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PHARMACOLOGY

Antihypoxic Effects of Some 3-Hydroxypyridine Derivatives in Isolated Rat Myocardium

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The electron-transporting function of the respiratory chain is known to be disturbed at its substrate-binding site in the early stages of hypoxia and ischemia [2,3,8]. Its first enzymatic complex is the limiting stage of this process. It is thus thought to be possible to activate compensatory metabolic pathways (alternative to the main NAD-dependent mitochondrial oxidation) acting as mechanisms of emergency adaptation to these pathological processes during hypoxia. A succinate oxidase pathway with oxidative thermodynamic advantages in the mitochondrial respiratory chain at low pO_2 values is the first of these [1]. Heterocyclic compounds are thought to facilitate succinate entry into the cell and its subsequent oxidation in the respiratory chain, this determining the antihypoxic properties of these compounds. The aim of the present study was to investigate the possibility of using mexidol (a succinate-containing 3-hydroxypyridine derivative) as an antihypoxant with a direct energizing effect [4].

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MATERIALS AND METHODS

Experiments were carried out on male albino rats weighing 180-200 g which were divided in a pressure chamber into rats with high resistance (HR) and with low resistance (LR) to hypoxia. The antihypoxic properties of the hydroxypyridine derivatives emoxypine and mexidol were studied on an isolated per-

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fused contracting rat heart (according to Langendorff's method modified by us [6]). The heart rate (HR) and heartbeat force (HBF) and their product (HR×HBF), myocardial respiration rate (V_r), and perfusion flow rate through the myocardium (V_p) were registered. The myocardium was perfused with carbogen-saturated Krebs-Henseleit solution (37°C, pH 7.4) (normoxia). Moderate hypoxia was caused by replacing this solution with perfusion fluid saturated with a gas mixture containing 50% O_2 + 45% N_2 + 5% CO_2. After 20 min the solution was replaced again with the "normoxic" one. Every 20 min the lactate and pyruvate concentrations were measured in the perfusion fluid. At the end of the reoxygenation period the myocardial ATP and CP content was measured. The test substances were administered in concentrations of 10^{-6}, 10^{-5}, and 10^{-4} M 10 min before hypoxia simulation. The results were statistically processed after Student.

RESULTS

The effects of the substances investigated under experimental hypoxia depended on their concentration and varied in the myocardium of HR and LR rats. Emoxypine inhibited myocardial contractility in hypoxia, the degree of inhibition being the most marked at a concentration of 10^{-6} M and minimal at a concentration of 10^{-5} M (Fig. 1). Its major influence was directed at HBF and V_r, i.e., at aerobic processes.

On the other hand, emoxypine in a concentration of 10^{-6} M reduced hypoxic disturbances of HR, HBF and V_r in the myocardium of LR rats, that is, it exhibited an antihypoxic effect (Fig. 1). This effect diminished when the concentration of the substance was lowered to 10^{-5} M, and it became prohypoxic at a concentration of 10^{-4} M. The heart was liable to stop in such a case, whereas without emoxypine or in HR rat myocardium this was never seen.

The effects of emoxypine correlated with the ATP and CP changes in the myocardium of HR and LR rats; the macroergic content increased in the case of a positive emoxypine influence on the hypoxic myocardium function, as compared to the influence of hypoxia alone. It decreased sharply under the prohypoxic influence of emoxypine (Table 1).

A prohypoxic effect of the succinate-containing 3-hydroxypyridine derivative mexidol on myocardial contractility was observed in HR rats under hypoxic conditions only at a low concentration (10^{-6} M) of the agent. Its negative influence on the cardiac functional parameters studied disap-

