Experimental Study of Hepatoprotective Activity of Hydroxymethyluracil

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Hepatoprotective activity of pyrimidine derivative hydroxymethyluracil was studied on the model of acute paracetamol-induced liver damage in rats. Hydroxymethyluracil was shown to produce a strong hepatoprotective effect.

Key Words: *toxic hepatitis; hepatoprotective properties; syringomyelia; hydroxymethyluracil; Carsil*

Hepatobiliary dysfunction is a prevalent pathology [6]. Liver damage often results from intoxication with drugs, including paracetamol [12], valproate, or carbamazepine (complication of anticonvulsant therapy) [8-10]. Liver diseases are accompanied by such neurological disorders as Wilson disease [11] and syringomyelia.

The search for new hepatoprotective drugs is an urgent problem.

Much recent attention was paid to pyrimidine derivatives as potential hepatoprotective drugs. They include 2,4-dioxo-5-hydroxy-6-methyl-1,2,3,4-tetrahydropyrimidine (hydroxymethyluracil, HMU) [3].

Pyrimidine bases are constituents of nucleic acids. They produce various effects on the organism [1]. A wide range of biological activity is also typical of derivatives of various natural nucleic acids, including HMU. Published data show that HMU has antioxidant, antitoxic, regenerative, and antiinflammatory properties [4,5]. HMU (Immureg) is recommended as an immunomodulatory drug.

Here we studied hepatoprotective activity of HMU on the model of paracetamol-induced liver damage.

MATERIALS AND METHODS

Experiments were performed on 46 male outbred rats weighing 200-250 g and maintained in a vivarium under standard conditions. The animals were divided into 4 groups. Group 1 included 10 intact animals (control). Other groups consisted of 12 animals with paracetamol-induced liver damage (acetaminophen). The suspension of powdered tablets of paracetamol (Nizhfarm) in 2% starch gel was administered *per os* in a dose of 2500 mg/kg for 2 days.

Group 2 animals did not receive therapy. Group 3 rats *per os* received HMU suspension in 2% starch gel (daily dose 50 mg/kg). Group 4 rats received a similar dose of Carsil suspension in 2% starch gel. Paracetamol was administered to animals of groups 3 and 4 on days 6 and 7 after the start of therapy with hepatoprotective drugs. Hence, treatment with paracetamol and hepatoprotective drugs was discontinued on the same day. The rats were euthanized 1 day after the last treatment.

The animals were tested for biochemical parameters of the blood, including activities of alanine transaminase (ALT) and aspartate transaminase (AST), concentrations of bilirubin and total protein, and activity of alkaline phosphatase (AP). Morphological characteristics of the liver were evaluated. Lipid peroxidation was studied by chemiluminescence methods. Malonic dialdehyde (MDA) content was measured in liver homogenates. ALT and AST activities were estimated using Lachema kits [2]. Total bilirubin concentration was determined spectrophotometrically by color reaction of bound pigment with sulfanilic acid. Total protein content was measured as described elsewhere [3]. AP activity was measured spectrophotometrically using standard

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TABLE 1. Effect of HMU on Biochemical Parameters of Blood Plasma from Rats with Paracetamol Intoxication (*M*±*m*)

Parameter	Group			
	1	2	3	4
ALT, μcat/liter/h	0.67±0.03	1.95±0.09*	1.10±0.08*+	1.16±0.11*+
AST, μcat/liter/h	0.92±0.02	1.71±0.05*	1.18±0.03*+	1.23±0.05*+
Bilirubin, μmol/liter	10.00±0.21	13.26±0.94*	10.58±0.23+	10.79±0.44+
Total protein, g/liter	72.00±1.71	66.00±3.81	77.20±2.42+	75.20±2.07+
AP, units	427.73±23.85	566.25±35.39*	430.31±23.20+	473.15±27.47+

Note. Here and in Table 2: $p<0.05$: *compared to group 1; +compared to group 2.

TABLE 2. Effect of HMU on Free Radical Oxidation in Liver Homogenates from Rats with Paracetamol Intoxication (*M*±*m*)

Parameter	Group			
	1	2	3	4
Total chemiluminescence, arb. units	5.25±0.25	14.22±0.50*	6.03±0.45+	6.60±0.41*+
MDA, nmol/sample	0.48±0.04	0.83±0.07*	0.55±0.06+	0.58±0.04+

Lachema kit. 4-Nitrophenylphosphate served as the substrate. Chemiluminescence was estimated on a KhL-003 device [7].

RESULTS

After 2 days, ALT activity in group 2 rats increased by 2.9 times compared to group 1 animals (Table 1). ALT activity in group 3 rats increased by 1.63 times compared to group 1 animals, but was lower than in group 2 (Table 1). In group 4, plasma ALT activity increased by 1.72 times and significantly exceeded that in group 1. However, ALT activity in group 4 rats was much lower than in group 2 animals. No statistically significant differences in enzyme activity were revealed in rats of groups 3 and 4.

Administration of paracetamol was followed by a 1.86-fold increase in plasma AST activity (compared to group 1 rats, Table 1). AST activity in group 3 rats significantly increased compared to group 1 animals (by 1.28 times), but was much lower than in group 2 specimens.

AST activity in group 4 rats increased by 1.33 times compared to group 1 animals, but was much lower than in group 2 specimens. No statistically significant differences in enzyme activity were revealed in rats of groups 3 and 4.

Our results indicate that HMU protects hepatocyte membranes from destruction and prevents the release of aminotransferases from these cells. Hence, HMU possesses hepatoprotective activity.

Analysis of chemiluminescence intensity and MDA content showed that HMU *in vivo* has high antioxidant activity. This drug probably protects hepatocyte membranes from peroxidation under conditions of paracetamol intoxication (Table 2).

We conclude that HMU produces a strong hepatoprotective effect on rats with acute paracetamol intoxication. In this respect HMU is highly competitive with extensively used hepatoprotective drug Carsil. The data indicate that hepatoprotective properties of HMU should be evaluated in clinical trials with chronic and acute liver damages.

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