EFFECT OF PCBs AND 2,4-DMA ON HEPATOCYTE ENZYME ACTIVITY IN RATS

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Introduction

The polyhalogenated aromatic hydrocarbons such as PCBs and phenoxy herbicide 2,4-D widely used in industry and agriculture and manufactured at many chemical plants, particular in Ufa, Russia, can be accumulated in environment and cause toxic influence on functional systems of the organism, resulting to metabolic disorders, pathology of endocrine organs, especially thyroid system [1-3]. Most of metabolic changes are generally the consequence of the liver tissue damage induced by these toxic agents. As we previously determined the changes of thyroid hormonal spectrum in rats in experimental 2,4-D action [4], the aim of the present study was to determine the liver functional status by activity of some enzymes in liver homogenate in conditions of experimental intoxication by PCBs and 2,4-D in possible connection with hormonal disturbances.

Materials and Methods

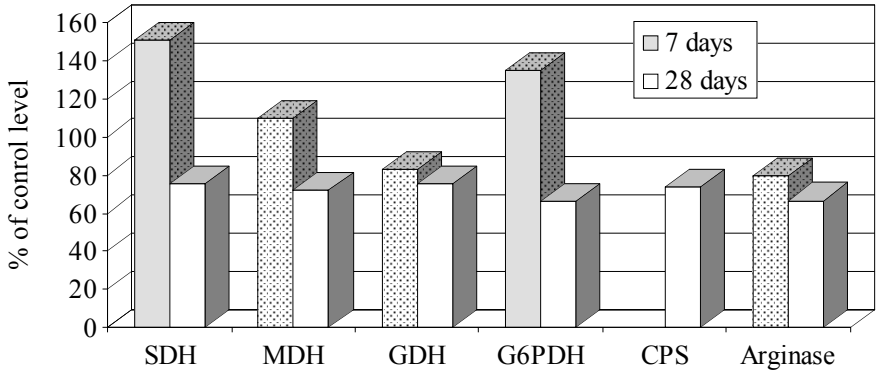
The experiments were performed on 140 male rats of 180-230 grams body weight who received (Groups 1 and 2) the water solution of 2,4-D-dimethylamine salt (2,4-DMA) and (Groups 3 and 4) the corn oil solution of Sovtol-10 (mixture of 70% PCBs and 30% chlorbenzenes) intragastrally during 7 days in total dose equivalent to 1/4 LD50 (Groups 1 and 3) and four weeks in total dose equivalent to LD50 (Groups 2 and 4) for all the period. Control animals were received sodium chloride solution and pure corn oil. Upon the termination of the experiment rats were decapitated and its liver homogenate was tested for the total protein content in cytosol and mitochondrial fraction, and activity of succinate dehydrogenase (SDH), NAD-dependent malate dehydrogenase (MDH), glutamate dehydrogenase (GDH), NADP-dependent glucose-6-phosphate dehydrogenase (G6PDH), carbamylphosphate synthase (CPS) and arginase as possible thyroid-dependent liver enzymes. All statistic data processing was performed using Student t-criterion.

Results and Discussion

The results of 2,4-DA intoxication study presented in Figure 1 show the significant increase of SDH and G6PDH activity (151% and 134,8%) at 7-day experiment, whereas the activity of MDH was only slightly increased and GDH and arginase were contralaterally decreased (down to 83% and 79,5% of control level). In conditions of 28-day toxicant action all the enzymes were inhibited (from 66,1 to 75,6% of control). One of the main reasons of these changes may be the elevation of peroxidation caused by 2,4-DMA. Small amounts of peroxides are known to activate some enzymes, though high level of peroxidation products inhibits it strongly.

