of the membrane, but have virtually no effect on intracellular conductance. Perfusion of erythrocytes "loaded" with fatty acids through a column containing the sorbent leads to opposite changes in their electrical characteristics: a sharp decrease in conductance and capacity of the membrane, much below the control levels obtained for intact erythrocytes. It can be postulated that not only the added fatty acids are removed from the plasma membranes of erythrocytes during hemoperfusion, but also membrane destabilizers present in them initially (hydrolysis products of phospholipids, peroxidation products of polyenic fatty acids, etc.) [2, 4].

It can thus be concluded from these results that the method of recording passive electrical properties of blood is a highly sensitive and informative means of monitoring the effectiveness of hemoperfusion. Further investigations will demonstrate to what extent the method can be used to select optimal conditions of hemoperfusion for different pathological states.

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REACTION OF NONENZYMIC FIBRINOLYSIS TO INTRAVENOUS INJECTION OF SMALL DOSES OF SALMONELLA ENDOTOXIN INTO RABBITS

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KEY WORDS: salmonella endotoxin; nonenzymic fibrinolytic activity of blood; heparin.

Acute intestinal infections are accompanied by endotoxemia, acute renal failure (ARF), and changes in hemostasis and the water-electrolyte balance.

The response of the blood clotting system to intravenous injection of endotoxins into animals has been studied in fair detail. One of the main symptoms reflecting the character of the effect of endotoxin on hemostasis is a state of hypercoagulation which arises when the endotoxin circulates in the blood stream, and if the toxin is injected continuously, intravenously over a period of several hours characteristic features of disseminated intravascular clotting (DIVC) develop [6, 10].

The main trigger mechanism leading to temporary thrombin formation in the circulating blood in response to intravenous injection of endotoxin may originate from different sources. For instance, according to some workers blood cells and, in particular, leukocytes of many species of animals, on contact with endotoxin, become the source of procoagulant activity. Platelets stimulated by endotoxin are one source of both procoagulant and proaggregating activity [8]. Experiments *in vitro* have demonstrated the cytotoxic action of granulocytes treated with endotoxin on a culture of endothelial cells [9]. Injury to the endothelium by endotoxin, mediated through granulocytes, is another factor in the stimulation of hypercoagulation and thrombin formation when endotoxin enters the blood stream.

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TABLE 1. Total and Nonenzymic Fibrinolytic Activity (TFA and NEFA respectively) of Blood, Antiplasmins, and R and K Indices of Thromboelastogram in Rabbits After Intravenous Injection of Salmonella Endotoxin (10 μg/kg) and of Endotoxin Together with Heparin (100-200 μg/kg + 120 IU/kg respectively) (M ± m)

Parameter studied	Group of animals				
	control (n = 15)	1 (n = 13)	2 (n = 14)	3 (n = 15)	4 (n = 7)
TFA, %	100,0±2,1	140,0±20,0	95,0±7,8	109,0±8,5	242,0±13,5*
NEFA, %	100,0±1,5	171,0±15,3*	135,0±10,1*	131,0±10,5*	209,0±16,5*
Antiplasmins, %	100,0±0,3	108,0±3,1	80,0±4,5*	109,0±3,4	—
R, mm	26,0±2,6	44,0±2,1*	45,0±3,1*	23,0±2,1	50,0±2,2*
K, mm	12,0±1,1	18,0±2,0*	16,0±2,5*	9,0±2,3	18,7±3,6

Legend. *P < 0.05 compared with control; n) number of animals.

When endotoxin is injected into animals it induces ARF, and this is the cause of the animals' death. Impairment of renal function may also affect the general state of hemostasis, for the kidneys synthesize blood clotting factors [5].

Excitation of function of the anticlotting system in response to thrombin generated in the blood stream activates a protective reaction against thrombus formation [1].

The object of this investigation was to study the dynamics of the reaction of the anticlotting system and, in particular, of its most important stage, namely nonenzymic fibrinolysis, to intravenous injection of small doses of salmonella endotoxin into rabbits, in the course of development of the pathological process. It was also interesting to determine the effect of heparin injections on the parameters of the reaction of nonenzymic fibrinolysis in animals with severe endotoxicosis. A further aim was to compare the development of ARF and morphological changes in the kidneys with the reaction of nonenzymic fibrinolysis in response to injection of endotoxin into the animals.

