3. R. Bergerhof (1954). Cited by V. P. Baluda, Z. S. Barkagan, E. D. Gol'dberg, et al., Laboratory Methods of Investigation of the Hemostasis System [in Russian], Tomsk (1980), p. 129.
4. M. R. Buchanan, R. W. Butt, J. Hirsch, et al., Prostagland. Med., 21, 157 (1986).
5. J. B. German and J. E. Kinsela, Biochim. Biophys. Acta, 877, 290 (1986).
6. P. Henriksson, M. Hamberg, and U. Diezjalusy, Biochim. Biophys. Acta, 834, 272 (1985).
7. H. Kuhn, R. Wiesner, L. Alder, et al., FEBS Lett., 208, 248 (1986).
8. M. Lecomte, D. Nunez, and J. M. Boeynaems, Prostaglandins, 32, 150 (1986).
9. L. G. Letts, M. Cirino, P. Yushko, et al., Prostaglandins, 29, 1049 (1985).
10. P. Mehta, J. Mehta, D. Lawson, et al., Thrombos. Res., 41, 731 (1986).
11. S. J. Rabiner and O. Hrodek (1968). Cited by V. P. Baluda, Z. S. Barkagan, E. D. Gol'dberg, et al., Laboratory Methods of Investigation of the Hemostasis System [in Russian], Tomsk (1980), p. 104.
12. C. Richards, A. R. Johnson, and W. P. Campbell, Biochim. Biophys. Acta, 875, 569 (1986).
13. T. Schewe, R. Wiesner, and S. M. Rapoport, Meth. Enzymol., 71, 430 (1981).
14. K. K. Wu and J. C. Hoak, Lancet, 2, 924 (1974).

ANGIOPROTECTIVE EFFECT OF CATHERGEN IN EXPERIMENTAL LIVER DAMAGE

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KEY WORDS: cathergen; microcirculation; mast cell

Disturbances of the microcirculation arising during pathological processes in the liver call for correction by drugs. For the treatment of liver disease, cathergen, a synthetic analog of bioflavonoids, has been used with success. However, the effect of cathergen on the microcirculatory system has not been studied. It was accordingly decided to undertake the investigation described below.

EXPERIMENTAL METHOD

A model of acute toxic liver damage with disturbance of the microcirculation was created by intraperitoneal injection of carbon tetrachloride (CCl_4) in a dose of 1 mg/kg (40 albino rats) or of ethanol in a dose of 6 g/kg (40 rats). Half the animals of both groups received preliminary injections of cathergen in a dose of 120 mg/kg once a day for 2 weeks. The control

TABLE 1. Changes in Number of Mast Cells and Their Degree of Degranulation under the Influence of Cathergen, CCl_4, and Ethanol (M ± m)

Parameter	Control	Cathergen	CCl_4	CCl_4 + cathergen	Ethanol	Ethanol + cathergen
Number of mast cells in 1 m^2	62,3±2,28	68,8±2,43	66,2±2,32	92,9±1,71	51,1±1,52	82,2±2,48
p_1		>0,05	>0,05	<0,001	<0,01	<0,001
p_2			>0,05	<0,001	<0,001	<0,001
p_3				<0,001		<0,001
Index of degranulation of mast cells	1,05±0,02	0,52±0,03	2,20±0,05	0,41±0,04	1,55±0,04	0,46±0,02
p_1		<0,001	<0,001	<0,001	<0,001	<0,001
p_2			<0,001	≤0,05	<0,001	>0,05
p_3				<0,001		<0,001

Legend. p_1) Significance of differences compared with control, p_2) compared with values obtained in animals receiving cathergen, p_3) between values obtained in animals receiving CCl_4 or ethanol and values obtained in rats receiving a combination of CCl_4 or ethanol with cathergen.

