HIGH TENSION ELECTRIC FIELDS AS THE FACTOR OF OXIDATIVE-DESTRUCTIVE INFLUENCE ON ERYTHROCYTES AT LOW ANTIOXIDATIVE STATUS

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ABSTRACT

High tension electric fields (EF) represent a physical environmental factor influencing on oxidative-destructive processes' rate in human organism. Resistance to this EF depends mainly on multicomponent system of natural antioxidants. Among trace elements selenium is of great importance. However the mechanism of its antioxidative action has not been investigated completely. Creating selenium-deficit conditions is essential for study of the action mechanism. Hence, both the erythrocytes of pregnant women and the erythrocytes at glucose-6-phosphatedehydrogenase (G-6-PD) enzyme deficiency are appropriate natural models. The results of investigations have shown that the erythrocytes of pregnant women (initial terms of pregnancy) proved to be more resistant to EF effect than the erythrocytes with G-6-PD pathology. Due to the intensive consumption of Se at late stages of pregnancy, erythrocytes of pregnant women are more labile to oxidation than the G-6-PD deficiency erythrocytes.

Keywords: hemoglobin, methemoglobin, hemolysis, lipid peroxidation, antioxidants, selenium, pregnancy, glucose-6-phosphatedehydrogenase, high tension electric fields

I. INTRODUCTION

Among the environmental physical factors affecting people health, electromagnetic fields of industrial frequency take an important place. Like the other environmental factors, EF can be an initial reason of some pathologies (or can complicate the course of already expressed diseases) associated with oxidation process acceleration [1, 2].

Natural resistance of organism to adverse factors, first of all to oxidative stress, is adjusted by the hemostasis system, which includes antioxidative (AO) protective complex. Study of natural antioxidant metabolism and oxidation promotors under the physical and chemical environmental factors influence is necessary for understanding of the primary mechanisms of the damage of bio-objects that, in any case, are connected with population health. One of the vital biological molecules is hemoglobin, of which oxidation condition is extremely important for the development of free radical oxidation of cellular structures.

A special interest represents the action of adverse environmental factors on people health at weak AO system. In particular, deficiency of glucose-6-phosphatedehydrogenase (G-6-PD) enzyme, which is frequently associated with the reduction of glutathione peroxidase (GP) enzyme activity [3]. The gene of (G-6-PD) enzyme deficit is widely distributed among the Azerbaijan population [4]. Besides, as a natural model of AO deficiency, the erythrocytes of pregnant women is of special interest (especially in the second half of pregnancy) [5].

Selenium and related AO proteins (GP, R-protein, some cellular intermediates, etc.) are the most important components of natural AO system. It is necessary to note, that the territory of our republic is a region characterized with low and average selenium supply that raises the urgency of study of the pathologies related to selenium deficiency and activation of oxidative-destructive processes [6].

Considering all the above mentioned, we have undertaken a study of oxidation process development induced by industrial frequency high tension EF in erythrocytes at glucose-6-phosphatedehydrogenase (G-6-PD) enzyme deficiency and erythrocytes of late stages of pregnancy with selenium deficit symptoms.

II. MATERIALS AND METHODS

1. For the creation of high tension EF an installation was assembled using cylindrical cored cables. The scheme of installation is presented in figure 1.

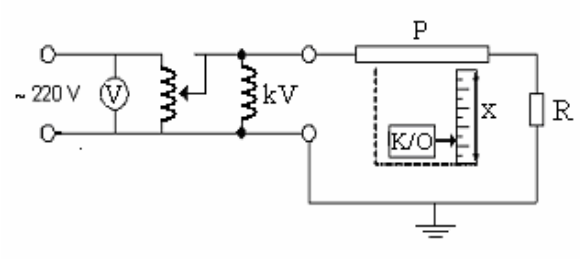

Fig. 1. (K/O – the cell, P - cored cylindrical cable)

The tension of electric field was calculated according to the equation below:

$$E_x = U_o/[x*\ln 2H/r_o],\ (r_o \ll x)$$

U_o - an initial tension (voltage) of conductor
x - distance from conductor to object
H - distance from conductor to "0" potential
r_o - radius of conductor

The tension of electric field affecting the objects changes depending on the distance according to the above formula.