EXPERIMENTAL METHOD

Experiments were carried out on 75 Chinchilla rabbits weighing 2.5-3.0 kg. Salmonella endotoxin was injected into the auricular vein of the rabbits in a dose of 10 μg/kg body weight. The severity of the pathological process developing after injection of endotoxin was assessed by the degree of dehydration of the animals, which characterizes the course of ARF. The animals were divided into four groups: 1) those whose loss of body weight was under 5% of the initial value, 2) from 5 to 10%, 3) from 15 to 19%; rabbits of group 4 were given an injection of a lethal dose of endotoxin (100-200 μg/kg) followed immediately by an intravenous injection of heparin in a dose of 120 IU/kg body weight.

Blood was taken in a volume of 5 ml with a syringe containing anticoagulant by cardiac puncture. Activity of total and nonenzymic fibrinolysis [2] and the antiplasmin level [7] were determined in samples of blood plasma. To characterize activity of the clotting system, indices R and K of the thromboelastogram were determined after addition of 0.1 ml of 1.29% $CaCl_2$ solution to 0.26 ml of citrated plasma.

Morphological and histochemical investigations of the kidneys in animals dying as a result of injection of the endotoxin or killed after blood sampling were undertaken on frozen sections 5 μ thick. Pieces of kidney tissue were embedded in paraffin wax after fixation in 10% buffered neutral formalin and stained with hematoxylin and eosin [3]. Material for electron-microscopic investigation of the kidneys was fixed in 1% osmic acid solution and the samples were embedded in Epon-812. The JEM-100 electron microscope was used.

EXPERIMENTAL RESULTS

The results of the comparative study of nonenzymic fibrinolytic activity depending on the severity of the pathological process after injection of endotoxin, given in Table 1, show that definite activation of nonenzymic fibrinolysis was taking place in the blood of the animals of group 1 with a degree of dehydration reflected in a loss of body weight of up to 5% of the initial value, and the total fibrinolytic activity of the blood was increased. The R and K indices of the thromboelastogram demonstrate a state of hypocoagulation and depression of activity of the clotting system. The protective mechanisms of the anticlotting system were thus completely preserved, mainly on account of nonenzymic fibrinolysis, in animals with dehydration by 5%.

Morphological investigation of the kidneys revealed marked spasm of vessels in the cortex, narrowing of their lumen, thickening of their wall, and edema of the stroma of the medulla. Signs of necrosis were found in the tubules of the proximal portion, and this was confirmed by ultrastructural examination.

A lower degree of activation of nonenzymic fibrinolytic activity was observed in the animals of group 2, which had lost from 5 to 10% of their body weight. However, in this group the background level of anticoagulant activity in the blood still remained quite high, as shown by the indices of the thromboelastogram.

At this stage of the experiment a more severe disturbance of the blood flow was observed in the kidneys, with more marked hypoxia of the cortex and edema of the medullary stroma. Ultrastructural investigations revealed swelling and destruction of mitochondria in the proximal and distal portions of the nephron.

Aggravation of the pathological changes (animals of group 3, loss of up to 15-19% of initial body weight) also was reflected in further depression of the defensive reaction of of nonenzymic fibrinolysis. In the animals' blood a definite tendency was found for the state of hypocoagulation to be replaced by the development of hypercoagulation, indicating exchaustion of the defensive reactions of the anticlotting system.

In this system of investigations the morphological disturbances in the kidney affected more extensive areas and evidence of necrosis of the eipithelium in all parts of the nephron, severe hypoxia and ischemia of the cortex, and marked edema of the medullary stroma were observed, accompanied by a dramatic decline of enzyme activity in the nephron.

In animals of group 4, receiving a lethal dose of endotoxin followed by injection of heparin, high activity of total and nonenzymic fibrinolysis was observed in the blood, together with low activity of the clotting system. The mortality among the animals of this group after injection of the lethal dose of endotoxin also was reduced. Morphological investigation of the kidneys of these animals showed qualitative differences compared with those of the previous experiments: Only moderate disturbance of kidney structure was observed, in the form of degenerative changes in the proximal part of the nephron, with moderately reduced enzyme activity. Ultrastructural disturbances also were found only in the proximal part of the nephron, and the distal part was completely intact.