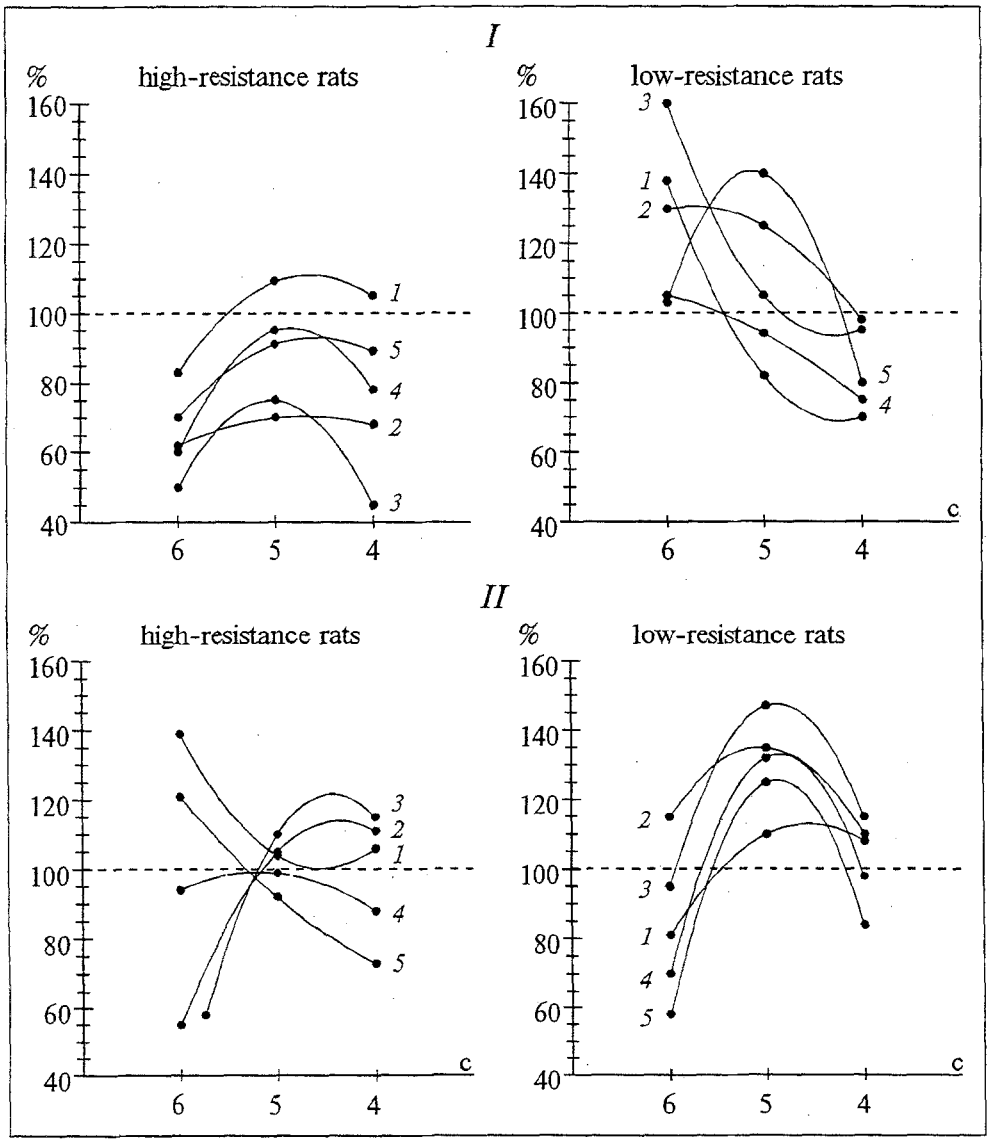

Fig. 1. Effectiveness of emoxypine (I) and mexidol (II) in different concentrations on the functional parameters of the myocardium at the 20th minute of reoxygenation. Abscissa: emoxypine and mexidol concentrations. Ordinate: changes of hypoxic effects in the presence of the agents (% of the values in hypoxia alone). 1) HR, 2) HBF, 3) HR×HBF, 4) V_r, 5) V_r

TABLE 1. Effect of Hydroxypyridine Derivatives on ATP and CP Content in the Isolated Rat Myocardium at the 20th min of Reoxygenation

