of these fetuses to changes in the hemodynamics of the maternal organism.

The drop of fetal body temperature observed in our investigations after the injection of trental and trental in combination with rheopolyglucin into the female is connected with increased heat transfer due to a boost of the MPBF more moderate than after the injection of rheopolyglucin, not accompanied by a rise of the venous pressure in the female. The increase of the TG between ND fetuses and the mother is related to the fact that in the case of a high initial blood flow rate in the uterus a drop occurred under the influence of trental and trental in combination with rheopolyglucin, i.e., after the combined infusion of trental and rheopolyglucin the effect of trental prevailed. The drop of the fetal body temperature went along with a qickening of the heart 1rate. Thus, after the infusion of rheopolyglucin, trental, and their combined application in the rabbit females, the changes in fetal heart activity depend more upon the changes in the rate of placental blood circulation, than upon the type of fetal temperature reaction (judging by the absence of a direct dependence between fluctuations of the fetal body temperature and heart rate). The changes in temperature gradient between fetus and mother for different influences on the mother can be used as an indication of the changes in the rate of the maternal-placenta l and interrelated fetal-placenta l blood flow.

Thus, the disturbances in temperature homeostasis of the fetuses for the injection of drugs into the female are due mainly to the changes of the rate of the MPBF, and hence in placental defficiency therapy and in the case of developmental delay of the fetus the possible adverse effect of these drugs on the temperature regime of the fetus and consequently on its metabolic processes must be taken into account.

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Effect of Defensin on Platelet Functional Activity

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Defensins comprise a family of nonenzymatic cation proteins accumulated in the phagocyte granular apparatus of mammals [7]. As established, defensins possess a broad spectrum of antimicrobial, antiviral, and cytotoxic activity and modulate hormonal responses [7, 8, 15]. Human defensin HNP-1 is a strong and

TABLE 1. Effect of Human Defensin (40 μg/ml) on Platelet Aggregation Induced by Thrombin, Collagen, and ADP (M±m)

Parameter	Thrombin, 0,5 U/ml (9)		Collagen, 4mkg/ml (7)		ADP, 5 mkM (5)	
	control	defensin	control	defensin	control	defensin
Aggregation amplitude, %	83,0±5,8	43,0±5,1*	51,9±7,6	39,3±9,4*	74,0±2,7	51,5±2,1*
Maximal rate of aggregation, %/min	99,5±12,0	78,7±7,8	74,3±7,5	52,4±6,4*	42,6±6,0	26,8±5,8

Note: Here and in Table 2 figures in parenthesis denote the number of experiments; values with $p < 0.01$ (in comparison with control) are marked with an asterisk.

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specific chemoattractant for monocytes, this probably promoting their accumulation in regions of inflammation [14]. The participation of defensins in the development of tissue inflammation, injury, and regeneration [5] makes it important to study their effect on the processes of hemostasis and platelet formation. However, the data on the changes in the activity of the blood anticoagulative system in rats after intraperitoneal administration of defensin are rather scant [2]. Besides, elucidation of the effect of defensin on platelet functional activity deserves the special attention of investigators.

The object of the present work was to study the effect of human defensin on platelet aggregational activity and to identify some of the possible mechanisms involved.

MATERIAL AND METHODS

Defensins were extracted from human donor blood according to Kokryakov [1]. The total fraction of human defensin was used in the experiments. According to electrophoretic analysis, it comprised three main components: HNP-1, HNP-2, and HNP-3.

Venous blood from healthy donors was taken into 3.8% sodium citrate (1:9). Platelets were pelleted down by centrifugation [10]. After two washings, they were resuspended in buffer containing 20 mM HEPES, 135 mM NaCl, 1 mM $CaCl_2$, 1 mM $MgCl_2$, and 10 mM glucose, at pH 7.4. The platelet number in suspension constituted 250,000 in 1 ml. Platelet aggregation in suspension was studied in a PICA lumi-aggregometer (Chrono-Log, USA). The amplitude and maximal rate of aggregation were measured in a Aggro-Link apparatus (Chrono-Log). Agonists were added to the platelet suspension in the 5th minute of their incubation with defensin. Collagen (4 μg/ml) (Chrono-Log), ATP (5 μM) (Sigma), and thrombin (0.5 U/ml) (Chrono-Log) were used as inductors of platelet aggregation. The ATP release by the platelets was registered concomitantly with the recording of their aggregation at a luminiescence channel of the aggregometer using luciferin-luciferase (Chrono-Log) and standard ATP (Chrono-Log). The level of malonedialdehyde (MDA) in the platelets was determined after analysis of their aggregation in the same samples by spectrophotometry [13] at a 532 nm wavelength in a Beckman spectrophotometer (model DU-50).

