Study of Hepatoprotective Effects of Xymedon

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Xymedon (1-(β-oxyethyl)-4,6-dimethyl-1,2-dihydro-2-oxopyrimidine), a regeneratory and wound-healing drug, exhibited hepatoprotective activity in laboratory animals with experimental toxic hepatitis. Oral drug reduced the severity of toxic involvement of the liver induced by CCl_4 and reduced animal mortality. Xymedon promoted recovery of the blood biochemical parameters characterizing the liver status.

Key Words: *xymedon; hepatoprotectors; toxic hepatitis; hepatic diseases*

Numerous adverse factors (environment pollution, infection, irrational nutrition, *etc.*) promote the increase in the incidence of liver diseases and dysfunctions [5,8]. This makes search for highly effective drugs with hepatoprotective activity an important problem for medicine and preventive (nootropic) pharmacology, as well as for labor and sports ecology.

Despite numerous available hepatoprotectors, there are no Russian drugs meeting all the requirements for drugs of this pharmacological group [2,3,7-12]. Active search for hepatoprotectors is in progress in Russia and in foreign countries [2,7,9-12].

Xymedon (1-(β-oxyethyl)-4,6-dimethyl-1,2-dihydro-2-oxopyrimidine) is an original Russian drug with a wide spectrum of pharmacological activities: wound-healing, regeneratory, anti-inflammatory, immunostimulatory, radioprotective, and antioxidant [1,4]. Importantly that the toxicity of the drug is low: LD_{50} above 7000 mg/kg.

We studied the hepatoprotective effects of xymedon in order to obtain more ample data on its biological activities and to contribute to search for safe Russian hepatoprotectors, needed in labor and sports ecology.

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MATERIALS AND METHODS

The study was carried out on outbred albino rats (250-400 g; *n*=30) and mice (12-27 g; *n*=45) using the method described in Manual for Experimental (Preclinical) Studies of New Drugs [6] in accordance with the regulations of the European Convention for Protection of Vertebrates Used for Experimental and Other Research Purposes.

Toxic hepatitis was induced by subcutaneous or oral administration of hepatotoxin (Hetox) (CCl_4 solution in olive oil, 1:1 volume proportion). Xymedon (studied drug) and hetox were administered according to the following protocols: 1) rats and mice: 4 days of subcutaneous hetox (2 ml/kg) 1 h after oral xymedon (5, 10, or 50 mg/kg); 2) rats: oral hetox in a single dose of 4 ml/kg and after 24 h oral xymedon in daily doses of 10 and 50 mg/kg for 3 days; 3) mice: oral xymedon (50 mg/kg) for 4 days and a single oral dose of hetox (4 or 5 ml/kg) 1 h after the last xymedon dose. Experiments with controls were carried out for each protocol. Experimental animals received the hepatotoxin and xymedon according to the above protocols, controls received hepatotoxin alone.

Groups of 3-6 animals were formed for experiments on rats, of 5-7 animals for experiments on mice. Controls received hetox according to similar protocols and oral water instead of xymedon. In addition, blood

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TABLE 1. Body Weight Increment and Mortality in Mice Treated by Xymedon and Hetox (*M*±*m*)

Experiment conditions	Body weight increment, g/day	Mortality, %
Protocol 1		
Control (H_2O+hetox 2 ml/kg)	-0.29±0.04	28.6 (2 из 7)
Xymedon 50 mg/kg+hetox 2 ml/kg	1.08±0.10***	28.6 (2 из 7)
Protocol 3		
Control (H_2O+hetox 4 ml/kg)	-0.35±0.07	0
Xymedon 50 mg/kg+hetox 4 ml/kg	-0.25±0.04	0
Control (H_2O+hetox 5 ml/kg)	-0.60±0.19	80 (4 из 5)
Xymedon 50 mg/kg+hetox 5 ml/kg	-0.70±0.24	40 (2 из 5)

Note. ***p<0.001 in comparison with control.

biochemical values were studied in intact rats (intact control; n=6).

