International Journal of Research Studies in Medical and Health Sciences
Volume 3, Issue 11, 2018, PP 01-09
ISSN : 2456-6373

The Stimulating Action of Low-Dose Blue Light Irradiation on Functional Activity of Retinal Pigment Epithelium Cells

Natalia Seryoznikova[1,2], Natalia Sakina[1], Pavel Zak[1], Alexander Dontsov[1]

[1]*Department of Photochemistry and Photobiology, Emanuel Institute of Biochemical Physics, Russian Academy of Sciences, Moscow, Russia*
[2]*Department of Biology, Lomonosov Moscow State University, Moscow, Russia*

***Corresponding Author:** Natalia Sakina, PhD, Senior Researcher, Department of Photochemistry and Photobiology, Emanuel Institute of Biochemical Physics, Russian Academy of Sciences, Moscow, Russia.*

ABSTRACT

***Background:** The Japanese quail, Coturnix japonica has been shown to be an efficient animal model for studying the retinal disorders. The retinal pigment epithelium (RPE) cells of this animal model and human monocyte cells were used to determine the conditions under which irradiation of cells in the blue spectral range not only does not lead to their death but has a stimulating effect on metabolic activity.*

***Methods:** A study of the effect of low-dose blue LED irradiation with a maximum of 450 nm and doses of ≤1 J/cm^2 was carried out on the models of RPE cells of the eye of the Japanese quail, Coturnix japonica and the human monocyte cell line THP-1. Metabolic activity of cells (resazurin test), their antioxidant status (inhibition of the process of chemiluminescence of luminol induced by hydrogen peroxide) and the value of the mitochondrial membrane potential (TMRE test) were registered as measurable parameters.*

***Results:** It was shown that irradiation with blue light at 450 nm in low doses causes an increase in the metabolic and antioxidant activity of RPE cells. It was established that low-dose irradiation with blue light, also leads to an increase in the mitochondrial membrane potential of RPE cells and human monocytes. At the same time, in the RPE cells of the Japanese quail, an increase in the number and specific volume of mitochondria is observed.*

***Conclusion:** The results of the study on RPE cells and human monocytes indicate that irradiation with blue light at 450 nm in doses less than 1 J/cm^2 not only does not cause damage but rather has beneficial effects on cells, as it was shown for irradiation by LED and laser sources in the red spectral range.*

Keywords: *Low dose blue light; Photobiomodulation; Retinal Pigment Epithelium; Mitochondrial membrane potential; Metabolic activity; Antioxidant activity; Japanese Quail; Human monocytes*

INTRODUCTION

The treatment with light or light therapy with laser or LED sources at low irradiation intensities is associated with the so-called photobiomodulation process, that is, the beneficial effect of light on certain cellular functions. As a general rule, the treatment with light is carried out in the far red or near infrared spectral ranges, since the shorter wave irradiation can have various toxic effects on the cells.

The facts of the beneficial (curative) effect of low-dose long-wave irradiation to heal wounds treat injuries, infections and common pathologies such as diabetic retinopathy and age-related macular degeneration of the retina have been well established [1, 2]. It is known that photobiomodulation suppresses inflammatory processes caused by a decrease in mitochondrial activity [1], restores the balance of the redox potential of the glutathione system at the toxic action on cells [3], enhances the process of phagocytosis RPE cells of the human eye [4].

The positive effects of photobiomodulation on cells can be associated with the activation of a variety of photosensitive molecules, including melanins, hemoglobin, cytochrome-c-oxidase, etc. The exact mechanism and specific molecular targets of photobiomodulation are not completely clear. At least seven different ways of the primary effect of low-dose laser and LED irradiation of cells have been proposed [5,6]. However, according to T. Karu [5,7], the universal mechanism of photobiomodulation is

associated with photoactivation of the cytochrome-*c*-oxidase, the mitochondrial respiratory chain enzyme. This leads to an increase in the mitochondrial membrane potential (MMP), an increase in the level of intracellular ATP, a change in the redox state of the cellular cytoplasm [7]. The spectrum of action of photobiomodulating effects indicates that the cytochrome-*c*-oxidase molecule acts as the primary photo-acceptor in one of its intermediate redox states, absorbing light in the range of 620-820 nm [7]. It can be assumed that since cytochrome-c-oxidase absorbs light also in the blue spectral range, this spectral range can also have photobiomodulation effects.

