Role of Leukotrienes in Stress-Induced Damage to the Heart

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Experiments on rabbits with epinephrine-induced damage to the heart showed that progression of necrotic degenerative processes in the myocardium is augmented by increased content of prostaglandin $F_{2\alpha}$ and predominance of constrictive over vasodilatory effects in the E and $F_{2\alpha}$ prostaglandin system. Twenty-four hours after injection of epinephrine we observed an increase in blood concentrations of myofibrillar fraction of creatine phosphokinase, serotonin and histamine, and the $F_{2\alpha}$/E prostaglandin ratio. The concentration of leukotriene B_4 increased during the urgent adaptation period, which correlated with a decrease in elastic properties of erythrocytes. *In vitro* passage of erythrocytes through a 2.5-μ capillary sieve sharply decreased, which played an important role in the progression of myocardial and cerebral hypoxia and ischemia. Myocardial necrosis and endothelial dysfunction developed 24 h later aggravate these pathological shifts.

Key Words: *myocardium; epinephrine stress; leukotrienes; prostaglandins; serotonin; histamine*

Leukotrienes C_4, D_4, and E_4 belonging to the class of cysteine-containing leukotrienes, produce potent effects on the cardiovascular system. They modulate cardiac function and induce vascular modification leading to the development of edema [7,9]. Leukotrienes C_4 and D_4 induce spasm of cerebral and cardiac vessels, increase blood pressure; leukotriene E_4 in a concentration of 10^{-7} M potentiates contraction caused by potassium depolarization and amplifies the contractile response to prostaglandin (PG) $F_{2\alpha}$. Since the effects of $PGF_{2\alpha}$ and thromboxane α_2 are realized through the same receptors, a synergistic effect of leukotriene E_4 and thromboxane was hypothesized. In contrast, PGI_2 reduces the vasoconstrictor effect of cysteine leukotrienes on smooth muscles [1]. All these facts confirm the hypothesis that leukotrienes not only participate in inflammatory reactions, but also exert systemic vasoconstrictor effects. Interrelated production of cysteine leukotrienes by neutrophils and endothelial cells (leukotriene C_4) and their increased plasma content are important risk factors in circulatory disorders, *e.g.* in shock, hypoxia, inflammation, and stress.

Here we measured the content of leukotrienes and PG in epinephrine-induced heart damage, when the hemodynamic shifts induced by diffuse necrotic myocardial injury were aggravated by secondary inflammatory reaction.

MATERIALS AND METHODS

The study was carried out on 18 Chinchilla rabbits weighing 2-4 kg: 6 controls and 12 experimental animals with epinephrine-induced myocardial damage [4]. The animals were kept under standard vivarium conditions and were sacrificed 2 and 24 h after epinephrine injection.

The content of epinephrine and histamine were measured by spectrofluorometry, leukotrienes C_4 and B_4 were assayed by fluorometry after their isolation on a column with *o*-ophthalmic dialdehyde [5,9]. The content of PG was measured by radioimmunoassay using commercial kits (Clinical Assays), myofibrillar fraction of creatine phosphokinase (CPK-MB) was measured using commercial kits (Boehringer Mannheim).

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The results were processed statistically by routine methods for small samples of dependent and independent groups; the significance of differences was evaluated using Student's *t* test.

RESULTS

The blood content of CPK-MB in animals with epinephrine-induced myocardial damage increased almost 4-fold in comparison with the control 2 h after injection of epinephrine, which confirmed the development of necrotic changes in the myocardium. In parallel, the content of histamine in the ventricular myocardium increased by 26.5%, indicating the development of inflammatory allergic response to the formation of necrotic foci. Epinephrine content in the myocardium increased by 71 times by the 2nd hour, while the norepinephrine/epinephrine ratio was by 81 times below the control (because norepinephrine content remained unchanged). Epinephrine-induced vasoconstriction was potentiated by increased content of vasoconstrictor $PGF_{2\alpha}$ and absence of significant changes in PGE content, which led to predominance of the vasoconstrictor components of the PG system over its vasodilatory factors (Table 1). These changes aggravate epinephrine-induced injury and underlie persistent myocardial injury in epinephrine stress.

Similar changes were observed in the blood. A direct correlation was found between norepinephrine/epinephrine and PGE/$PGF_{2\alpha}$ ratios in the myocardium and blood (r=0.73, p<0.01 and r=0.69, p<0.05, respectively). The content of cysteine leukotriene B_4 synthesized in erythrocytes increased by 123% by the 2nd hour, while the content of leukotriene C_4 produced by endothelial cells remained virtually unchanged. An inverse correlation between the content of leukotriene B_4 and elastic characteristics of erythrocytes (r=0.74, p<0.01) was observed.