It can thus be concluded from the results that after a single intravenous injection of small doses of salmonella endotoxin into rabbits the protective reaction of nonenzymic fibrinolysis in connection with the function of the anticlotting system remains intact in the initial stage of development of the pathological process, but in the later stages of the disease this reaction becomes depressed. Changes in the kidneys revealed by morphological and ultrastructural investigations suggest that pathological changes in the renal vessels (spasm, disturbance of the circulation) and in the cortex contribute to thrombin formation, in addition to thrombin generation in the general blood flow (activation of blood cells, changes in vascular endothelium), and a protective reaction of the anticlotting system develops in resonse to its formation. The parallel noted between the reaction of nonenzymic fibrinolysis and the morphological and ultrastructural changes in the kidneys, depending on development of the pathological process after endotoxin injection, indicates that the nonenzymic fibrinolysis test can be used under certain conditions as a diagnostic test of the degree of severity of this pathological state.

The results of experiments with intravenous injection of heparin into animals receiving the endotoxin also are in agreement with existing evidence that heparin has a protective action against endotoxin by forming a complex with it. Acitivity of the endotoxin in the complex thus formed is abolished or considerably reduced [4].

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TIME COURSE OF LYMPH NODE FUNCTION AFTER DENERVATION

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KEY WORDS: lymph node; denervation; lymph flow.

The aim of this investigation was to study lymph flow in the popliteal lymph nodes of dogs in two groups of experiments: in the course of 1 week and 3 and 5 months after unilateral division of the sciatic and femoral nerves. A parallel investigation was undertaken on lymph nodes of the contralateral limb. Lymph nodes of intact dogs served as the control. Altogether 38 popliteal lymph nodes from 19 mature mongrel dogs were used. The operative access to the lymph node was obtained under thiopental anesthesia.

EXPERIMENTAL METHOD

Movements of the surface of the lymph nodes were measured by means of an apparatus, the sensory element of which was a 6MKh1S mechanotron, designed for the high-precision measurements within the rate of ±100 μ. The mechanotron was secured to a massive stand, and a contact surface measuring 3.5 mm^2 was fixed to the end of its rod, and pressed with a force of 200-300 mg force against the surface of the lymph node. Rapid movements of the lymph node capsule caused by pulsations of arteries, interfering with the recording of the slow oscillations, were eliminated by means of an RC circuit with time constant of 1.6 sec. The power source of the mechanotron consisted of a bipolar voltage source with stabilization factor of 5000 and with voltage pulsation of under 0.4 mV. The mechanogram was recorded for between 15 and 40 min, on an automatic X-Y Recorder, with built-in voltage amplifier.

EXPERIMENTAL RESULTS

In the course of 1 week after the experiment the results recorded on the mechanograms demonstrated acceleration of filling of the lymph nodes with lymph in the denervated and contralateral limbs. The period of oscillations of the lymph node capsule (filling and emptying of the node) in the denervated lymph nodes averaged 2 min 27 sec ± 3 sec (Fig. 1B), and in the contralateral nodes 1 min 42 sec ± 3 sec (Fig. 1C), which is much shorter than the period of oscillations of the capsule in the lymph nodes of intact dogs, namely 3 min 26 sec ± 4 sec (Fig. 1A). At the time of maximal filling of the denervated and contralateral lymph nodes with lymph the amplitude of the oscillations showed a tendency to increase compared with the initial level (49.2 ± 2.2 μ).

In the later postoperative stages (3-5 months) the period of oscillation of the capsule of the denervated lymph nodes remained virtually unchanged (2 min 8 sec ± 3 sec Fig. 1D), but in the lymph nodes of the contralateral limb it became statistically significantly slower (2 min 34 sec ± 5 sec, Fig. 1E), although as before the period of oscillation was less than that in intact animals. The amplitude of the oscillations during maximal filling of the denervated (22.6 ± 2.5 μ) and contralateral nodes (21.7 ± 2.7 μ) was less than half of that both in intact nodes (49.2 ± 2.2 μ) and at the previous postoperative stage.

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