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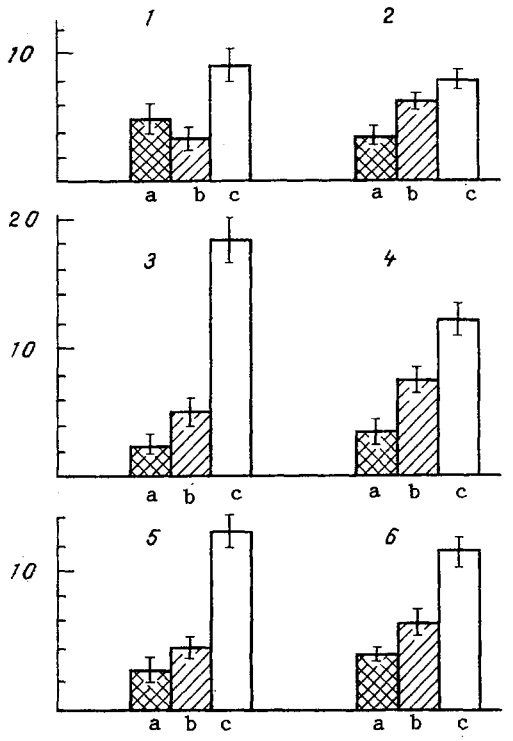

Fig. 1

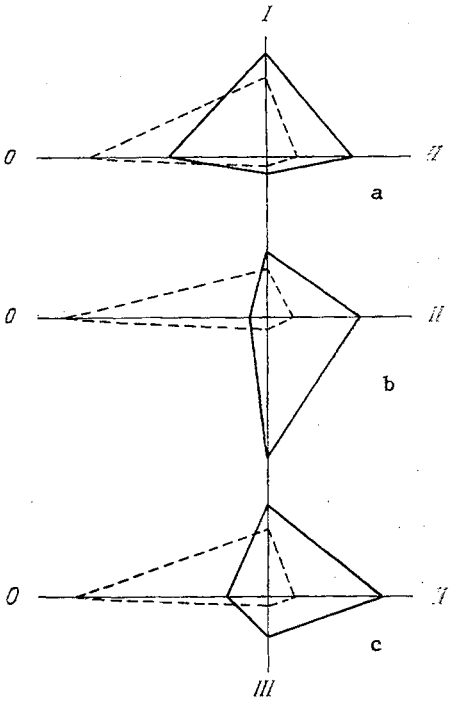

Fig. 2

Fig. 1. Changes in bulk density (in %) of capillary, venous, and arterial components of microcirculatory bed under the influence of CCl_4, ethanol, and cathergen. a) Arterial bed; b) capillary bed; c) venous bed.

Fig. 2. Effect of cathergen on relative numbers (in %) of mast cells of form 0, I, II, and III in intact rats (a), and following injection of CCl_4 (b) and ethanol (c). Broken line denotes ratio between numbers of forms of mast cells in animals receiving cathergen.

group consisted of 20 intact rats and 20 albino rats receiving cathergen without the poison. The action of cathergen was tested 24 h after production of liver damage. Microangiometry was carried out with measurement of the diameter of the vessels and the bulk density of the microcirculatory bed and with assessment of the mast cells for their degree of degranulation, with distinction of their 0, I, II, and III forms, were carried out on film preparations of the falciform ligament of the liver and the mesentary, stained with azure-eosin.

EXPERIMENTAL RESULTS

After injection of CCl_4 and ethanol the arteriolovenular ratio decreased on average to 0.3 and 0.38 respectively (0.46 in the control). This is evidence of disproportion between the inflow and outflow of blood in the microcirculatory system. Preliminary administration of cathergen restored equality of the ratio of the arterial inflow to the venous outflow. The arteriolovenular ratio was 0.44-0.46 after administration of cathergen.

