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THE STATE OF PRO/-ANTIOXIDANT ENZYMES AND HISTOLOGICAL CHANGES IN RAT KIDNEYS UNDER SUBLIMATE-INDUCED DAMAGE

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The activity of catalase and glutathione peroxidase in the kidneys of rats was studied 72 hours after administering a 5 mg/kg body weight dose of mercury dichloride solution. This research is important for understanding the effects of mercury salts on the antioxidant system of the kidneys under induced diuresis. In the post-nuclear supernatants of kidney layers, the following were determined: the content of TBA-reactive substances (TBARS); the level of protein oxidative modification products (POMP); and the activities of catalase and glutathione peroxidase. Histological sections were stained with hematoxylin and eosin for the morphological evaluation.
A decrease in glutathione peroxidase activity was found in the cortical, medullary, and papillary regions of the kidneys under water and salt loading after mercury dichloride exposure. This was accompanied by an increase in lipid and protein oxidative modification products and morphological changes in kidney tissues. The intensification of oxidative protein and lipid modification in kidney tissues under mercury dichloride action, in our opinion, is associated with the sharp suppression of antioxidant defense enzymes in the kidneys. Specifically, the glutathione peroxidase activity in the kidneys of animals administered HgCl2 decreased by an average of threefold across all kidney layers.
The results indicate the suppression of antioxidant defense enzymes in rat kidneys due to mercury dichloride exposure.

Keywords: glutathione peroxidase, catalase, protein oxidative modification products, TBA-reactive products, sublimate, kidneys.

Introduction. When mercury salts enter the body, 50 % accumulates in the kidneys. It should be noted that there are two forms of mercury fixation in the kidneys: a labile part of the ion, which determines its excretion level through urine due to the secretory activity of cells, and a less mobile form, which leads to its gradual accumulation (Hazelhoff, Torres, 2021). The toxic effects of mercury ions are linked to lysosome destruction and the release of hydrolytic enzymes, which degrade mitochondrial membranes. Lipid peroxidation (LPO) activation causes significant changes in cellular metabolism and membrane function, playing a crucial role in the pathogenesis of various diseases, including kidney disorders (Temel, Taysi, 2019; Perrone et al, 2023). It is known that under 3 % salt loading during $HgCl_2$ intoxication, free radical oxidation processes are activated in the rat kidneys. Therefore, it is important to investigate the state of the antioxidant system in rat kidneys under mercury dichloride intoxication and to determine changes in antioxidant defense enzymes under water and salt loading conditions (Perrone et al, 2023).

Materials and Methods. The study was conducted on white non-linear sexually mature male rats, weighing 180 ± 10 g, which were housed under standard vivarium conditions. The animals were divided into five groups. Sublimate intoxication was induced using a 0.1 % mercury dichloride solution at a dose of 5 mg/kg body weight. The effect of mercury dichloride on the studied parameters was investigated under conditions of induced water and salt diuresis. The experiments were conducted according to the requirements of the European Convention for the Protection of Vertebrate Animals (86/609/EEC). In the post-nuclear supernatants of kidney layers, the following were measured: 1. TBA-reactive products (TBARS) based on the reaction between malondialdehyde derivatives and thiobarbituric acid (TBA); 2. Protein oxidative modification products (POMP) using the reaction with 2,4-dinitrophenylhydrazine; 3. Catalase activity [EC 1.11.1.6] based on the reaction of unbroken hydrogen peroxide with ammonium molybdate; 4. Glutathione peroxidase activity [EC 1.11.1.9] based on the amount of reduced glutathione. For morphological evaluation, histological sections were stained with hematoxylin and eosin.

Results and Discussion. It was shown that morphological changes occurred in kidney structure as early as 72 hours after treatment (Fig. 1). In intact rats, occasional cells in the cortical substance exhibited signs of clasmatosis, a separation of cytoplasmic fragments into the tubular lumen. These fragments, in sufficiently large quantities, can later form granular or hyaline casts distally; however, this is not a pathological condition. In animals administered mercury dichloride solution, significant morphological changes were observed, particularly in the epithelium of the proximal tubules of the

kidney cortex. Coagulative necrosis was noted in 39.4 ± 3.64 % of the proximal tubules. It should be noted that the number of necrotic epithelial cells could not be counted due to complete nuclear destruction – karyolysis. One hundred percent damage to the epithelial cells of the proximal tubules of the kidneys by the alterative process can be stated. The lumens of most convoluted tubules were filled, fully or partially, with fragments of necrotic and desquamated cells (Fig. 1).

