EFFECT OF CARNOSINE ON THE SYSTEMIC HEMODYNAMICS AND MYOCARDIAL METABOLISM IN RATS IN THE EARLY POSTRESUSCITATION PERIOD

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The early recovery period after resuscitation circulatory failure (CF) may develop. A pathogenetic role in its development is played by deficiency of the circulating blood volume [5], disturbance of the rheologic properties of the blood [9], and endogenous toxemia [14]. Besides the extracardiac factors mentioned above, damage to the heart muscle itself, induced by an excess of catecholamines, by extreme activation of lipid peroxidation (LPO) processes, and by disturbance of the energy metabolism of the myocardium [4], may also be of great importance in the development of CF at definite stages of the recovery period.

The aim of the present investigation was to attempt to reduce hemodynamic disturbances and metabolic changes in the heart after acute lethal blood loss with the aid of the dipeptide carnosine (β-alanyl-L-histidine).

METHODS

Experiments were carried out on 120 noninbred male rats anesthetized with pentobarbital (25 mg/kg), in two stages. In stage I the effect of the duration of clinical death on parameters of the systemic hemodynamics and on myocardial metabolism was estimated, and in stage II the possibility of diminishing postresuscitation disturbances of the hemodynamics and myocardial metabolism with the aid of carnosine was studied. Clinical death with a duration of 4 and 6 min was induced by acute blood loss, and resuscitation carried out by centripetal injection of the withdrawn blood and artificial ventilation of the lungs. The time of restoration of cardiac contractions and of respiration and the corneal reflexes was estimated. To record parameters of the systemic hemodynamics the RPG2-02 rheograph [6], ÉKTG-02 electrocardiograph, and N338-4P automatic writer were used. The mean blood pressure (BP) was measured by means of a mercury manometer. The heart rate (HR), cardiac output (CO), and total peripheral vascular resistance (TPVR) were calculated. The heart was removed 1.5 h after resuscitation, when CF was at its severest [4], and its content of lactate [15], pyruvate [1], and malonic dialdehyde (MDA) [11] was determined. The content of conjugated dienes in the lipid extract was studied by the method in [12]. Activity of superoxide dismutase (SOD) was determined in the postmitochondrial fraction of the myocardium and calculated per milligram protein [13]. Carnosine, in the previously chosen optimal dose of 25 mg/kg [10], was injected simultaneously with the returned blood during resuscitation. The results were subjected to statistical analysis by Student's test.

RESULTS

Table 1 shows that with an increase in the duration of clinical death from 4 to 6 min the times of recovery of cardiac contractions, respiration, and the corneal reflexes were delayed, and the efficacy of resuscitation was reduced

