Armenian Journal of Physics, 2016, vol. 9, issue 1, pp. 76-79

Chemiluminescence of Rats' Whole Blood after X-ray and γ-irradiation

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Received 15 January 2016

Abstract: The effects of X-ray and γ-radiations on H_2O_2-chemiluminescence of rats' whole blood were investigated. It was found out that when cells and biomaterials were exposed by X-ray radiation an intensification of oxidative processes took place leading to the free radical reactions increase, which is reflected in rise of the level of chemiluminescence (CL). However, by γ-irradiation in small doses the level of CL is slightly increases and at a relatively high dose intensity of H_2O_2-chemiluminescence decreases, which in our opinion is due to degradation processes, which takes place in biomaterials.

Keywords: X-ray, γ-radiations, H_2O_2-chemiluminescence, rats' blood, biomaterials

Introduction

At present, from the viewpoint of importance of free-radical oxidation processes in biomedical research, that issue was studied by many authors in a variety of biological materials in the presence of various pathological processes, chemical and physical impacts, including ionizing radiation [1, 5, 10].

Under the influence of ionizing radiation, the substances with high chemical activity are formed in organism such as free atoms or radicals, which destroy the organism cells. Ionizing radiation can also have a direct impact on molecules [3, 8, 9, 11].

At this stage, to address these issues, the method of chemiluminescence (CL) found widespread application in research laboratories and clinical practice both for study the pathological processes and to create biophysical prognostic and additional diagnostic tests. The dependence of the CL intensity in biological fluids, including blood components under influence of various physical and chemical factors were established [2, 4, 6].

In those studies appropriateness of the use of CL data for studying the mechanisms of these effects both on the whole organism and in its separate parts was shown. It was clearly shown that CL of biological objects was caused by free radical (FR) processes and by peroxidation of the lipids (LPO) and lipid containing structures [2, 6, 7].

The purpose of this study was to investigate the chemiluminescence of rats' whole blood after X-ray and γ-ray irradiations.

Material and methods

The materials for the study were samples of rats' whole blood after X-ray irradiation at different doses. X-ray irradiation was carried out in a glass test tube in X-ray device.

CL registration was carried out in quantum metric device, which was assembled by using a new developed electronic and amplifier units. As a detector of weak light fluxes, a highly sensitive low-noise photoelectronic multiplier (PEM-140) was used operating in the regime of constant cooling with tap water in order to reduce the level of thermal noises of the photomultiplier's photocathode. Experiments were performed at room temperature. Since the whole blood does not have its own chemiluminescence because of a weak signal, which, by the way, is absorbed by the red medium, the method of CL activation by hydrogen peroxide was used. In this case, CL was due to the interaction of the test sample with H_2O_2. Upon decomposition of hydrogen, the peroxide $^{\bullet}OH$ radical is generated, which gives rise to a chain reaction accompanied by emission of light quanta.

In order to study the H_2O_2 - CL in 2.7ml blood samples, 0.3ml of 3% hydrogen peroxide was added and placed for 3-5 seconds in an optical cell in front of photomultiplier cathode and was recorded by computer with NATIONAL INSTRUMENTS LabVIEW software [12].

Results and discussion

The results of experiments on the effect of X-ray irradiation of blood are given in Fig. 1, 2.

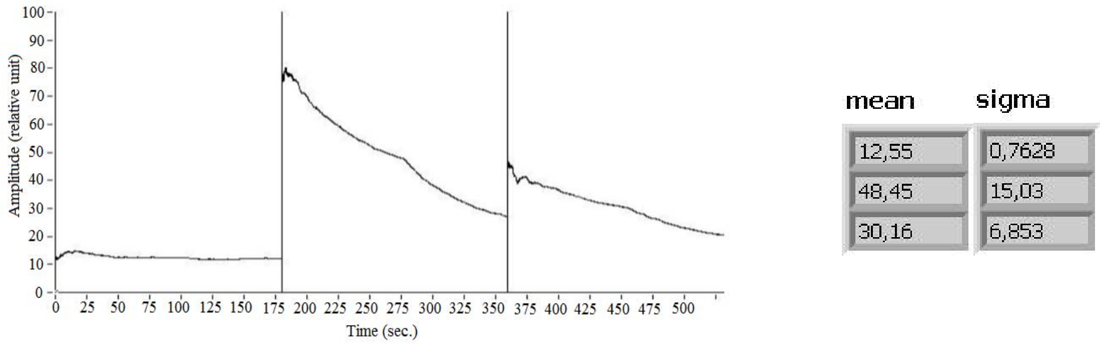


Fig. 1. H_2O_2-CL irradiated for 5 minutes. X-ray of rats' whole blood: 1 – background of quantum metric device, 2 – irradiated blood sample, 3 – non-irradiated blood sample.