The animals were weighed before and after the experimental procedures. After experiments, the animals were decapitated under ether narcosis and the blood was collected for macro- and micromorphologic analysis. Biochemical values were measured on a DaytonaRandox biochemical analyzer using Randox kit. The status of the liver was evaluated visually, and micromorphology was studied in histological sections (fixation in 10% formalin, paraffin sections of 7-10 μ, hematoxylin and eosin staining). Microphotographs were made on a device consisting of an AxioImager M2 microscope, AxioCamHRc digital camera (Carl Zeiss), and PC.

The values were processed by Origin 6.1 software. The differences between the samples were compared by Student's t test.

RESULTS

In the controls, the highest mortality (80%) was recorded after hetox in a dose of 5 ml/kg according to protocol 3. In experimental group that received xymedon in a dose of 50 mg/kg according to protocol 3, the mortality was 2-fold lower (Table 1). Control animals did not die after hetox in a dose of 4 ml/kg according to the same protocol, but body weight loss was less marked in experimental animals receiving xymedon.

The mortality of experimental and control animals receiving protocol 1 treatment was virtually the same. However, the time course of body weight changes differed significantly (p<0.001). Body weight loss was recorded in controls under the effect of hetox, indicating deterioration of animal status. Experimental animals gained weight (Table 1).

Biochemical analysis showed a reduction of blood glucose level and an increase of transaminase (ALT and AST) activities (Table 2). These changes in the blood biochemistry in comparison with intact animals indicated liver involvement – disorders in the synthetic function and destruction of hepatocytes [3,5,8,12].

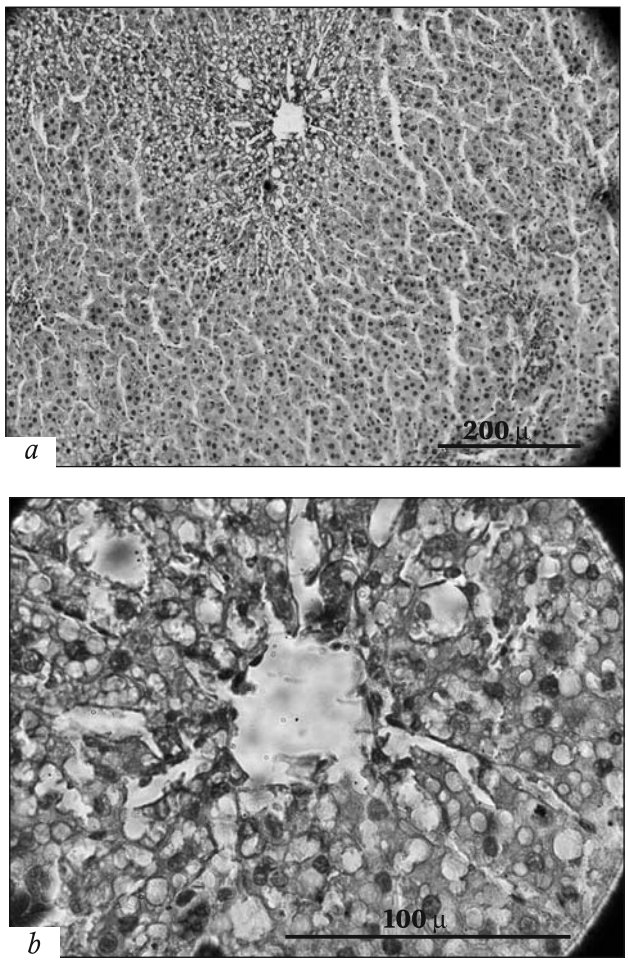

Fig. 1. Rat hepatic lobe after water and hetox for 4 days according to protocol 1; ×100 (*a*), ×1000 (*b*).