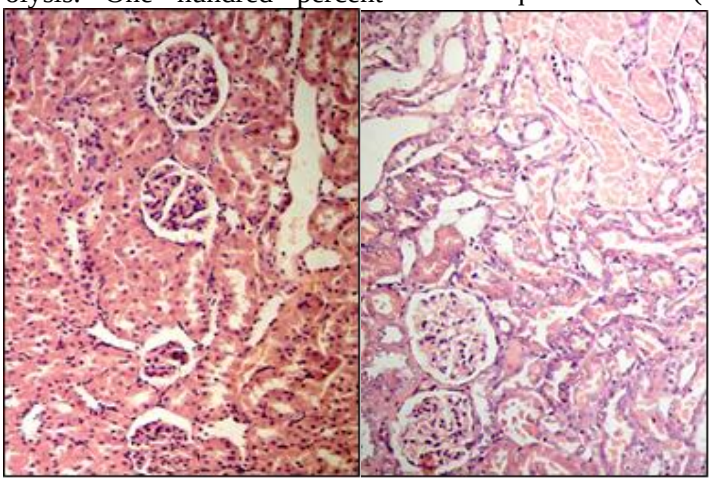

Fig. 1. Histological section of the renal cortex of rats: A – control, B – mercury dichloride. Staining with hematoxylin and eosin. Obj. 10x. Eyepiece 10x.

Catalase activity in kidney tissues (Bevzo, 2017), which detoxifies not only hydrogen peroxide but also macromolecule peroxides, decreased 3.5 times in the cortical and medullary layers under water loading, leading to an increase in both TBA-reactive products and protein oxidative modification products – 2,4-dinitrophenylhydrazones. We found that under water and salt loading, mercury dichloride exposure was accompanied by the activation of free radical oxidation in all parts of the rat kidneys. Thus, the TBARS levels in the cortex and papilla increased by 22 % under sublimate nephropathy with water loading. Salt loading after $HgCl_2$ exposure caused a 23 % increase in TBARS in the cortex and a 30% increase in the papilla of kidneys. Proteins in the kidneys also underwent oxidative modification upon mercury(II) chloride solution administration, as indicated by increased 2,4-dinitrophenylhydrazone levels: under water loading (86 % in the cortex, 72 % in the medulla, and a fivefold increase in the papilla) and salt loading (81 % in the cortex, 90 % in the medulla, and a threefold increase in the papilla) compared to the control groups.

The intensification of oxidative modification of proteins and lipids in kidney tissues under mercury dichloride exposure is linked to a sharp suppression of the antioxidant defense enzymes. Specifically, glutathione peroxidase activity in the kidneys of $HgCl_2$-treated rats decreased threefold across all kidney layers.

The increased oxidative damage to proteins and lipids in the kidneys of animals exposed to mercury dichloride (HgCl2) is likely a consequence of a significant decline in antioxidant enzyme activity within the kidney. Specifically, a substantial three-fold reduction in glutathione peroxidase activity across all kidney layers in these animals is observed.

However, the kidney's antioxidant system appears to exhibit adaptive responses to HgCl2 intoxication. We found that catalase activity was elevated in all kidney layers under both water and salt loading conditions. Under water loading, catalase activity increased by 27% in the cortex and 70% in the papilla, while under salt loading, it increased by 59% in the cortex, 67% in the medulla, and 58% in the papilla

Conclusion. The increase in catalase activity in the kidneys under mercury dichloride exposure may be one of the mechanisms of antioxidant defense.

Thus, subcutaneous administration of a 0.1% mercury dichloride solution at a dose of 5 mg/kg body weight under water and salt loading leads to a suppression of glutathione peroxidase activity, which is accompanied by enhanced oxidative modification of proteins and lipids, as well as morphological changes in kidney tissue. The pro-/antioxidant balance under toxicant exposure is maintained by an increase in catalase activity.

Interests disclosure*. The authors declare no conflict of interest.*

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СТАН АНТИОКСИДАНТНИХ ЕНЗИМІВ НИРОК ЩУРІВ ПРИ УРАЖЕННІ СУЛЕМОЮ

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Вивчали активність каталази, глутатіонпероксидази у нирках щурів через 72 години після введення розчину меркурію дихлориду в дозі 5 мг на 1 кг маси тварин, що є важливим для з'ясування впливу солей меркурію на антиоксидантну систему нирок за умов індукованого діурезу. У постядерних супернатантах шарів нирок визначали: вміст ТБК-реакційних продуктів (ТБК-РП); вміст продуктів окисної модифікації протеїнів (ПОМП); активність каталази та глутатіонпероксидази. Для морфологічної оцінки гістологічних зрізів зразки забарвлювали гематоксиліном і еозином.
Встановлено зниження активності глутатіонпероксидази у кірковій, мозковій речовині та сосочку нирок за умов водного та сольового навантаження після дії меркурію дихлориду, що супроводжувалося підвищенням вмісту продуктів окислювальної модифікації ліпідів і білків та морфологічними змінами у тканині нирок. Посилення окиснювальної модифікації протеїнів і ліпідів у тканинах нирок за умов дії меркурію дихлориду пов'язано, на нашу думку, з різким пригніченням ензимів системи антиоксидантного захисту в нирках. Так, активність глутатіонпероксидази нирок тварин, яким вводили HgCl2, знизилась у всіх шарах нирок у середньому в 3 рази.
Отримані результати свідчать про пригнічення ферментів антиоксидантного захисту у нирках щурів за дії меркурію дихлориду.

Ключові слова: глутатіонпероксидаза, каталаза, продукти окисної модифікації білків, ТБК-реакційні продукти, сулема, нирки

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