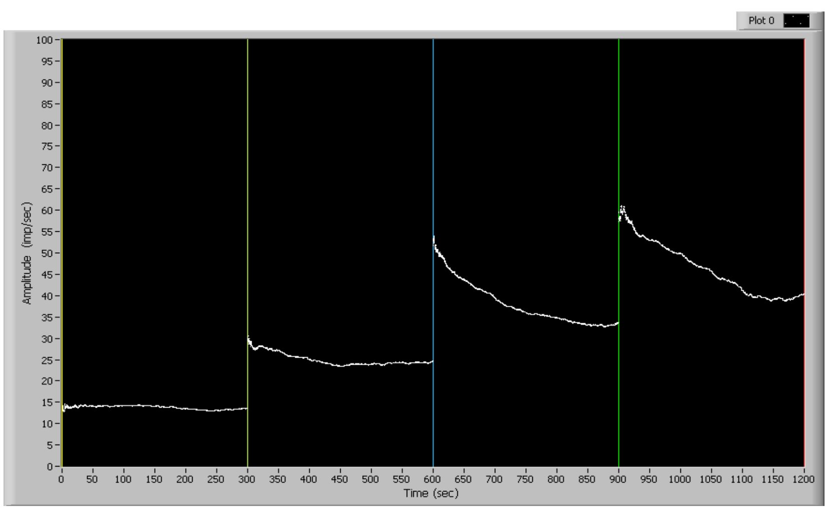


Fig. 2. H_2O_2-CL of rats' whole blood irradiated by X-ray: 1 – background quantum metric device, 2 – non-irradiated blood sample, 3 – blood sample irradiated for 5 minutes, 4 – blood sample irradiated for 20 minutes.

With the purpose to clarify the question in what biological structures the basic processes associated with radiation exposure took place, in the following experiments peroxidation of rats' whole blood lipids (LPO) and rats' blood plasma was investigated (Table 1).

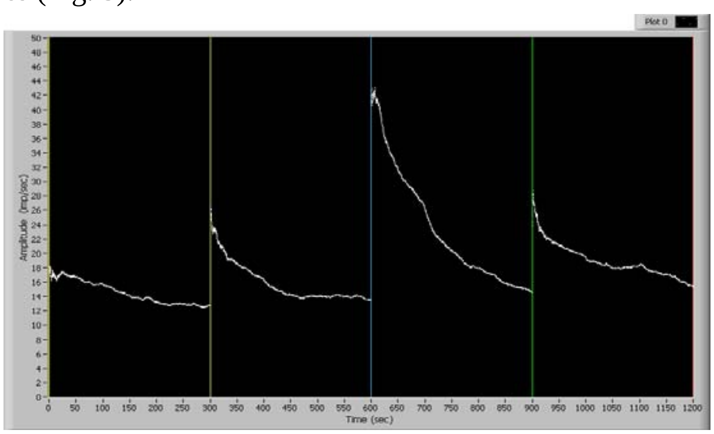
Table 1. LPO of the whole blood and blood plasma of irradiated and non-irradiated rats.

Non-irradiated whole blood samples (control) The average values	0.43, 0.38, 0.42 0.041 ± 0.022
Irradiated samples of the whole blood The average values	0.105, 0.103, 0.104 0,104 ± 0.02
Non-irradiated samples of blood plasma The average values	0.156, 0.149, 0.152 0,179 ± 0.021
Irradiated samples of whole blood plasma The average values	0.177, 0.181, 0.179 0.179 ± 0.02

The Table above shows that at irradiation, LPO increase is observed both in the whole blood and in plasma. This indicates that irradiation mainly acts on the membrane and the lipid containing structures inducing in them free radical oxidation reaction accompanied by the chemiluminescence.

By analyzing the above data on X-ray exposure of rat's whole blood it can be certainly established that depending on the irradiation exposure (dose dependency), an intensification of the H_2O_2-chemiluminescence takes place. Intensification of H_2O_2-CL is due to the oxidation of the blood components, mainly containing lipid membrane structures. At interaction of the oxidized products with H_2O_2 the free radical chain processes are generated accompanied by light quanta release. H_2O_2-CL intensity increases with increasing doses of X-ray irradiation.

However, under the influence of hard γ- radiation an opposite picture is observed: after irradiation of rats' whole blood with relatively low doses, intensity of H_2O_2-CL increases (at 5 min of exposure), while at increasing of γ- irradiation dose (at 20 min of exposure) it significantly decreases (Fig. 3).



1 2 3 4

Fig. 3. H_2O_2-CL of γ- irradiated rats' whole blood: 1 – background of quantum metric device, 2 – non-irradiated blood sample, 3 – blood sample irradiated for 5 minutes, 4 – blood sample irradiated for 20 minutes.

Based on these data, we can assume that (i) at hard γ- irradiation in small doses a partial destruction of the biological material takes place, in which the mechanism is triggered of lipids peroxidation of the membrane structures followed by the increase in chemiluminescence; and (ii) with increasing doses of γ- radiation, probably, the processes of degradation of biomolecules and destruction of peroxide radicals begin that result in decrease in the levels of chemiluminescence.

Thus, analyzing the data obtained it can be concluded that under the action of X-ray radiation on cells and biomaterials, intensification of oxidative processes takes place leading to the increase in free radical reactions, which is reflected in increasing the H_2O_2 - chemiluminescence. However, at γ- irradiation in small doses, the level of chemiluminescence slightly increases, and at a relatively high dose the H_2O_2-CL intensity decreases, which in our opinion is due to the degradation processes taking place in the biomaterial.

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