Twenty-four hours after modeling of epinephrine-induced myocardial damage the content of epinephrine in the myocardium decreased 10-fold, while the norepinephrine/epinephrine ratio increased almost 8-fold (Table 1). However, the content of inflammation markers serotonin and histamine increased by 33% com-

TABLE 1. Content of Epinephrine, Norepinephrine, Serotonin, Histamine, Prostaglandins, and Leukotrienes in the Myocardium and Blood in Epinephrine-Induced Myocardial Damage (*M*±*m*)

Parameter	Normal control	Epinephrine damage, h	
		2	24
Myocardial ventricles			
Epinephrine, μg/g	0.07±0.01	5.0±1.1*	0.5±0.1*$^{+}$
Norepinephrine, μg/g	1.25±0.4	1.0±0.2	0.8±0.1
Norepinephrine/epinephrine	17.9±1.8	0.21±0.04*	1.63±0.16*$^{+}$
Serotonin, μg/g	6.3±0.5	6.1±0.3	10.3±1.3*$^{+}$
Histamine, μg/g	3.4±0.5	4.3±0.4***	7.8±0.6*$^{+}$
PGE, ng/g	3.0±0.8	3.6±0.9	2.2±0.9**
$PGF_{2\alpha}$, ng/g	3.4±0.6	5.8±0.5**	7.4±0.9**$^{++}$
PGE/$F_{2\alpha}$	0.88±0.07	0.62±0.07	0.30±0.10*$^{+}$
Blood			
Epinephrine, pg/ml	4.5±0.8	14.5±1.8*	5.9±0.8***$^{+}$
Norepinephrine, μg/ml	2.3±0.6	4.3±0.8**	2.1±0.2
Norepinephrine/epinephrine	0.51±0.08	0.30±0.05**	0.36±0.09***
CPK-MB, nmol/ml	2.3±0.8	8.9±2.8**	12.5±1.8***$^{+++}$
Serotonin, nmol/ml	39±5	49±3***	56±3***$^{+++}$
Histamine, nmol/ml	0.24±0.05	2.85±0.15*	1.58±0.13*$^{+++}$
PGE, ng/g	65±3	189±21*	102±15**$^{+}$
$PGF_{2\alpha}$, ng/g	105±10	385±16*	402±21*
PGE/$F_{2\alpha}$	0.62±0.06	0.49±0.05**	0.25±0.04*$^{+}$
Leukotriene C_4, nmol/ml	5.7±0.8	6.7±0.8	14.3±1.5*
Leukotriene B_4, nmol/ml	5.8±0.8	8.2±0.8***	10.2±2.1**$^{++}$
Deformability, arb. units	12.5±3.4	20.6±4.5	89±9*$^{+}$

Note. *p<0.001, **p<0.01, ***p<0.005 compared to the control; ^{+}p<0.001, ^{++}p<0.01, ^{+++}p<0.05 compared to epinephrine damage (2 h).

pared to 2-h epinephrine damage and by 27 and 129% compared to the control (Table 1). The balance in the PG system was further shifted towards the predominance of the vasoconstrictor component. Blood concentration of CPK-MB increased almost 6-fold compared to its initial level and was 40% higher than 2 h after epinephrine injection, which attested to progression of necrotic changes in the myocardium. In contrast to serotonin, blood content of histamine decreased. PGE content also decreased in comparison with its level 2 h postinjection, while the content of $PGF_{2\alpha}$ did not change significantly, which determined predominance of vasoconstrictor components of the PG system.

In comparison with the previous term (2 h), we observed the increase in blood concentration of not only leukotriene B_4 (produced by activated neutrophils from leukotriene A_4 and converted into B_4 by specific hydrolase in erythrocyte [6,8]), but also leukotriene C_4 (produced in endothelial cells in the reaction catalyzed by leukotriene A_4 synthase [8,10] with participation of glutathion [10]). This probably suggests that necrosis and inflammatory reaction were complicated by endothelial dysfunction. The increase in leukotriene B_4 content correlated with further impairment of erythrocyte elasticity (r=0.8, p<0.001). *In vitro* erythrocyte passage through a 2.5-μ capillary filter sharply decreased, which, together with impaired capillary permeability [11] played an important role in the progression of hypoxic and ischemic phenomena not only in the myocardium, but also in the brain. New drugs correcting the leukotriene component during necrotic degenerative and inflammatory damage to the heart are needed.

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