Animal type	Concentration of substances, M	ATP				CP			
		emoxypine		mexidol		emoxypine		mexidol	
		μmole/g dry weight	%	μmole/g dry weight	%	μmole/g dry weight	%	μmole/g dry weight	%
HR	—	11.35±0.65	100	11.35±0.65	100	16.64±0.50	100	16.06±0.37	100
	10^{-6}	8.35±0.62	74*	11.24±0.8	99	16.34±0.57	98	19.31±0.81	120*
	10^{-5}	10.45±0.36	92	11.55±0.9	102	15.86±0.63	95	16.06±0.42	100
	10^{-4}	8.57±0.20	76*	11.75±0.85	104	13.18±0.54	79*	16.64±0.54	104
LR	—	10.90±0.21	100	10.55±0.52	100	16.46±0.76	100	15.56±0.98	100
	10^{-6}	12.41±0.68	114*	11.71±0.63	111	19.67±1.3	120*	18.46±1.2	118*
	10^{-5}	6.63±0.43	61**	11.11±0.71	105	11.52±2.24	70*	18.56±0.10	119*
	10^{-4}	2.56±0.30	23***	10.86±0.58	103	4.46±0.32	27***	15.55±0.95	100

Note: asterisk: $p<0.1$; two asterisks: $p<0.05$; three asterisks: $p<0.01$.

peared or even became antihypoxic when its concentration was increased (Fig. 1). A reduction of hypoxic disturbances of HBF together with an absence of any pronounced influence on V_r was particularly characteristic of its effect. During the posthypoxic period, only a prohypoxic effect of mexidol alone took place in the myocardium of HR rats.

In contrast to this, mexidol in a concentration of 10^{-5} M produced a marked antihypoxic effect, manifested in a decrease of HBF, V_r, and HR disturbances during hypoxia (Fig. 1). The drug accelerated recovery of these parameters in comparison with that in hypoxia alone. Mexidol in the tested concentration intensified both aerobic and anaerobic processes in the myocardium of LR rats. Moreover, it promoted an increase of the pyruvate content in the perfusion fluid, which probably reflected oxidation restriction. An increase or decrease of the mexidol concentration was associated with a weakening (at a concentration of 10^{-4} M) of its antihypoxic effect, or the latter became a prohypoxic one (Fig. 1).

Mexidol did not influence the ATP content in the myocardium of HR rats. Still, the ATP level was higher in the presence of mexidol than in the presence of emoxypine, correlating with the absence of a pronounced negative influence of mexidol on the contractile function of the myocardium during hypoxia, such as was characteristic of emoxypine. The ATP and CP content in the myocardium of LR rats was somewhat higher in exposure to mexidol in any concentrations than in hypoxia alone (Table 1).

Hence, succinate introduced into the hydroxypyridine molecule promotes the transformation of the prohypoxic effects typical of emoxypine into the antihypoxic effects characteristic of mexidol. These antihypoxic effects are more marked in the myocardium of LR rats, this correlating with the protective effect of mexidol exhibited *in vivo* under pressure chamber conditions just in LR rats [7]. Taking into account the effect of mexidol on the energy status of the myocardium, one may speak of its direct energizing effect, associated probably with succinate oxidation in the respiratory chain.

A noteworthy fact is that emoxypine, in possessing marked antioxidative properties, exerts various effects on the hypoxic myocardium; this suggests diverse levels of LPO processes in the hypoxic hearts of HR and LR animals. Keeping in mind the more pronounced inhibitory effect of emoxypine observed in the myocardium of HR rats, we may assume that this level is lower in HR than in LR rats. Its further decrease in the presence of an antioxidant leads to the suppression of heart functional activity. Hence, antioxidant protection in hypoxia can aggravate hypoxic disturbances. These data, in turn, suggest that the antihypoxic effect of mexidol is due not to the antioxidant, but to other properties of the substance, namely the presence of succinate in the molecule of the hydroxypyridine derivative, which is capable of being oxidized in the respiratory chain under such conditions.

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