2. Erythrocytes of persons, the carriers of the gene of (G-6-PD) enzyme deficiency and erythrocytes of the women at normal proceeding pregnancy in various stages served as materials of research. Erythrocytes were extracted by standard method: centrifugalized, washed three times and diluted by the phosphate buffer (0,1 M, pH 7,4) 1:50 (hematocryte ≈ 1), erythrocyte shadows were obtained by Dodge's method [7]. Suspensions of erythrocytes were placed in glass flasks at 25°C temperature and exposed to electric field, 50-150 κV/m. Aliquots were taken in each 3 hours. The condition of hemoglobin oxidation was determined in suspension and buffer solution. The rate of oxidative hemolysis (λ_{670}) was studied in supernatant by spectrophotometric method. The rate of oxidative-destructive processes was evaluated on the accumulation of products reacting with thiobarbituric acid (TBA) in suspension and in erythrocyte shadows [8]. The hemoglobin oxidation conditions was determined by the following formulas [9]:

$$[HbO_2] = 29{,}8A_{577} - 9{,}8A_{630} - 22{,}2A_{560}$$
$$[MetHb] = 7{,}0A_{577} + 76{,}8A_{630} - 13{,}8A_{560}$$
$$[Hi] = -33{,}2A_{577} - 36{,}0A_{630} + 58{,}2A_{560}$$

A_{577}, A_{560}, A_{630} extinction of samples on corresponding wave lengths.

Statistical processing of measurement results was carried out using Student's criterion [10].

III. RESULTS AND DISCUSSION

It is well known that increased tendency to oxidative hemolysis is one of the consequences of G-6-PD deficiency [11]. In many respects, it is associated with the fact that selenium containing glutathione peroxidase (GP) connected with G-6-PD in pentosephosphate shunt is also low active. As a result, hemolysis is accelerated during the promotor action of various oxidizers including medicals such as hydrazine and primaxine, and also at the use of beans in meals (*Vicia Fava*) [11, 12]. From modern point of view, it is associated with the fact that beans contain high concentration of lipoxigenase enzyme, accelerating lipid peroxidation (LPO) membranes of erythrocytes that causes hemolysis under low concentration of a natural antioxidant - selenium and low GP activity [13].

Selenium plays an important role during reproductive period. Due to double consumption, the demand for selenium in pregnant women considerably increases. Additional content of selenium in the diet of pregnant women will help prevent progress of some diseases [14]. However, the necessary and safe level of selenium receipt in the organism of pregnant woman, which is based on the regional features of selenium supply, is not established up to now.

As can be seen from fig. 2, there are threshold values of EF intensity (at fixed 5 hour expositions), under which marked acceleration of oxidative processes is observed. The threshold for control erythrocytes is ≈ 83 κV/m, for initial stages of pregnancy E ≈ 74 κV/m, for G-6-PD deficiency E ≈ 67 κV/m and for late stages of pregnancy E ≈ 55 κV/m. Under the influence of high tension EF in erythrocytes at G-6-PD pathology the accumulation of TBA of active products proceeds faster than in erythrocytes of pregnant women (the first trimester). But at the late stages of pregnancy in conditions of selenium deficiency the accumulation of TBA of active products proceeds faster than in erythrocytes at G-6-PD pathology.

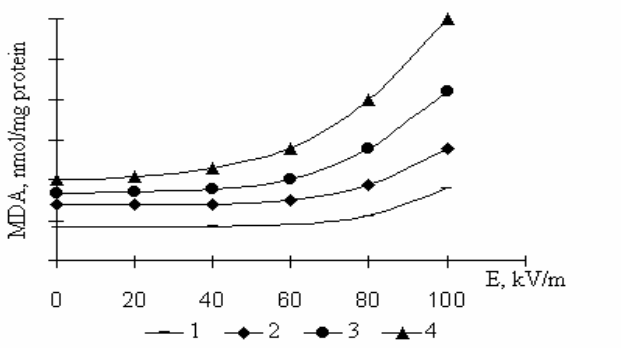

Fig 2 Kinetics of TBA-active products (MDA) accumulation in erythrocytes under electrical field effect
1 – Control, 2 – Initial stage of pregnancy, 3 – erythrocytes with G-6-PD deficiency, 4 – Late stage of pregnancy

Erythrocytes contain at least two significant "targets" for oxidative influence: hemoglobin itself (MetHb and other oxidative forms of hemoglobin) and membrane structures (LPO). These two oxidative processes are interconnected. However, their sequence is unknown. Therefore, kinetics of these two processes are considered simultaneously. The curves of methemoglobin accumulation and hemolysis under electric field effect are presented in fig. 4. As can be seen from the figure, the curve of MetHb accumulation has more acute shape that may indicate that oxidative degradation of erycthrocytes is induced by hemoglobin oxidation. Interestingly, erythrocytes of the women at late stages of pregnancy show greatest tendency to oxidation (both hemoglobin and membrane structures).

