THE INFLUENCES OF COMBINED TREATMENT WITH XENOBIOTICS ON NEUROPHYSIOLOGICAL FUNCTIONS OF THE BRAIN IN RATS

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ABSTRACT

The whole population is, especially in the developed countries, exposed to environmental pollutants (heavy metals, pesticides) which can affect the nervous system causing considerable changes in certain sensitive processes of the central nervous activity. As people are generally exposed not only by one chemical, and among them there are also the chronic ethanol consumers, the aim of the study was to investigate the functional changes of brain induced by combined treatment with these neurotoxic substances. Low doses of mercury and dimethoate were given daily by gavage to the rats, or the same amounts with parallelous administration of 5 % ethanol in the drinking water in different stages of intra-and/or extrauterine development. The data showed that the combined exposure by the substances caused more expressed alterations of the investigated neurophysiological parameters (electrocorticogram, cortical evoked potential) than their single administration. The results emphasize the importance of further analysis of the combined effects of environmental xenobiotics not only in animal experiments but in the exposed population.

KEY WORDS: mercury, dimethoate, ethanol, electrocorticogram, evoked potential, rat.

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The population is continuously exposed by environmental xenobiotics (heavy metals, organophosphate pesticides) which can affect the nervous system causing pathological (biochemical, functional and morphological) changes (10, 11). At the same time there is a steady, in some regions increasing, number of users of ethanol which, among other symptoms, can induce neurological diseases (1).

In our previous experiments it was stated that relatively low doses of mercury and dimethoate, given in different periods of nervous system development, caused dose- and stage-dependent changes of certain neurofunctional parameters such as electrocorticogram or cortical evoked potentials (4, 7). The aim of this study was to investigate the functional changes of brain caused by combined administration of mercury and dimethoate with ethanol in different stages of intra- and extrauterin development.

10 weeks old female Wistar rats (10 per group) were given by gavage 0.4 (L – low dose) and 1.6 mg/kg (H – high dose) mercury ($HgCl_2$); 5.0 (L) and 20.0 (H) dimethoate; or the same doses in combination with 5 % ethanol in drinking water – from day 5 to 15 during pregnancy (P variation), or from day 5 to 15 of pregnancy + for 4 weeks of lactation (P+L variation), from day 5 to 15 of pregnancy + for 4 weeks of lactation plus the male offspring (F1 generation) were further treated for 8 weeks after weaning (P+L+P variation). The control rats were orally given saline only.

At 12 weeks of the age, from F1 male rats electrocorticogram (ECoG) was simultaneously recorded from the primary somatosensory, visual and auditory cortical centres of anesthetized rats (1000.0 mg/kg urethane ip.) for 15 minutes. The analyzed ECoG parameters were: mean amplitude, mean frequency, power spectrum of the frequency bands. From these cortical centres, sensory evoked potentials were recorded with the same electrodes. Fifty evoked potentials were averaged and latency of the averaged potentials was measured. The statistical analysis of the data was performed by two-factorial ANOVA.

Compared to the control, dose- and treatment variation-dependent changes of the ECoGs were found in the groups given mercury or dimethoate. The mean amplitude was decreased, while the mean frequency was increased, and the activity of slow frequency bands was lower and that of the fast ones was higher (Fig. 1). The 5 % ethanol treatment caused lowered frequency of ECoGs, together with depressed activity of faster part of frequency wave bands. In rats administered with the combination of mercury or dimethoate with ethanol, the changes in the analyzed parameters differed from that caused by the single treatment with the compounds: the ECoG mean frequency in all dose and treatment variation were, compared to the controls, higher but lower than in the single mercury or dimethoate groups (the latter