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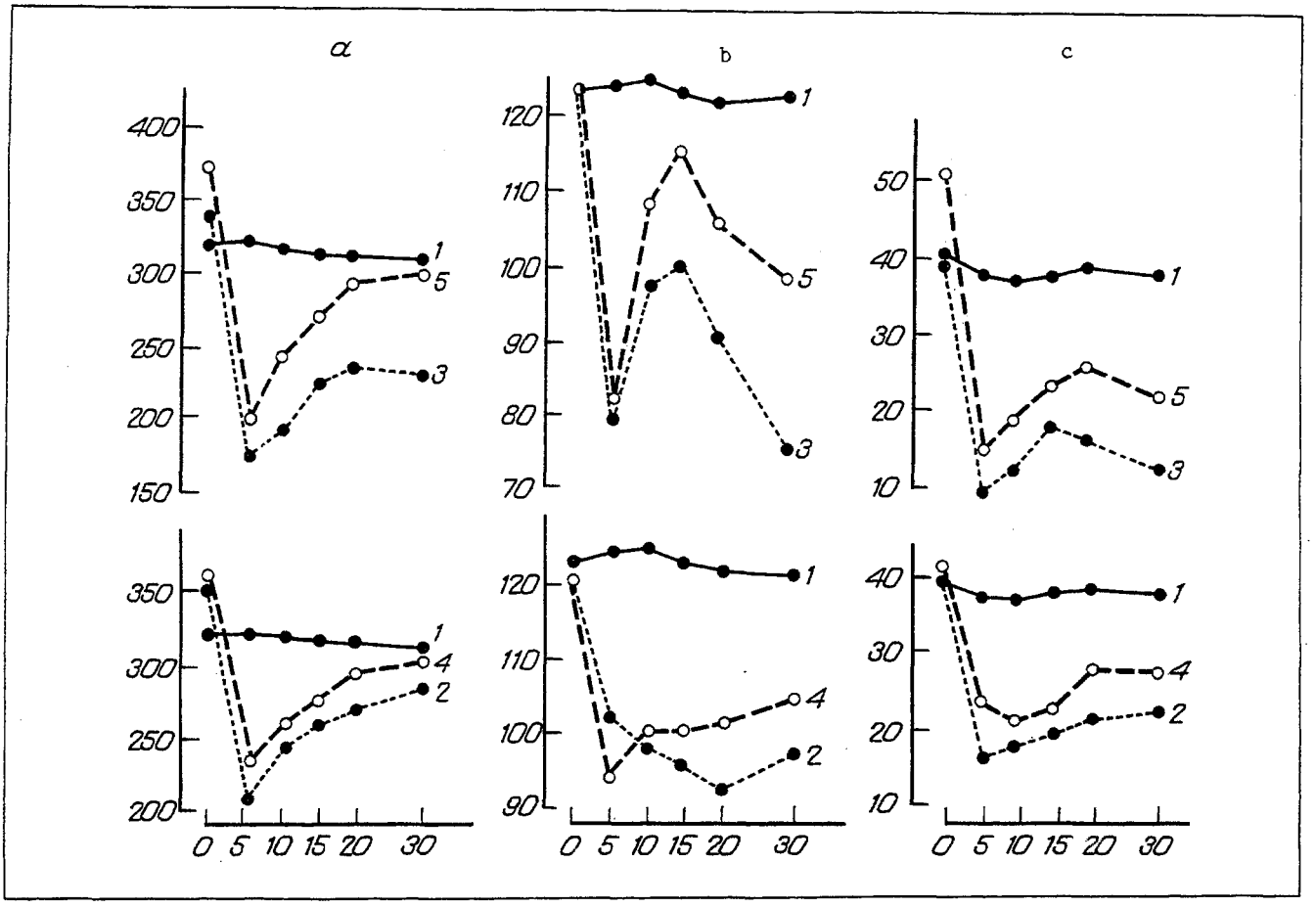

Fig. 1. Effect of carnosine on systemic hemodynamics of rats in early postresuscitation period: a) heart rate (min^{-1}); b) blood pressure (mm Hg); c) cardiac output (ml/min); 1) control; 2, 3) animals surviving clinical death for 4 and 6 min, respectively; 4, 5) animals surviving clinical death for 4 and 6 min and treated with carnosine. Abscissa) time after resuscitation (min).

TABLE 1. Effect of Carnosine on Restoration of Functions of Vital Organs and Systems during Early Postresuscitation Period (M ± m)

Parameter	Duration of clinical death			
	4 min	4 min + carnosine	6 min	6 min + carnosine
Time of restoration of				
Cardiac contractions, sec	95±10,5	76±4,6·	144±10,6*·	101±8,8·
Respiration, min	9,2±0,55	7,3±0,32·	11,3±0,73*	9,0±0,55·
Corneal reflexes, min	19±0,7	18±0,7	22±0,9*	17±0,7·
Efficacy of resuscitation, %	87,1	90,0	66,7	74,2
Mortality in the course of 1.5 h, %	18,5	3,7	31,8	17,4

Note. *) Significant differences between parameters in animals surviving clinical death for 4 and 6 min; ·) the same for animals resuscitated and treated with carnosine.

TABLE 2. Effect of Acute Lethal Blood Loss and Carnosine on Postresuscitation Metabolic Disturbances in the Heart (M ± m)

Parameter	Number of experiment					
	I	II	III	IV	V	VI
Conjugated dienes, mmoles/kg lipids	11,4±2,47	15,8±2,15	78,4±10,30*	42,7±4,20*·	81,4±13,23*	26,5±5,09*·
Malonic dialdehyde, mmoles/kg	5,0±0,86	6,4±0,45	12,7±1,72*	6,5±1,32·	14,2±0,53*	6,9±1,32·
SOD, units/(min·mg protein)	4,6±0,22	5,2±0,54	3,6±0,16*	4,9±0,33·	3,4±0,16*	4,7±0,31·
Pyruvate, mmoles/ kg	0,78±0,09	0,85±0,08	1,54±0,18*	1,12±0,10*·	2,08±0,22*	1,21±0,15*·
Lactate, mmoles/kg	2,26±0,14	2,20±0,20	6,11±0,15*	3,67±0,26*·	7,91±0,39*+	2,92±0,17*·

Notes. I) Control animals; II) animals receiving carnosine; III, V) animals surviving clinical death for 4 and 6 min, respectively; IV, VI) animals surviving clinical death for 4 and 6 min, respectively, and treated with carnosine. *) Significant differences from control; +) between groups III and V; ·) between experiments of series III and IV, and V and VI.

to 66.7%. One third of the animals developed asystole, fibrillation, or pulmonary edema actually during the period of clinical death or at the time of resuscitation. The early postresuscitation mortality reached 31.8%.

