ACADEMICIA
An International Multidisciplinary Research Journal
(Double Blind Refereed & Peer Reviewed Journal)

DOI: 10.5958/2249-7137.2021.02328.4

PECULIARITIES OF HEMORHEOLOGICAL DISORDERS IN THE PATHOGENESIS OF MICROCIRCULATOR DISORDERS OF THE LIVER DURING THE DEVELOPMENT OF HYPOXIC HYPOXIA

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ABSTRACT

The experiments were carried out on 50 white rats of a mixed population with an initial weight of 150-220 grams, kept in a vivarium on a standard laboratory diet. The state of the microvasculature of the liver has been studied, and a quantitative assessment of the changes occurring during experimental hypoxic hypoxia has been given. A comprehensive analysis of the state of the rheological properties of blood in close relationship with the properties of the erythrocyte membrane in the dynamics of the development of experimental hypoxia is presented.

KEYWORDS: *Hypoxia, Microcirculation, Hemorheology, Liver.*

INTRODUCTION

The problem of oxygen starvation of biological systems continues to be the focus of attention of researchers. This is due to the fact that hypoxia is a leading factor in the occurrence and development of many pathological processes associated mainly with dysfunctions of the blood, circulatory and respiratory systems [1,8].

Modern ideas about the mechanisms of pathological changes in hypoxic conditions are based on experimental and clinical data. In this aspect, the study of the hypoxia of critical states, which is a complex multifaceted biological phenomenon arising as a result of disorders in the activity of various organs and systems of the body, is of great importance.

In recent decades, the problem of hypoxia has been intensively developed in various directions [2,3,4]. At the same time, along with the indisputable successes, a number of issues of fundamental nature and requiring in-depth methodological analysis were clearly identified.

Various aspects of the hypoxic state were studied with a predominant study of the function of external respiration, respiratory failure and the state of arterial oxygenation [5].

However, in most cases, hypoxia is formed at practically all stages of the body's vital activity and at all stages of oxygen transport from the alveoli to the cell, i.e. represents changes in essentially all bodily functions. That is why the assessment of hypoxia as a clinical phenomenon requires the study of many body functions.

Modern methods of studying the manifestations of hypoxia, unfortunately, do not allow us to obtain comprehensive information about the essence of physiological events occurring in the body. Understanding hypoxia, its pathogenesis and clinical essence is impossible without simultaneously studying the processes occurring in the liver, blood, the microcirculatory component of oxygen transport and the state of tissue metabolism.

The development of hypoxia is accompanied not only by an increase in energy imbalance, but also by structural and functional damage to cells and tissues, changes in the reactivity of blood vessels, a violation of their nutrition, and sharp violations of the rheological properties of blood [6,7]. These disorders lead to altered functions of vital organs with high metabolic activity, characterized by abundant blood supply and sensitive to oxygen starvation. Therefore, a comprehensive study of the rheological properties of blood and the state of microcirculation of internal organs (liver, kidneys, pancreas) is of great scientific interest in order to deepen the understanding of the pathogenesis of hypoxia.

According to some authors, during the first hours of hypoxic hypoxia, the alkaline reserve changes little, there is only a decrease in pCO2 due to hyperventilation, followed by an increase in blood pH. After a few hours, the alkaline reserve begins to decrease, and the degree of its decrease depends on the severity of the hypoxic state [8,11]. The authors found in the terminal stage of hypoxic hypoxia a decrease in the alkaline reserve to 8.8 vol%. The accumulation of carbon dioxide leads to the expansion of arterioles and capillaries, however, Shvets D.A. [9], express doubts that carbon dioxide really increases blood flow, since, in this case, respiration is stimulated.

Studies to study the effect of hypoxia on the cardiovascular system were undertaken with the aim of accumulating factual material and elucidating the mechanisms of hemodynamic reactions. The main results of these studies can be summarized as follows. A short stay at high altitude is accompanied by an increase in heart rate and slight changes in systolic blood pressure. Enhanced blood circulation due to increased heart rate begins at an altitude of 2000 m, slowly increases to 7000 m, and then develops faster, and it is usually pronounced in untrained individuals, while in trained individuals, adaptation is carried out mainly due to an increase in the stroke volume of the heart. At an altitude of about 5000 m and above, disorganization in the activity of the

cardiovascular system is observed, the strength of the heart contraction decreases, but the increase in the heart rate can continue. With the ineffectiveness of compensatory reactions providing an increase in the activity of the circulatory apparatus, the redistribution of blood and the centralization of blood circulation, i.e. maintaining normal blood flow in vital organs such as the brain and heart [4,10].

