EFFECT OF ZIXORYN ON THE PHARMACOKINETICS OF ANTIPYRINE IN INTACT AND SENSITIZED GUINEA PIGS

T. G. Khlopushina, A. V. Krinskaya, L. P. Kovalenko, V. P. Zherdev, and A. V. Val'dman*

UDC 616-056.3-02:613.2]-092.9-07:
616-008.949.4:615.212.3]-033.1

KEY WORDS: food-related anaphylaxis; zixoryn; pharmacokinetics; antipyrine and its metabolites

The pharmacokinetics of antipyrine indirectly reflects changes in monooxygenase activity and in the state of the functioning mass of hepatocytes. In the investigation described below we studied the character of biotransformation and the pharmacokinetics of the model preparation antipyrine on an experimental model of alimentary anaphylaxis, which is a delayed-type hypersensitivity reaction, in the presence and absence of zixoryn, an inducer of hepatic cytochrome P-450 [12].

EXPERIMENTAL METHOD

Experiments were carried out on noninbred male guinea pigs weighing 250-300 g, kept on the standard animal house diet.

An experimental model of alimentary anaphylaxis to hen's egg albumin (HEA) in guinea pigs was produced by the method in [7], which we modified. Guinea pigs were sensitized by peroral administration of a 0.3% solution of HEA in a dose of 2 ml/250 g body weight daily for 3 days. Starting 12 days after the beginning of sensitization, the animals were given intraperitoneal injections of zixoryn, in a dose of 50 mg/kg, in 0.5% starch suspension, daily for 3 days; the control sensitized animals received the corresponding volume of starch suspension. HEA was injected intravenously into the animals 24 h after the last injection of zixoryn in a dose of 1 mg/300 g body weight, after which the development of the anaphylactic reaction was recorded. Surviving animals from this group were used in the pharmacokinetic investigations.

Antipyrine was injected intraperitoneally into the animals in a dose of 100 mg/kg. Blood samples were taken at the following times: 0, 1, 2, 3, 4, and 5 h; urine was collected during 5 h. Quantitative analysis of the content of antipyrine and its metabolites (4-OH-antipyrine and norantipyrine) in the biological material was carried out by gas chromatography [10, 11]. Antipyrine and its metabolites were analyzed on a Soviet model 3700 gas chromatograph with ionization-flame detector and "Interkhrom 1" integrator (Bulgaria). The conditions of chromatography were: glass column with internal diameter 2 mm and length 2 m, mobile phase 3% OV=17, filler "Chromosorb G" (100-200 mesh), column thermostat temperature 240°C, carrier gas nitrogen, flow rate 30 ml/min, hydrogen and air flow rate 30 and 300 ml/min respectively. The test substances were dissolved in methanol and introduced into the chromatograph in a volume of 1 μl. Quantit;ative determination of antipyrine and its metabolites was carried out by the absolute calibration method (based on areas of the peaks).

The pharmacokinetic parameters of plasma concentration versus time were calculated by regression analysis on a computer of IBM AT type; the constant of the excretion and clearance rates of the metabolites in the urine were calculated as described in [6]. The results were subjected to statistical analysis by Student's test [3].

Research Institute of Pharmacology, Academy of Medical Sciences of the USSR, Moscow. Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 112, No. 7, pp. 67-69, July, 1991. Original article submitted October 22, 1990.

 0007-4888/91/0007-0990$12.50

TABLE 1. Effect of Zixoryn on Pharmacokinetics of Antipyrine in Intact and Sensitized Guinea Pigs (M ± m; n = 6)

Experimental conditions	K_a, h^{-1}	AUC, $h \cdot \mu g \cdot ml^{-1}$	$T_{1/2}$, h	Cl_t, $ml \cdot h^{-1} \cdot kg^{-1}$	Vd, $ml \cdot kg^{-1}$
Intact control	0,50±0,01	190,6±8,1	1,39±0,03	525,7±22,5	1055,5±165,8
Sensitization	0,35±0,03***	260,4±23,0***	1,97±0,18**	387,2±35,2***	1096,9±128,9
Sensitization + zixoryn	0,74±0,14***	127,9±49,7***	0,97±0,17***	770,8±278,9*	948,4±311,2

Legend. Asterisk indicates data for which $p < 0.05$.

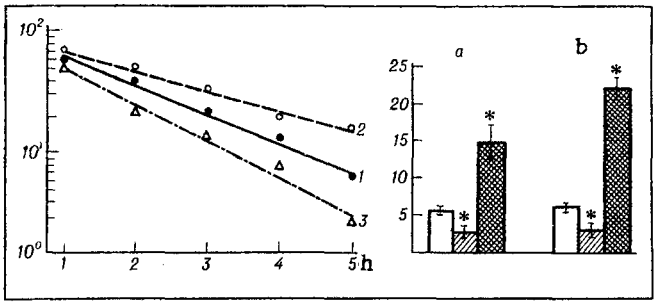

Fig. 1

Fig. 2

Fig. 1. Kinetic curves of unchanged antipyrine in plasma of guinea pigs: intact (1), sensitized (2), and sensitized and receiving zixoryn (3). Abscissa, time (in h); ordinate, concentration of antipyrine (in $\mu g \cdot ml^{-1}$).

Fig. 2. Effect of zixoryn on renal clearance of antipyrine metabolites in intact and sensitized guinea pigs. a) Norantipyrine, b) 4-OH-antipyrine. Unshaded columns — intact control; obliquely shaded — sensitization, cross-hatched — sensitization + zixoryn. Ordinate, Cl_r (in $ml \cdot g^{-1} \cdot kg^{-1}$). *$p < 0.05$.