We found a significant activation of SDH - one of key enzymes in energy metabolism - in both 7-day intoxication by 2,4-DMA and PCBs (Figures 1,2). Such a "switching" of energy metabolic pathways from NAD-depending substrates to succinate seems to be a universal mechanism of any

Figure 1. Enzyme activity in liver homogenate of rats received 2,4-DMA.



chemical stress, helping the cell to restore the level of ATP in conditions of its elevated consumption on the aims of detoxication (induction of microsomal oxidation enzyme synthesis and conjugation). To approve these suggestions a relative increase of cytosol protein content in hepatocytes was found in our investigations (Table 1). The SDH activation was more expressed at 2,4-DMA intoxication; the same and more significant changes were observed on G6PDH. Meaning that G6PDH activation accompanies much extremal situations, this enables to assume more effective realizing of compensative and adaptational reactions in hepatocytes at early 2,4-DMA intoxication than in case of PCBs. In late intoxication period (28-day) the further elevation of cytosol protein content in hepatocytes was found, up to 20,4% at PCBs and 10,1% at 2,4-DMA. This can testify the weaker inductive abilities of 2,4-DMA by comparison with PCBs at the end of intoxication period, probably by the reason of its more hydrophylity and smaller molecular size.

Table 1. Protein content (mg/g of native tissue) in liver homogenate of rats at 28-day 2,4-DMA and PCBs intoxication (M±m; n=16).

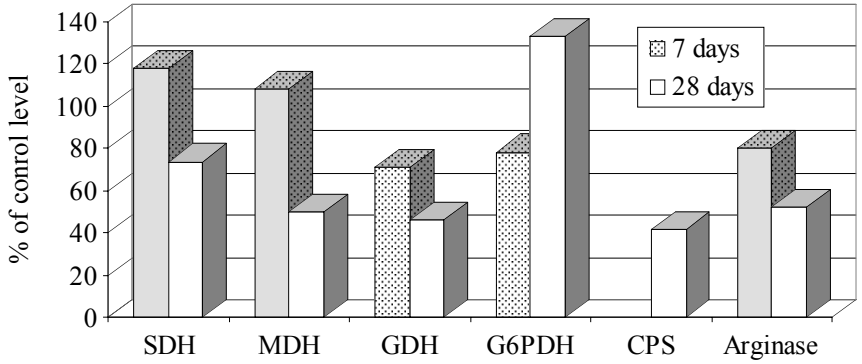
Group of animals	Control	LD50 2,4-DMA	LD50 PCBs
Protein in cytosol fraction	61.60±2.34	70.03±3.19*	74.16±4.83*
% of control level	---	113.6	120.4
Protein in mitochondrial fraction	11.57±0.83	10.08±0.61*	8.63±0.59*
% of control level	---	87.1	74.6

*- $P<0.05$

Found at 28-day PCBs intoxication 46,1% to 49,8% inhibition of NAD-dependent MDH and GDH is possible to consider a sign of general depletion of oxydative reactions in hepatocytes. Under such conditions SDH can become one of the main energy suppliers, as its activity level remained relatively high (73,8% of control). The significant activation of G6PDH (133% of control) reflecting also the G6P utilization in PMP shunt, may both be a result of NADPH/NADP decrease (reducted form was consumed at microsomal oxydation) and at the same time accelerate the generation of NADPH required in high quantities for detoxication, and cause parallel lipogenic reaction in cell resulting in accumulation of triglycerides and cholesterol in liver observed in other investigations [5].

Our results show that GDH and enzymes of urea cycle (CPS and arginase) were inhibited during all periods of intoxication, more significantly at 28-day experiment and in case of PCBs (73.8 to 41.7% of control level). Meaning that these enzymes are mainly located at mitochondria and the

Figure 2. Enzyme activity in liver homogenate of rats received PCBs.



protein content in these organells was also decreased, this can generally testify some delay of amino acid catabolism in hepatocytes in conditions of intoxication. Besides it a probable disorders of specific protein transport into mitochondria may also occur.

The determining role in formation of hepatocyte enzyme activity changes can belong to direct influence of toxicants upon cell metabolic systems alongside with hormonal changes (reduction of blood thyroid hormone level we previously determined), as the most of investigated enzymes and/or its synthesis are thyroid-dependent. This way the correction of liver enzyme activity disorders by administration of thyroid hormones and/or correction of its peripheric conversion has to be principally possible.

References

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