The Purpose of the Study: The aim of this work was to assess the relationship between vascular liver lesions and changes in the rheological properties of blood in experimental hypoxic conditions.

Materials and Methods of Research: The experiments were carried out on 50 white rats of a mixed population with an initial weight of 150-220 grams, kept in vivarium conditions on a normal laboratory diet.

Hypoxic hypoxia was induced by placing rats in a pressure chamber SPT-200 Vaccum-DRIER. The animals were kept in a pressure chamber at an altitude of 9000 m for 3 hours.

The studies were carried out at 1, 3, 24, 96 hours after the reproduction of hypoxia.

Study of the Viscosity And Shear Rate Of Blood: One of the main indicators of the rheological properties of blood, viscosity (or fluidity) was determined by the Copeley method modified by V.M. Udovichenko. For this purpose, a system consisting of a measuring capillary preostat and a thermostatic installation was assembled at the Department of Basic Medical Disciplines of the Fergana branch of the Tashkent Medical Academy. Viscosity indices were determined in the following shear stresses 2, 4, 8, 12, 16 mm of water. Art. The results were calculated using the following formula:

$$Z = \frac{100\ gz}{8Я.1.L}\ P.t\ (cP)$$

where, $Я$ is the radius of the capillary in the wide part.

z - radius of the narrow part.

L - wide part length.

t - blood flow time.

P - pressure supplied from the capillaries from the preosstat.

g - acceleration of gravity equal to 980 cm/s

The shear rate of blood at a given viscosity was also calculated using the following formula:

$$V = \frac{4R2L}{z\ t}\ sec\text{-}1$$

Based on the obtained values, a dynamic viscosity curve was constructed.

Intravital biomicroscopy was performed under general anesthesia (the animals were injected intraperitoneally with Na etaminal at a dose of 8 mg / 100 g of body weight).

In order to optimize the study and improve the quality of the results obtained, we used an interactive system for monitor digital analysis of microcirculation parameters with computer data processing. The system consists of a luminescent microscope "Lumam IZ", a monitor camera, a monitor capillaroscope, a digital memory device with an adjustable recording interval, a video monitoring device and a Pentium-IV type personal computer.

Biomicroscopy was performed with contact objectives 10x0.40 and 25x0.40.

Such parameters of microcirculation as the diameter of microvessels and the linear velocity of blood flow were assessed.

The digital data were processed by the method of variation statistics. Numerical differences were considered significant under the condition when $t \geq 2$, and $P < 0.05$.

RESULTS AND ITS DISCUSSION: Metabolic processes in internal organs depend on the preservation of the structure and function of erythrocytes, the most important of which is the transport of oxygen to organs and tissues.

The blood viscosity in animals was determined after 1, 3, 24, 96 hours, as well as at the pressure values applied to the blood flow: 2,4,8,12,16 mm. water Art. The results of changes in blood viscosity in white rats during hypoxic hypoxia, depending on the duration of the experiment and blood pressure, are shown in Table 1.

In the intact group of animals, it was found that the viscosity of blood at a pressure of 2 mm cP. is 5.0 ± 0.42 cP, and at 16 mm cP. 3.81 ± 0.26 cP. In experimental animals, in the dynamics of the development of a hypoxic state, there is a significant increase in blood viscosity and a significant decrease in the shear rate during the study period at various values of hydrostatic pressure.

TABLE 1 INDICATORS OF BLOOD VISCOSITY AND SHEAR RATE DEPENDING ON THE PERIOD DURING HYPOXIC HYPOXIA.

Study groups	2 mm water column		16 mm water column	
	Viscosity	shear rate	viscosity	shear rate
Intact	5,0±0,42	7,63±0,08	3,8±0,25	76,5±0,27
1 hour	6,2±0,13*	4,55±0,18*	4,3±0,14*	73,6±2,9
3 hours	9,3±0,26*	3,97±0,13*	6,5±0,27*	50,7±2,2*
24 hours	7,5±0,16*	6,84±0,18*	5,1±0,12*	69,4±1,2*
96 hours	5,8±0,31	7,35±0,35	4,2±0,37	75,6±2,8

Note: * - The results are reliable in relation to intact gr. ($P < 0.05$)

With hypoxic hypoxia, the blood viscosity indices after 1 hour at the minimum pressure increased by 24%, and at the maximum one exceed the value of intact animals by 13.1%.