Injection of carnosine for therapeutic purposes led to a more rapid recovery of functions of vitally important organs and systems, increased the efficacy of resuscitation, and lowered the mortality in the most vulnerable period, i.e., during 1.5 h after resuscitation.

Analysis of the hemodynamic parameters (Fig. 1) shows that the low cardiac ejection syndrome was clearly demonstrable in rats in the early postresuscitation period. Minimal values of CO were reached in the first 10 min of the recovery period under conditions of marked bradycardia, lowered BP, and increasing TPVR, especially in animals surviving clinical death for 6 min. A tendency toward a small increase in CO appeared after 15 min, mainly due to quickening of the heart rate. However, 30 min after resuscitation CO was still only half of the control values in rats resuscitated after clinical death for 4 min, and two-thirds lower after clinical death lasting 6 min, grounds for the development of irreversible damage to the animal [8].

The pathogenetic importance of extracardiac factors (circulating blood volume deficiency, disturbance of rheologic properties of the blood, endotoxemia, metabolic acidosis, etc.) in the formation of the low cardiac ejection syndrome has been described in the literature [5, 8]. In our view, an essential role in the development of this syndrome may be played by damage to the heart itself, causing depression of its contractile function [4]. Table 2 shows clearly that acute lethal blood loss and subsequent resuscitation and reoxygenation activate LPO processes, as is shown by an increase in the content of CD and MDA. This process is facilitated by inhibition of SOD, an enzyme responsible for antiradical protection of the cell by dismutation of superoxide anion-radicals into H_2O_2 [3]. Hyperproduction of CD and MDA and of other peroxide compounds is known to cause damage to the bilipid layer of cardiomyocyte membranes, including membrane-localized enzymes of the Krebs' cycle, transport ATPases [7]. That is evidently why, with the lengthening of clinical death, the pyruvate concentration in the heart muscle rises in the early postresuscitation period (Table 2). In the intact heart, pyruvate formed during glycolysis undergoes oxidative decarboxylation with the form of acetylcoenzyme A, which is incorporated into the Krebs' cycle, which takes place completely in the mitochondria. Accumulation of pyruvate in the myocardium of the resuscitated rats is evidence of injury to the mitochondria and impairment of its incorporation into the tricarboxylic acid cycle. Reduction of energy formation in oxidative phosphorylation leads to compensatory activation of anaerobic glycolysis, as shown by the threefold increase in the lactate concentration in the heart muscle. Lactate itself has a marked cardiodepressive effect, which is regularly reflected in the parameters of the systemic hemodynamics (Fig. 1).

In the second stage of the investigations we showed that injection of carnosine during resuscitation reduces the metabolic disturbances in the heart muscle. In particular, the content of CD and MDA (products of LPO) was reduced by 2-2.5 times, but SOD activity was the same as in the control. Thus, carnosine, by restricting excessive activation of LPO, a characteristic feature of the early postresuscitation period, reduced the accumulation of toxic peroxide compounds, which have a detergent-like effect on the cardiomyocyte membranes. By exhibiting membrane-

protective properties [2], carnosine evidently prevented destruction of the mitochondrial membranes and inhibition of enzymes of the Krebs' cycle. Ultimately, the pyruvate content in the heart muscle was reduced by 37 and 72% (after clinical death for 4 and 6 min, respectively). The preparation limited the intensity of glycolysis as shown by a reduction of the lactate content in the myocardium by half.

Reduction of the metabolic disturbances in the myocardium of the animals treated with carnosine improved the hemodynamic parameters: CO and BP were higher than in animals not treated with carnosine, and the bradycardia did not reach such low values.

Thus carnosine, injected during resuscitation, has a direct beneficial effect on myocardial metabolism and an indirect effect on the systemic hemodynamics, for it increases the efficacy of resuscitation and reduces early postresuscitation mortality.

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