So it was shown [6] that irradiation with a blue laser at 442 nm restores mitochondrial respiration suppressed by nitric oxide, which is probably associated with photoactivation of cytochrome-*c*-oxidase. It is well known, however, that the light in the blue range is extremely toxic to the cell [8]. Therefore, in order to register the stimulating effect of blue irradiation, it is important to choose conditions so that its toxicity is minimized or absent at all. Studies of the effect of sublethal doses of blue light on human RPE (ARPE-19) cells showed that light at 405 nm, even at doses of 3.2 J/cm^2 and 10 J/cm^2, not only did not lead to apoptosis of cells, but practically did not change their metabolic activity and MMP values [9]. The purpose of this study was to select the conditions for irradiation of cells with blue light, under which the metabolic characteristics will improve, as it does under the effect of long-wave irradiation. To solve this problem, we used a low dose ($\leq$ 1 J/cm^2) irradiation of the LED with a blue light source with a maximum at 450 nm.

Materials and Methods

Exposure of Cells to Blue Light

A suspension of the RPE cells of the Japanese quail, Coturnix japonica, and the human monocyte cell line THP-1 were used in the study as model systems in vitro. The experiments complied with the ethical principles and regulations as recommended by the European Science Foundation (ESF) and the declaration on the Humane Treatment of Animals.

Microscopic control showed that the RPE suspension contains exclusively a suspension of small clusters of 5-10 free-floating cells with a characteristic fringe of melanin apical processes and a submicroscopic suspension of single melanin granules. RPE and monocyte THP-1 cells were irradiated with a blue (450 nm) LED light source. The time and distance to the object were varied in order to change the irradiation doses, which were from 0.1 J/cm^2 to 1 J/cm^2. Measurements of the irradiation dose of light were made by a spectrometer "Avantes – 2048" (Netherlands). The time of irradiation, as a rule, did not exceed 50 minutes. Irradiation was carried out at a temperature of 37°C or room temperature. The control samples were stored in the darkness.

Morphogenesis of Mitochondria of RPE Cells

Evaluation of the mitochondrial state in RPE cells at low dose blue irradiation was carried out by morphometric analysis of the electron microscopy images. Transmission electron microscopy (JEM-1011, JEOL Ltd., Japan) of transverse sections of the central (macular) zone of the retina (about 1 mm^2 in size) was used. The specimens were fixed in a mixture of 2.5% glutaraldehyde ("Pan Real", Spain) with 2% formaldehyde ("MP Biomedicals ", France) on phosphate buffer (pH 7.2-7.4). A visual analysis of mitochondrial ultrastructure of RPE cells was carried out at a magnification of 10,000 x. Quantitative morphometry was performed on digitized electrograms using Adobe Photoshop CS4 software, in which the numerical density and the specific volume of mitochondria were determined within a square test grid (64 μm^2). For the measurements, the eyes of 3 birds were used; at least 25 RPE cells from the different parts of the macular zone were examined from each bird. The statistical analysis of the data was carried out in the Statistica10 program using the non-parametric Mann-Whitney test, the differences were recognized as significant at $p < 0.05$. The Kolmogorov-Smirnov criterion was used to estimate the normality of the distribution.

Measurement of Antioxidant Activity of RPE Cells

The antioxidant activity (AOA) of a suspension of RPE cells from Japanese quail was measured with a chemiluminescent model oxidation system consisting of hemoglobin, hydrogen peroxide and luminol [10]. Suspensions of RPE cells incubated in Hanks buffer solution supplemented with 10% fetal serum were

studied at 37°C and constant stirring. The duration of the latent period of development of the maximum luminescence intensity and/or the intensity of luminescence before and after addition of the cell suspension was used as the measured parameters. As the control samples were used RPE cells stored in the darkness under constant mixing, and also a buffer solution containing no cells. The freshly prepared initial suspension of the RPE cells was divided into two equal parts to minimize the melanin selection effect. A part of the suspension remained in the dark, the other was irradiated with blue light with an energy of 1 J/cm^2. The chemiluminescence kinetics were recorded on a "Shimadzu RF5301PC" spectrofluorometer at a luminescence wavelength of 470 nm. AOA activity was assessed by the degree of increase in the latent period of chemiluminescence as a function of the protein concentration of the added suspension of RPE cells. The deviation of data is presented by measuring five different samples.