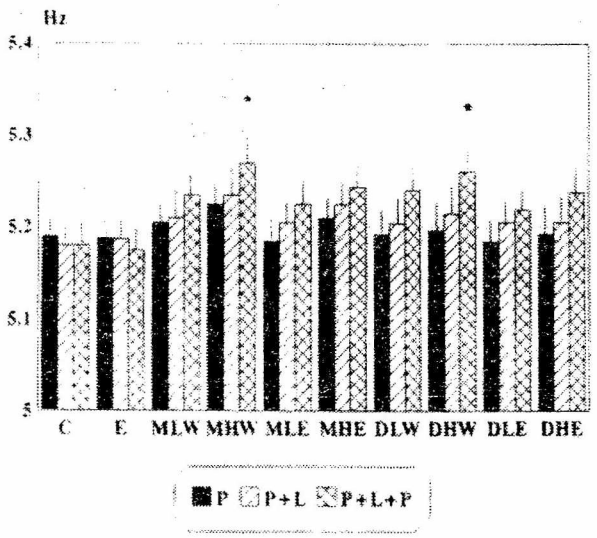

Fig. 1. *Changes of somatosensory ECoG mean frequency*

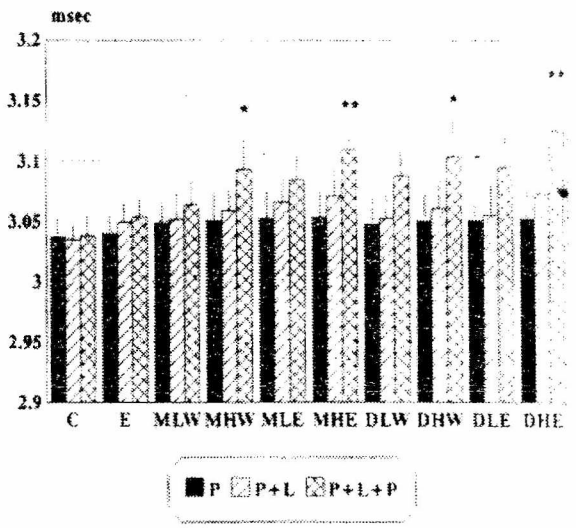

Fig. 2. *Changes of somatosensory latency*

(C: control; E: ethanol; MLW/MHW: mercury low/high dose with water; MLE/MHE: mercury low/high dose with ethanol; DLW/DHW: dimethoate low/high dose with water; DLE/DHE: dimethoate low/high dose with ethanol) *p<0.05; **p<0.01

differences of ECoGs were not significant). The investigated parameters of the visual and auditory ECoGs showed the same tendencies.

Dose- and treatment-variation-dependent increase of the latency of the cortical evoked potentials, caused by the single administration of the substances, was also observed. In all treatment variation groups, the latency of the somatosensory evoked potential of rats receiving mercury, dimethoate or ethanol was lengthened compared to the controls (Fig. 2). The changes were more pronounced in the animals administered with the combination of the compounds. The trends of lengthening of the latency of the visual and auditory evoked potentials were practically the same, the alterations caused by the combined treatment were also more expressed in all treatment variations.

The results pointed to a dose- and developmental stage-dependent susceptibility of the central neurofunctional processes to single inorganic mercury, dimethoate and ethanol exposure. It was also found that the combined mercury/dimethoate + ethanol exposure caused more expressed functional alterations in both the spontaneous and evoked electrical activity of the brain than the single administration of the same dose of chemicals.

Although in case of P and P+L treatment variations the changes of the investigated spontaneous and evoked functional processes were not significant, the tendencies indicated the vulnerability of the intra- and early extrauterine development of functional processes. It is necessary to emphasize that the measurements of the neurophysiological parameters were made at the end of 12th week of age, and that in P variations the rats received no

mercury from their birth, and in P+L one, from their weaning. That is why, it is a fairly substantial trend that the low-level inorganic lead/dimethoate + ethanol exposure during pre- and postnatal brain development considerably altered certain bioelectical functions. The reason of the more expressed or even significant changes in the P+L+P groups is the additive, postweaning treatment which increased the effect of pre- and postnatal single or combined administration of the compounds.

The investigated substances can pass the placental barrier so they can directly affect the central nervous system (10, 11). The functional changes seen may be explained by the influence of Hg, dimethoate and ethanol on some neurotransmitter processes. The organophosphates alter mostly the cholinergic system, mercury can affect several others (dopaminergic, GABAergic, glutaminergic), and ethanol also has effect on neurotransmitter mechanisms (2, 3, 5, 6, 9). The ECoG changes caused by the single administrations, and also the combinations, could result from alterations of these transmitter systems. At the same time, the lengthening of the latency of evoked potentials can be the consequence of the effects on ion channels slowing down the nerve impulse conduction (8, 9).

