enriched rations may be attributed to a modification of the CA tissue levels resulting from the dietary supply of the modulators of metabolism of other inhibitory neurotransmitters. This is supported by data on the mutual regulation of the level of inhibitory neurotransmitters in the brain, in particular, an elevation of the NE and dopamine levels in different structures of the brain [12], an increase of NE release from brain sections [10], and activation of dopamin synthesis in neurons of the striatum [7] in response to exogenous taurine. There is also some evidence on a stimulation of the activity of the central NE-ergic receptors after injection of the serotonin precursor 5-hydroxytryptophan [13].

The rats of series I (control ration) had a decreased GSH-Px activity in the brain cytoplasm in comparison with the intact animals (see Table 1). The enriched rations (II and III) resulted in an elevation of all the indexes of the thiol-dependent protective system, with the increase of GSH-Px activity

TABLE 1. Activity of Glutathione–S–Transferase and Glutathione Peroxidase, and Content of SH–Groups in Rat Brain ($M \pm m$)

Ration	Conditions	SH-groups in cytoplasm, μmol/mg protein	GS-T, nmol prod/min×mg protein		GSH-Px in cytoplasm, nmol oxid. NADPH/min×mg protein
			in cytoplasm	in microsomal fraction	
I	intact	31.6±0.6	48.6±8.0	9.7±0.8	58.0±3.6
	kindling	33.0±1.5	46.3±2.8	12.1±2.5	37.9±6.3*
II	intact	42.0±1.9**	87.1±12.6**	17.5±1.6**	105.1±5.7**
	kindling	45.6±4.2**	96.4±3.7**	21.8±0.7**,*	78.8±10.5*,**
III	intact	44.8±2.6**	93.6±17.3**	20.1±3.1**	106.4±6.3**
	kindling	43.1±1.8**	86.7±3.0**	23.6±2.9**	71.4±6.7*,**

Note. Asterisk denote reliably differences ($p<0.05$): * – in comparison to intact rats of the corresponding series, ** – in comparison to rats of the corresponding series, maintained on the control ration. Each experimental series consisted of 6 animals.

in the brain cytoplasm being less pronounced under conditions of kindling than in the intact animals of the corresponding series. Taking into account the fact that the experimental rations were not enriched with immediate modifiers of the thiol-dependent system, the stimulation of this system may be connected with the above-indicated regulatory influence of CA. This is confirmed by the correlation analysis, which revealed a strong positive correlation (r=0.8 and 0.76) between the lipoperoxidase activity of GSH-Px and the levels of E and NE. A strong positive correlation was also found between the level of DOPA, on the one hand, and microsomal GS-T activity (r=0.82), cytoplasmic GS-T activity (r=0.86), and the content of SH-groups (r=0.88) in the brain, on the other.

Taking into account the role of FRLP of neuronal membranes in epileptogenesis, it may be surmised that the protective effect of specialized rations is related not only to the elevation of tissue levels of CA under conditions of an exogenous supply of alimentary precursors and regulators of the synthesis of the inhibitory neurotransmitters, but also to the antioxidant effect occurring through the maintenance of the thiol-dependent protective mechanisms.

Thus, dietary supplementation with inhibitory amino acids, the precursor and regulators of the synthesis of inhibitory neurotransmitters in kindling and under conditions of antiseizure therapy promote the preservation of the tissue levels of CA, which leads to the activation of thiol-dependent protective mechanisms and the further retention of CA in the brain by preventing its oxidative degradation, which is aggravated in the chronic seizure syndrome.

REFERENCES

1. F. G. Vetrogon, *The role of Bioactive Compounds in the Pathogenesis of Epilepsy*. Author's Abstract of Doctoral Dissertation. Tbilisi (1974).
2. L. S. Kolesnichenko, N. S. Mantorova, and L. A. Shapiro, *Biokhimiya*, **52**, № 5, 743 (1987).
3. G. N. Kryzhanovskii, E. V. Nikushkin, V. E. Braslavskii, and R. N. Glebov, *Byull. Eksp. Biol.*, **89**, № 1, 14, (1980).
4. E. Sh. Matlina and A. D. Solov'eva, *The Pathogenesis of Epilepsy* [in Russian], Sofia (1971).
5. V. I. Okladnikov, *Sov. Meditsina*, № 4, 39 (1981).
6. E. M. Stabrovskii and K. F. Korovin, *Nauchn. Trudy Leningr. Inst. Usoversh. Vrachei*, **105**, № 3 (1971).
7. L. Antee and M. L. Vahalu, *Prog. Clin. Biol. Res.*, **179**, 331 (1985).
8. G. L. Ellman, *Arch. Biochem.*, **85**, 70 (1959).
9. W. H. Habig, M. J. Pabst, and W. B. Jakoby, *J. Biol. Chem.*, **249**, № 22, 7130 (1974).
10. R. Kontro and E. R. Korpi, *Neuroscience*, **13**, № 3, 663 (1984).
11. C. Little, R. Olinescu, and K. G. Reid, *J. Biol. Chem.*, **245**, № 14. 3632 (1970).
12. K. Muralidhar, M. A. Udayakumar, P. B. Venkateswara, and B. Saclasivudu, *Neurochimia*, **3**, № 3, 244 (1984).
13. G. A. Smythe and R. M. Gluson, *Neuroendocrinology*, **47**, № 5, 389 (1988).