It was found that after 3 hours after hypoxic exposure, the severity of these processes increases and the studied parameters at minimum and maximum applied pressures increase by 86 and

71.5% compared to the intact group, and by 50 and 51.1% compared to the previous period, respectively.

After 24 hours, the severity of changes in comparison with the previous period decreases somewhat: the increase in viscosity at the minimum and maximum applied pressures is 19.4 and 21.6%, but at the same time, in relation to the intact group of animals at the minimum pressure, the blood viscosity remains high 50 %, and at the maximum by 34.2%.

In the subsequent period - after 96 hours, the tendency to a decrease in viscosity at the minimum and maximum applied pressures continues. So, in relation to the intact group of animals, at the minimum pressure, the blood viscosity remains increased by 16% and at the maximum pressure by 10.5%, which indicates the preservation of blood hypercoagulation.

The blood shift rate in the intact group of animals at the minimum pressure was 7.65 ± 0.08 C^{-1}, and at the maximum pressure 76.5 ± 0.27 C^{-1}.

It is noted that already 1 hour after hypoxic exposure at a minimum pressure, the blood shear rate decreases by 32.5%, and at a maximum pressure by 35.7%. 3 hours after the experiment, the severity of these changes increases with the minimum and maximum applied pressures; by 48.0 and 43.8% in relation to the intact group of animals. One day after hypoxic exposure, an increase in the shear rate is observed - at a minimum pressure compared to the previous period by 72.2%, and at a maximum pressure by 36.8%, respectively. 96 hours after the hypoxic state, the shear rate at minimum and maximum pressures continues to increase in comparison with the previous period and almost returns to the initial level.

Thus, the results obtained indicate that in hypobaric hypoxia, the most pronounced changes are observed after 3 hours, and by the end of the experiment they tend to recover.

The viscosity of the blood at the minimum pressure was found to be higher than the values at the maximum pressure and high shear rates.

A decrease in the shear rate of blood and an increase in its viscosity will certainly affect the state of the vessels of various organs, the functional failure of which can lead to stagnation of blood and the development of organ hypoxia. Taking this into account, we subsequently investigated the state of microcirculation of internal organs.

The State Of Liver Microcirculation In Experimental Hypoxic Hypoxia: The most important physiological parameter of microcirculation is the blood flow velocity in microvessels and their diameter, which determine, in particular, the conditions for oxygen transport to the liver tissues [3,6,8].

Proceeding from this, the task of our work was to study possible disorders of the microcirculatory bed of the liver, determine its static and dynamic parameters, fluctuations in these values in the norm and their changes in some forms of experimental hypoxia.

When observing the surface layer of the liver of intact rats using a contact lens, it was found that at a depth of 20-30 microns there are a large number of hepatic venules with sinusoids flowing into them. Portal terminals, from which the sinusoids originate, are located at great depths and therefore were available to our observation much less frequently. They had a length of about 200-400 μm and a diameter of 9.5 ± 1 μm on average. The diameter of the portal venules was

32.1 ± 0.4 μm, the blood flow velocity was 0.345 ± 0.01 mm/sec. The diameter of the sinusoids was 9.5 ± 0.2 μm, with a blood flow rate of 0.290 ± 0.002 mm / sec. In the central collecting venule, the diameter was 45.1 ± 0.8 μm, and the blood flow velocity was 0.206 ± 0.002 mm / sec. In the sinusoids in the center of the lobules, the blood flow velocity is faster than in the sinusoids located to the periphery. The results of morphometric studies are shown in Table 2.

As can be seen from the table, 1 hour after hypoxic hypoxia, the diameter of the portal venules increased by 23.6%, sinusoids by 50.5%, central collecting venules by 13.7% and a slowdown in the blood flow velocity in them by 43.2, 36.3 and 26.9% compared to the intact group. In the microcirculatory bed of the liver, there is a decrease in the number of functioning portal, collecting venules and sinusoids. In the vessels of the liver, empty sinusoids alternate with dilated ones, filled with blood. There was a significant decrease in the rate of hepatic blood flow. Hepatic venules became full-blooded, somewhat dilated, with areas of microaneurysms.