Although it is well known that the results of animal experiments cannot be directly transferred to man, our data call the attention to the more expressed, hazardous effect of the combined low-level heavy metal/organophosphate + ethanol exposure which can be a real functional neurotoxicological risk firstly for the more susceptible human subpopulations, such as pregnant and nursing women, their fetuses and suckling babies. That is why, monitoring and prevention of the combined human exposure by neurotoxic compounds seems to be necessary.

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REFERENCES

1. Chang LW, Slikker W. (Eds) *Neurotoxicology. Approaches and Mthods.* Academic Press, 1995.

2. Druse MJ, Tajuddin N, Kuo A, connerty M. Effects of in utero ethanol exposure on the developing dopaminergic system in rats. *J. Neurosci. Res.* 1990; 27: 233-240.

3. Kaneko WM, Riley EP, Ehlers CL. Elecrophysiological and behavioral findings in rats prenatally exposed to alcohol. *Alcohol* 1993; 10: 169-178.

4. Nagymajtényi L., Schulz H, Dési I., Changes of brain evoked potentials caused by dimethoate treatment in three generations of rats. *Neurotoxicology* 1994; 15: 741-744.

5. Narahashi T, Ma JY, Arakawa O., Reuweney E, Nakahiro M., GABA receptor-channel complex as a target site of mercury, copper, zinc, and lanthanides. *Cell Mol. Neurobiol.* 1994; 14: 599-521.

6. Rajanna B, Rananna S, Hall E, Yallapragada PR., In vitro metal inhibition of N-methyl-D-aspartate specific glutamate receptor binding in neonatal and adult rat brain. *Drug Chem. Toxicol.* 1997; 20: 21-29.

7. Schulz H, Nagymajtényi L, Dési I., Behavioral and

neurophysiological changes caused by subchronic mercury exposure in rats. *Cent. Eur. J. Occup. Envir. Med.* 1997; 3: 210-223.

8. Sirois JE, Atchinson WD., Effects of mercurials on ligand- and voltage-gated ion channels: a rewiev. *Neurotoxicology* 1996; 17: 63-84.

9. Weinberg F., Bickmeyer U, Wiegand H., Effects of inorganic mercury (Hg^{2+}) on calcium channels currents and catecholamine release from bovine chromaffin cells. *Arch. Toxicol.* 1995; 69: 191-196.

10. WHO: Dimethoate. Enviromental Health Criteria 90, Geneva, 1989, pp. 1-168.

11. WHO: Inorganic mercury. Enviromental Health Criteria 118, Geneva, 1991, pp. 1-168

INFLUENȚA ADMINISTRĂRII COMBINATE DE XENOBIOTICE ASUPRA FUNCȚIILOR NEUROFIZIOLOGICE ALE CREIERULUI LA ȘOBOLANI

REZUMAT

Întreaga populație, în special cea din țările dezvoltate, este expusă la poluanții mediului înconjurator (metale grele, pesticide) care pot afecta sistemul nervos determinând modificări iremediabile în desfășurarea unor procese senzitive ale activității nervoase centrale. Cum în general oamenii sunt expuși nu numai unui singur poluant, iar printre ei există și consumatori cronici de etanol, cercetarea noastră a avut ca scop studierea modificărilor funcționale ale creierului induse de administrarea combinată a acestor substanțe neurotoxice. Studiul a fost efectuat pe șobolani la care s-au administrat zilnic prin gavaj doze reduse de mercur si dimethoat, la un alt lot administrandu-se în paralel în apa de băut și etanol 5 %. Cercetarea a fost realizată în diferite stadii de dezvoltare intra și/sau extrauterină. Datele obținute au arătat că expunerea la mai multe substanțe nocive a indus alterări mai pronunțate ale parametrilor neurofiziologici urmăriți (electrocorticograma, potențiale corticale evocate) decât cele produse de administrarea unuia singur. Aceste rezultate subliniază importanța analizării în viitor a efectelor combinate induse de xenobiotice din mediul inconjurător nu numai în cadrul experimentelor pe animal, dar și asupra populației.

CUVINTE CHEIE: mercur, dimethoat, etanol, electrocorticograma, potențiale evocate, șobolani.