Following administration of cathergen the diameter of the microvessels of the capillary-venular portion of the microcirculatory bed was less than in animals not receiving cathergen therapy. For instance, when administration of cathergen was followed by injection of CCl_4 the following diameters of the microvessels were observed: capillaries 7.3 ± 0.13 μ (without cathergen 9.6 ± 0.34 μ), postcapillaries 13.1 ± 0.41 μ (without cathergen 24.7 ± 0.48 μ), venules 32.2 ± 1.03 μ (without cathergen 42.7 ± 2.69 μ). In the group of animals receiving cathergen and ethanol, only the diameter of the postcapillaries was significantly reduced (to 13.2 ± 1.1 μ; without cathergen 19.3 ± 0.21 μ). Cathergen evidently increases the resistance of the microvessels and stabilizes their throughput at the optimal level under altered hemodynamic conditions. Meanwhile cathergen increases the bulk density of the capillary bed under normal hemodynamic conditions and maintains high values of bulk density of the capillary bed when the density of the venous bed is relatively reduced (Fig. 1). The microvascular effects are linked

with the normalizing action of cathergen on autoregulation in the "mast cell–microvessel" system. Administration of cathergen lowered the intensity of degranulation of the mast-cell apparatus, with an increase in the proportion of 0-forms of mast cells (Table 1; Fig. 2). It can be tentatively suggested that cathergen has an angioprotective action when the circulation of the blood is disturbed.

ACID HYDROLASE ACTIVITY AND CYCLIC NUCLEOTIDE CONTENT IN THE RAT HEART DURING MYOCARDIAL ISCHEMIA AND POSTISCHEMIC REPERFUSION

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Myocardial ischemia causes significant changes in cyclic nucleotide levels [2, 6, 13], destabilizes lysosomal membranes, reduces the latency of lysosomal enzymes, and enhances hydrolase activity in the cell cytoplasm [3, 6, 8, 12]. Cyclic nucleotides play an important role in the regulation of the state of the lysosomal membranes; cAMP, moreover, has a stabilizing action on lysosomal membranes whereas cGMP has the opposite effect [1, 4, 5, 9]. Since methylxanthines, by blocking phosphodiesterase [5], may lead to an increase in the cAMP concentration in the cardiomyocytes, it was thought worthwhile to use substances of this group to study the possible mechanisms of damage to lysosomal membranes in myocardial ischemia and to develop methods of correcting these lesions. In the light of the view that damage to cardiomyocytes is aggravated on restoration of the blood flow along the coronary vessels after myocardial ischemia lasting about 40 min or more [2, 3, 10, 11], there is also an urgent need for a study of the effect of methylxanthenes on cyclic nucleotide concentrations and activity of lysosomal enzymes during postischemic reperfusion.

The aim of this investigation was to study activity of lysosomal hydrolases and cyclic nucleotide levels in tissues of the heart associated with ischemic damage to the myocardium and during the period of postischemic reperfusion, and also to examine the effects of caffeine, a phosphodiesterase blocker, on the state of the lysosomes and cyclic nucleotide levels.

EXPERIMENTAL METHOD

Experiments were carried out on 60 isolated hearts of noninbred male albino rats, perfused by Langendorff's method. The hearts were perfused with Krebs' solution, aerated with a gas mixture consisting of 95% O_2 and 5% CO_2 (temperature 37°C, pH 7.4, perfusion flow rate 10 ml/min, isovolumic cardiac contractions). In the experiments of series I, after perfusion for 20 min under the conditions described above, the perfusion rate was reduced to 1 ml/min, and 40 and 60 min after the beginning of ischemia, tissues of the left ventricle were removed for investigation. In some experiments, after 40 min of ischemia, reperfusion was carried out in the course of 20 min, the perfusion rate being increased to its initial value. In the experiments of series II, 30 min after the beginning of ischemia the hearts were switched to perfusion with Krebs' solution containing 50 μM of caffeine (from "Serva," USA). The conditions of perfusion of the control hearts remained unchanged throughout the experiment. Concentrations of cAMP and cGMP in tissues of the left ventricle were determined by radioimmunoassay using kits from the firm "Amersham International" (England). Activity of cathepsin D and acid phosphatase was determined spectrophotometrically [7]. Two forms of hydrolase activity were investigated: the so-called free activity (FA), i.e., activity of the enzyme in supernatant obtained by centrifugation of the homogenate at 30,000g for 30 min, and bound (lysosomal) activity (LA), or activity of the enzyme in the lysosome-rich fraction obtained by differential centrifugation followed by treatment of the lysosomal fraction to release the bound enzyme with Triton X-100 in a final concentration of 0.1%.

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