EXPERIMENTAL RESULTS

The writers previously found correlation between the intensity of manifestations of alimentary anaphylaxis in guinea pigs and the state of the liver cytochrome P-450 system [1]. Alimentary sensitization of animals to HEA was found to be accompanied by a decrease in the content of cytochrome P-450 in the microsomal fraction of the liver by 20-25%, and by weakening of monooxygenase activity: the rate of N-demethylation of amidopyrine was reduced by 16-23%, and the rate of hydroxylation of aniline by 20-30%. It was also found that injection of inducers of cytochrome P-450 of different chemical nature into sensitized guinea pigs not only restored the monooxygenase activity of the liver, when depressed by sensitization, but also considerably potentiated it, and this was accompanied by a decrease in the intensity of the anaphylactic reaction [1, 4, 5].

The results of the study of the functional state of the cytochrome P-450-dependent monooxygenase system of the liver by the antipyrine test in intact and sensitized guinea pigs receiving and not receiving zixoryn are given in Table 1. The pharmacokinetic tests showed that antipyrine, if injected intraperitoneally in a dose of 100 mg/kg, was detectable in the blood plasma for 5 h (Fig. 1). The half-elimination time ($T_{1/2}$) of antipyrine from the plasma in sensitized animals was 1.4 times higher than $T_{1/2}$ in intact animals, whereas injection of zixoryn into the sensitized guinea pigs lowered the value of this parameter by half compared with the "sensitization" group. The area under the pharmcokinetic curve (AUC) also changed in a similar manner. Total clearance (Cl_t) was reduced by 1.3 times in the sensitized animals, but administration of zixoryn doubled the value of this parameter. The distribution volume (V_d) in animals of all groups studied did not differ statistically significantly.

The results of the investigations of 4-hydroxylase and N-demethylase activity of the liver monooxygenases in intact and sensitized guinea pigs are given in Fig. 2. They show that clearance of metabolites (Cl_r), namely 4-OH-antipyrine, in the sensitized animals was reduced by half compared with intact animals. Injection of zixoryn into sensitized animals increased the norantipyrine clearance by 6.7 times and the 4-OH-antipyrine clearance by 7.7 times.

Thus the results of investigation of the pharmacokinetics and excretion of unchanged antipyrine and of its metabolites also show that sensitization of guinea pigs by HEA reduces the functional activity of the liver monooxygenases, whereas the use of zixoryn as inducer of liver cytochrome P-450 leads to a sharp increase in the 4-hydroxylase and N-demethylase activity of the monooxygenases in the microsomal fraction of the liver. Our results are in good agreement with data [9] also showing that zixoryn significantly reduced $T_{1/2}$ for antipyrine and increased renal excretion of D-glucuronic acid, i.e., that zixoryn is a powerful inducer of the reactions of phases I and II of biotransformation.

The experiments showed that differences in basal liver monooxygenase activity in guinea pigs are reflected in the sensitivity of the animals to anaphylactic shock: increased activity of the cytochrome P-450 system of the liver increases the resistance of the animals to anaphylaxis [2]. There is evidence that a "weak" oxidation phenotype (poor metabolizer status) is connected with food allergy: most patients in the food allergy group are "poor metabolizers" [8]. It has been suggested that the phenotype of the enzymes in these patients makes them more or less sensitive to the chemical components of the diet.

The possibility cannot be ruled out that many persons suffering from food allergies are people with undiagnosed disturbances of cytochrome P-450-dependent metabolic processes. The study of the character of the changes in metabolism of such patients may allow the level of the metabolizing ability of the liver in such patients to be indirectly determined, and the use of cytochrome P-450 inducers may be a new approach to the treatment of allergic diseases.

LITERATURE CITED

1. E. Kovalev, T. O. Khlopushina, I. N. Marokko, and E. M. Lysenkova, Patol. Fiziol., No. 4, 22 (1983).
2. I. N. Marokko, T. O. Khlopushina, E. M. Lysenkova, et al., Byull. Éksp. Biol. Med., No. 12, 713 (1984).
3. N. A. Plokhinskii, Algorithms of Biometrics [in Russian], Moscow (1980).
4. T. G. Khlopushina, A. V. Krinskaya, I. N. Marokko, and I. E. Kovalev, Farmakol. Toksikol., No. 4, 86 (1988).
5. T. G. Khlopushina, I. N. Marokko, and I. E. Kovalev, Khim.-Farm. Zh., No. 11, 1298 (1987).
6. L. E. Kholodov and V. P. Yakovlev, Clinical Pharmacokinetics [in Russian], Moscow (1985), pp. 69-72.
7. V. A. Shaternikov, I. N. Marokko, N. N. Pyatnitskii, et al., Vopr. Pitaniya, No. 2, 27 (1982).
8. J. Brostoff and S. J. Challacombe, Food Allergy and Intolerance, London (1987), pp. 443-447.
9. B. Gachalyi, Cytochrome P-450: Biochemistry, Biophysics and Induction, Budapest (1985), pp. 269-271.
10. D. H. Huffman, D. W. Shoeman, and D. L. Azarnoff, Biochem. Pharmacol., **23**, No. 2, 197 (1974).
11. T. Inaba and N. E. Fischer, Can. J. Physiol. Pharmacol., **58**, No. 1, 17 (1980).
12. S. Szeberenyi, Cytochrome P-450: Biochemistry, Biophysics and Induction, Budapest (1985), pp. 253-259.