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EFFECT OF OXIDATIVE ACTION OF POTASSIUM HEXACYANOPHERATE (III) ON HEMOGLOBIN

Introduction. Hemoglobins are structurally related proteins in the erythrocytes of vertebrates. They perform two important biological functions: they transfer O2 from the lungs to peripheral tissues and CO2 and protons from peripheral tissues to the respiratory organs for further excretion from the body [1: 55]. The interaction of hemoglobin (Hb) with the oxygen molecule is a complex process that includes changing the spin state of the iron ion, increasing the size of the porphyrin cycle nucleus, changing the tertiary structure of the corresponding subunit, rearranging the quaternary structure of the whole Hb molecule. Under certain conditions, equilibrium oxygen binding can switch to an irreversible autooxidation reaction to methemoglobin (metHb). This process is the main source of oxygen free radicals in the erythrocyte, namely superoxide radicals ($O_2^{-}\cdot$) [2: 42-48].

Despite the fact that hemoglobin has been studied quite well, there are still many unresolved issues related to its functioning. Some of them are of interest for general and theoretical biology; others are relevant for practical medicine. The first group includes the study of molecular mechanisms of signal transmission from hemoglobin to other components of the cell, which is closely related to the study of mechanisms of erythrocyte adaptation, damage and resistance, the second – the development of erythrocyte stabilization methods and the use of data on various functional forms of hemoglobin [3: 3-23].

Review of recent publications. The mechanism of hemoglobin autooxidation is well studied. It is shown that the rate of autooxidation of Hb under the action of potassium hexacyanoferrate (III) depends on the state of hemoglobin. It is known that organic phosphates (ATR, bisphosphoglycerate, but not AMR), stabilizing the T-conformation of the protein molecule, increased the rate of oxidation of Hb by ferricyanide, while the modification of sulfhydryl groups Cys-93 - probably reduced by suppressing the electron transfer of ferricyanide molecule. The resistance of oxyhemoglobin (oxyHb) to the action of chemical oxidants reflects the reactivity of heme, which may be associated with a particular conformation of the protein molecule, the availability of heme pockets to the solvent [2: 42-48].

The peripheral blood of almost healthy donors of the same sex and approximately the same age was used in the experiments. Erythrocytes were washed three times by centrifugation with Na-phosphate buffer (0.015 mol, pH 7.4) containing 0.15 mol NaCl. Plasma-washed and packed erythrocytes were resuspended in the same buffer containing different glucose. The amount of glucose was administered according to the number of packed erythrocytes in the suspension. The cells were incubated for 5 hours at 20 ° C. After certain time intervals, the sample was washed by centrifugation with Na-phosphate buffer (pH 7.4) and subjected to lysis. The hemolysate was used to determine the rate of autooxidation of hemoglobin in the sample. The rate of autooxidation was determined by automatically recording the change in optical density of the sample at 540 nm after adding potassium ferricyanide to the hemoglobin sample. According to the obtained kinetic dependences, the reaction rate constant was determined [4].

Based on information about the functioning of Hb, we can say that hemoglobin is able to accumulate information about the state of erythrocytes and blood plasma, functioning as a signaling molecule. Signal-regulatory functions of Hb are determined by its conformational transitions associated with the cycle of oxygenation-deoxygenation, as well as changes in the valence state of iron. Therefore, the additional functions of hemoglobin are clearly manifested in conditions of hypoxia and / or oxidative stress, when there is a conformational R-T transition, and the proportion of its oxidized non-physiological forms increases [3: 3-23].

The objective of the paper is to use the Hb autooxidation reaction under the action of potassium hexacyanoferrate (III) as a rapid test to determine the state of the cytoplasmic and membrane-bound fraction of hemoglobin in human erythrocytes, the level of its oxidative modification.

Results of the research. The presence of an external oxidizing environment and glucose changes the tendency of hemoglobin to auto-oxidation.

Conclusion. The data obtained in this work indicate that the rate of oxidation of hemoglobin by erythrocytes by ferricyanide can be used as an indicator of the propensity of hemoglobin to autooxidation. The rate constant of hemoglobin oxidation by ferricyanide can be used as a stand-alone test for hemoglobin.

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DEVELOPMENT OF THE PROGRAM "DEPARTMENT"

Introduction. The process of solving any complex problem can be divided into three interrelated global stages, which are shown in figure 1 [1].

First, as we see, we must build a model of our system, or in other words, formalize the problem.

Once the model is built, the stage of developing the solution algorithm begins, and then we have the stage of writing the program. The algorithm allows you to implement the model on a computer, and the program in the modern sense is interpreted, of course, much wider than just the program implementation of the described algorithm. It is a complex software package consisting of many related components (such as a software kernel, database, graphical interface, etc.), and is