TABLE 2 MORPHOMETRIC ANALYSES OF THE VESSELS OF THE MICROVASCULATURE OF THE LIVER DURING HYPOXIC HYPOXIA

Research objects	Portal venules		Sinusoids		Central collecting venules	
Timing of the experiment	diameter (μm)	blood flow velocity (mm/s)	diameter (μm)	blood flow velocity (mm/s)	diameter (μm)	blood flow velocity (mm/s)
Intact	32,1±1,16	0,345±0,002	9,5±0,18	0,290±0,002	45,1±0,59	0,206±0,002
1 hour	39,7±0,23 *	0,196±0,008 *	14,3±0,09*	0,185±0,002 *	51,3±1,95*	0,130±0,001 *
3 hours	45,3±2,25 *	0,114±0,004 *	18,5±0,94*	0,136±0,001 *	57,2±1,36*	0,104±0,003 *
24 hours	36,4±1,43 *	0,245±0,01*	12,7±1,23*	0,213±0,001 *	48,1±1,82	0,168±0,002 *
96 hours	33,5±1,42	0,278±0,002 *	10,8±0,064	0,256±0,003 *	46,7±0,89	0,186±0,003

Note: * - The results are reliable in relation to intact gr. (P <0.05)

An hour after the experiment, microcirculatory disorders in the liver are aggravated. There is a statistically significant increase in the diameter of the portal venules, sinusoids and central collecting venules by 41.1, 94.7 and 26.8%, with a slowdown in the blood flow velocity in them by 67, 53.2 and 49.6% compared to the intact group of animals , in some areas there is a presinusoidal edema, especially in the center of the lobules, and along the periphery of the lobules there is an increase in the number of "plasma" sinusoids. The tendency to aggregation of blood corpuscles is intensified and areas of the so-called "dumb zones" appear in which signs of diapedesis are noted. In this case, the vessels are sharply dilated, tortuous, filled with aggregates of blood corpuscles. And the blood flow is sharply slowed down, in some places even completely stopped (stasis). Arteriolovenular anastomoses function, shunt blood flow is observed.

After 24 hours of the experiment, plethora and expansion of the sinusoids are still observed. Thus, the diameter of the portal venules, sinusoids and central collecting venules was significantly increased by 13.3, 33.6, and 6.6% in comparison with the intact group, and the blood flow velocity in them decreased by 29, 26.6 and 18.5%, respectively. In this period, areas of petechial hemorrhages are noted in the liver parenchyma, at the same time there is a separation of plasma in the capillaries and the functioning of arterio-venular anastomoses. In all visible terminal hepatic venules and sinusoids, the blood flow is slowed down, intermittent, plasma gaps are clearly traced, which indicates a pronounced aggregation of blood corpuscles, which are caused by a change in the aggregation of erythrocytes and the rheological properties of blood.

We have found that by 96 hours of the experiment in rats with hypoxic hypoxia, the above-described mosaicity of changes is somewhat preserved in the microcirculatory picture, but in some areas there is a tendency to recovery. This is confirmed by changes in morphometric data. Thus, the diameter of the portal venules, sinusoids and central collecting venules, in comparison with the previous period, decreased by 8.15 and 3%, and the blood flow velocity increased by 13.4, 20.1 and 10.7%. But, at the same time, in relation to the intact groups, the diameter increased by 4.3, 13.6 and 3.5%, and the blood flow velocity decreased by 19.5, 11.8 and 9.8%, respectively. The number of functioning sinusoids is noticeably increased mainly along the periphery of the lobules, mainly due to the inclusion of previously non-functioning sinusoids. The perisinusoidal edema is somewhat reduced compared to the previous period, but the vessel walls remain blurred.

It is known that vasoactive substances that come from foci of tissue hypoxia and affect microcirculation play an important role in the development of primary microcirculatory and hemorheological disorders. Experiments have shown that the linear velocity of blood flow in the vessels of the liver after 3 hours with hypoxic hypoxia decreases by more than 2 times. In venules, due to pronounced rheological disorders, the blood flow rate decreases, vasodilatation is observed. Aggregates of shaped elements begin to appear, while there is a decrease in the speed of blood flow. Plasma separation from erythrocytes, multiple micro thrombosis, stasis phenomena in venules, capillaries and sinusoids lead to the development of tissue hypoxia.

CONCLUSIONS

1. Acute hypoxic hypoxia is accompanied by pronounced disturbances in the rheological properties of blood.

2. Disorders in the liver microcirculation during hypoxic hypoxia are unidirectional and depend on its genesis: at 98 hours of hypoxic hypoxia, normalization of the diameters of the liver vessels is observed, against the background of a reduced blood flow rate in them.

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