TABLE 2. Blood Biochemistry in Laboratory Rats (*M*±*m*)

Parameter	Protocol 1					Protocol 2		
	Intact animals	Control	Xymedon			Xymedon		
			5 mg/kg	10 mg/kg	50 mg/kg	10 mg/kg	50 mg/kg	
Glucose, mmol/liter	7.99±1.19	3.59±1.00*	6.14±0.78	5.96±0.35	7.30±1.37	7.24±1.05	–	
ALT, U/liter	42.22±5.57	220.14±97.05**	169.56±23.30***	142.15±33.80**	119.39±42.88	99.22±17.56**	42.77±14.01	
AST, U/liter	141.66±11.71	277.81±14.30***	281.43±12.12***	194.54±26.98	207.01±61.09	209.86±14.63*	149.57±8.48	

Note. Control values for protocol 2 are not presented, as the animals died. *$p<0.05$, **$p<0.01$, ***$p<0.001$ in comparison with intact animals.

Fig. 2. Rat hepatic lobe after xymedon in a dose of 50 mg/kg and hetox during 4 days according to protocol 1; ×100 (*a*), ×1000 (*b*).

The parameters less differed from the normal values in experimental rats treated by xymedon. The differences were the least in the groups which received xymedon in a dose of 50 mg/kg according to protocols 1 and 2 (Table 2).

Histological studies of the liver showed disorders in the organ morphology after hetox in all control animals: extensive regions of destroyed and morphologically changed hepatocytes, mainly adjacent to the central vein (Fig. 1). Bile ducts were stenosed, slit spaces between the cells were virtually absent. The morphological disorders in the organ were less severe in animals which received xymedon; the structural organization of hepatocytes was retained, and foci of cell destruction were significantly smaller (Fig. 2).

The data of experiments carried out according to protocols 1 and 3 indicated that xymedon reduced the severity of hepatic dysfunction after CCl_4 intoxication. The results of experiment by protocol 2 suggested that xymedon treatment during several days after liver intoxication led to recovery of its function.

Hence, one more biological activity of xymedon — hepatoprotective, heretofore unknown, has been detected.

REFERENCES

1. S. G. Izmailov, G. A. Izmailov, M. Yu. Averyanov, and V. S. Reznik, *Xymedon in Clinical Practice* [in Russian], Nizhn. Novgorod (2001).
2. N. F. Kushnerova, V. G. Sprygin, and Yu. A. Rakhmanin, *Biomed. Khim.*, **50**, No. 6, 605-611 (2004).
3. V. E. Novikova and E. I. Klimkina, *Obzory Klin. Farmakol. Lekarstv. Ter.*, **4**, No. 1, 2-20 (2005).
4. V. I. Pogoreltsev, V. Yu. Tereshchenko, A. A. Chirkin, *et al.*, *Kazansk. Med. Zh.*, **86**, No. 4, 346-347 (2005).
5. M. G. Romantsov, D. S. Sukhanov, A. Yu. Petrov, *et al.*, *Fundament. Issled.*, No. 3, 131-141 (2011).
6. *Manual for Experimental (Preclinical) Studies of New Drugs* [in Russian], Moscow (2000).
7. A. S. Saratikov, V. S. Chuchalin, A. E. Rat'in, *et al.*, *Eksp. Klin. Farmakol.*, **68**, No. 2, 51-54 (2005).
8. A. N. Tauki, V. N. Fyodorov, Z. A. Kunitsa, *et al.*, *Ros. Med. Biol. Vestn. Im. Akad. I. P. Pavlova*, No. 1, 66-73 (2010).
9. I. V. Shilova, T. V. Zhavoronok, N. I. Suslov, *et al.*, *Bull. Exp. Biol. Med.*, **146**, No. 1, 49-51 (2008).
10. K. Demirdag, I. H. Bahcecioglu, I. H. Ozercan, *et al.*, *J. Gastroenterol. Hepatol.*, **19**, No. 3, 333-338 (2004).
11. A. S. Kabankin and L. I. Gabrielyan, *Pharm. Chem. J.*, **39**, No. 3, 135-139 (2005).
12. A. I. Vengerovskii, N. O. Baturina, V. S. Chuchalin, and A. S. Saratikov, *Pharm. Chem. J.*, **30**, No. 2, 69-71 (1996).