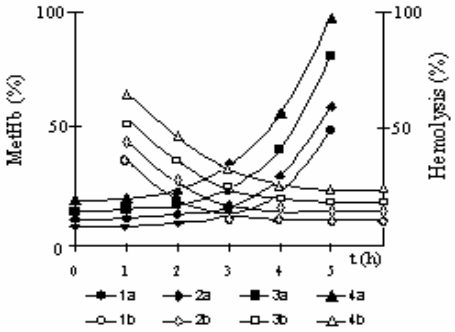

Fig.4 Accumulation MetHb and hemolysis in erythrocytes under electrical field effect
1 – Control, 2 – Initial stage of pregnancy, 3 – erythrocytes with G-6-PD deficiency, 4 – Late stage of pregnancy

Comparative examination of the kinetics of accumulation of active products' TBA in suspension and in erythrocyte shadows (fig. 5, 6) shows significant difference both in initial level of LPO and in oxidative process intensity. It is explained by the presence of antioxidative factors in cytosole of erythrocytes (GP, superoxidedismutase, catalase, etc.). It should be noted that in erythrocyte shadows oxidative process proceeds more faster in late terms of pregnancy that indicates considerable reduction of antioxidants.

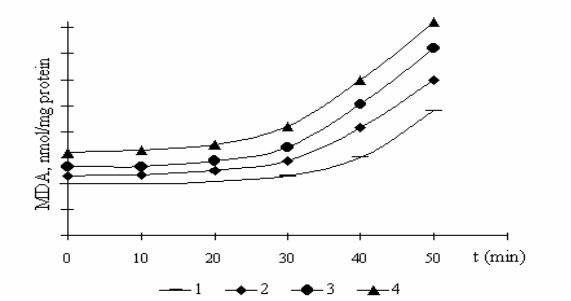

Fig.3 Kinetics of TBA-active products (MDA) accumulation in erythrocytes under electrical field effect (E = 100 kV/m, during 5 hours)
1 – Control, 2 – Initial stage of pregnancy, 3 – erythrocytes with G-6-FD deficiency, 4 – Late stage of pregnancy

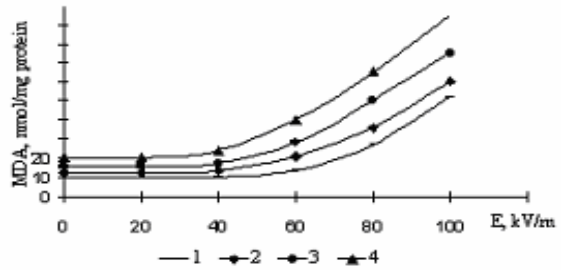

Fig.5 Kinetics of TBA-active products (MDA) accumulation in shadows of erythrocytes under electrical field effect
1 – Control, 2 – Initial stage of pregnancy, 3 – erythrocytes with G-6-PD deficiency, 4 – Late stage of pregnancy

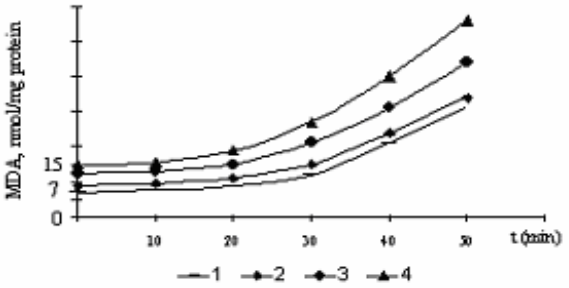

Fig.6 Kinetics of TBA-active products (MDA) accumulation in shadows of erythrocytes under electrical field effect
1 – Control, 2 – Initial stage of pregnancy, 3 – erythrocytes with G-6-PD deficiency, 4 – Late stage of pregnancy

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