TABLE 2. Changes in MDA Production in Platelets Induced by Thrombin and Collagen in the Presence of Defensin (40 μg/ml) and without the Peptide (M±m)

Parameter	Basal level	Thrombin 0,5 U/ml (7)	Collagen, mkg/ml (7)
MDA, nM/10^9 platelets control	0,6±0,3	3,3±0,7	3,9±0,8
defensin	1,0±0,3	2,1±0,3*	2,3±0,7*

Statistical analysis of the data was performed using the Student and paired Wilcoxson tests.

RESULTS

In the concentrational range of 0.1-40 μg/ml defensin did not induce platelet aggregation. As the defensin concentration increased to 100 μg/ml, insignificant platelet aggregation was registered which reached the maximal value approximately by the 10th minute and constituted 10.3+2.1%.

Preliminary incubation of platelets with defensin (40 μg/ml) reduced their aggregation induced by thrombin, collagen, or ADP in comparison with the control, *i.e.*, in the absemnce of defensin (Table 1). Defensin caused a significant decrease in both the intensity and the rate of agonist-induced platelet aggregation, independently of the type of inductor used. It is noteworthy that when spontaneous platelet aggregation occurred in some experiments, the presence of defensin in the sample prevented its further development.

The quantity of ATP released from platelet granules in the course of thrombin- and collagen-induced aggregation was reduced in the presence of defensin by 45.7 ($p<0.01$) and 21.1% ($p<0.01$), respectively, as compared to the control. Together with the absolute quantity of ATP, defensin reduced the rate of ATP release after thrombin or collagen stimulation of platelets.

The basal level of MDA in the platelets after their incubation with defensin (40 μg/ml) did not change significantly. However, in the presence of defensin the intensity of MDA production induced by thrombin and collagen decreased by 35.4 ($p<0.01$) and 40.3% ($p<0.01$), respectively, in comparison with the control (Table 2).

The problem of the possible involvement of neutrophils into regulation of platelet functional activity is now being widely discussed [9, 11]. At present, several mechanisms of down-regulation of platelet activity by neutrophils are known, one of them being related to the production of an NO-like endothelium-dependent relaxing factor by neutrophils [11]. Besides, neutrophils are capable of inhibiting platelet activity through the release of elastase, which causes limited proteolysis of platelet membrane glycoproteins [3] as well as through neutralization of ATP secreted by cell granules [12].

It seems possible that defensins contained in the neutrophil granular apparatus may, due to their positive charge and ability to interact with polyanionic platelet aggregates [5], also participate in cell-cell interactions. The results obtained on platelet aggregation induced by defensin at a concentration exceeding 40 μg/ml confirm the above data. However, at lower concentrations defensin did not elicit platelet aggregation, preventing at the same time the development of thrombin-, collagen-, and

ADP-induced aggregation. According to published data, defensin molecules contain lipophilic regions and can accomplish passive diffusion through the membrane lipid bilayer by hydrophobic interactions [5]. As demonstrated [4], defensins of human neutrophils act as inhibitors of protein kinase C which, in turn, mediates platelet aggregation, secretion from granules, and activation of arachidonic acid metabolism [6]. Without questioning the importance of this observation, we are inclined to believe, however, that the decrease in platelet functional activity in the presence of defensin revealed in the present study may be accounted for by a direct membrane-protective effect of the peptide.

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Myoglobin as an Indicator of Stress-Induced Damage

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Significantly elevated activity of cytoplasmic and lysosomal enzymes in the blood (i.e., enzymemia) is generally regarded as an important criterion for judging the severity of stress-associated damage to the body [4]. It has also been shown that elevated blood myoglobin is the earliest marker of damage sustained by the heart muscle in acute myocardial infarction [2, 13], which is known to be accompanied by a stress reaction [14]. However, the questions of whether myoglobin is released into the blood in the event of stress-associated damage and to what extent myoglobinemia may be relied upon as a criterion of such damage and of the efficacy of protective measures against stress have remained open. In view of this, the present study was undertaken to evaluate the impact of a relatively short single exposure of animals to stress on their blood content of myoglobin and to see how adaptation to periodic hypoxia, which is known to afford protection against stress [10-12], might affect myoglobinemia in the stressed animals.

MATERIAL AND METHODS

The experiments were carried out on four groups of male Wistar rats weighing 300±50 g: intact rats (group 1, control), those exposed to stress on a single occasion (group 2, stressed), those adapted to periodic hypoxia (group 3, hypoxia-adapted), and those exposed to such stress after adaptation to periodic hypoxia (group 4, hypoxia-adapted + stressed). The rats were stressed by being kept immobile in the supine position for 1 h. For adaptation to hypoxia, they were kept in a pressure chamber during 4 h daily over 40 days, starting with an "altitude" of 1000 m above sea level

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