Measurement of the Mitochondrial Membrane Potential (MMP)

Measurement of MMP in Human Monocytes by flow Cytofluorimetry

The studies were performed on the human monocyte cell line THP-1. The cell suspension was cultured in RPMI medium with 10% fetal bovine serum. To study the changes in the mitochondrial membrane potential (MMP), a protocol for the intravital staining of THP-1 cells in suspension with fluorescent potential-dependent TMRE dye (tetramethyl rhodamine ether) was worked out [11, 12]. For this, the cells were incubated at 37°C for 60 minutes with different concentrations of TMRE (10 nM, 30 nM, 50 nM, 100 nM and 250 nM).Then a recording of positive control was taken on the FACSAria SORP flow sorter. The excitation wavelength for TMRE was 561 nm, the emission intensity was recorded using a photoelectron multiplier equipped with a 505LP + 585/20BP set of filters. Separation of living single cells was carried out according to the parameters of direct and lateral light scattering. Populations of intensely fluorescent cells (colored with TMRE+) and weakly fluorescing (non-colored TMRE-apoptotic) cells were evaluated as a percentage. The percentage of uncolored cells in our experiments did not exceed 1% of the total number of treated cells. For both cell populations, the average fluorescence intensity and standard deviation from the mean (SD) were calculated. Since the studied populations showed an asymmetric distribution according to the level of intensity of TMRE coloring, the fluorescence intensity median was also analyzed, since the median distribution was stable against abnormal deviations of the indices. As a negative control, a reduction of potential of the inner mitochondrial membrane was made with the help of the FCCP oxidative phosphorylation uncoupler (10 μM and 100 μM, 30 minutes) followed by recording on a FACSAria SORP flow sorter. Since the best results in the separation of populations of TMRE-colored cells and cells losing color due to the release of MMP by the uncoupler were obtained by coloring with 10 nM TMRE and inhibition of 10 мM FCCP, these concentrations were used by us in further experiments. After irradiating the cell suspension, all samples were incubated with 10 nM TMRE for an hour. Positive control was incubated with 10 nM TMRE for an hour in a darkened box. Negative control was incubated with 10 nM TMRE for an hour in a darkened box and then with 10 мM FCCP for 30 minutes. After that, all samples were recorded on the flow sorter FACSAria SORP.

Measurement of MMP in a Suspension of the RPE Cells of the Japanese quail Eye

MMP of the RPE cell suspension was also evaluated using a fluorescent specific mitochondrial potential-sensitive TMRE dye with fluorescence measurement at a wavelength of 575-585 nm according to the described procedure [11, 12]. The wavelength of the exciting light was 549 nm. Samples of RPE cells were incubated in Hanks buffer solution (HBSS "Sigma") supplemented with 10% fetal calf serum ("Sigma"). TMRE concentrations ranging from 50 nM to 300 nM were used, depending on the concentration of homogenates and the intensity of their pigmentation with melanin. The experiments were performed in two versions. In the first version, the cells were preloaded with a dye, and then they were irradiated already in the presence of a dye. In the second variant, dye-free cells were irradiated, then the dye was added and the kinetics of its distribution between the cells and the environment were recorded. In separate experiments, it was shown that blue light in the doses used by us and at the used dye

concentrations did not cause a change in the fluorescence characteristics of TMRE. The accumulation of the dye in RPE cells was evaluated by its content in a centrifuged cell pellet (5 min at 12000g, followed by extraction with dimethylsulfoxide to eliminate the effect of fluorescence quenching by melanin), as well as its residual content in the extracellular incubation solution. The mitochondrial specificity of TMRE accumulation was monitored by the FCCP oxidative phosphorylation uncoupler (20 μM).

Measurement of Total Metabolic Activity of RPE Cells

The metabolic activity of the cells was quantified using a vital resazurin dye, according to the Sigma-Aldrich instructions given in the studies [13,14].

Bioremediation of resazurin by metabolically active cells leads to a decrease in the concentration of its blue oxidized form and to an increase in the concentration of the red fluorescent intermediate. Resazurin (final concentration 50 μM) was added to a suspension of RPE cells previously irradiated with blue light, then it was incubated for 10-15 minutes, after which a zero sample was taken and incubated at 37 °C for 2-6 hours. The incubation medium is Hanks buffer with 10% fetal serum. Control samples contained buffer and resazurin without the addition of cells, as well as samples with cells incubated in the darkness. Estimation of resazurin recovery was made by increasing the intensity of fluorescence at an excitation wavelength of 560 nm and an emission maximum of 590 nm.

Statistical Analysis

The data were expressed as the mean ± SD. For the statistics, Student's t-test was used. $P < 0.05$ was considered as statistically significant.

RESULTS

Metabolic Activities

To determine the effect of low-dose blue irradiation on RPE cells, a quantitative assessment of cell damage was carried out by measuring the recovery of a vital resazurin dye.

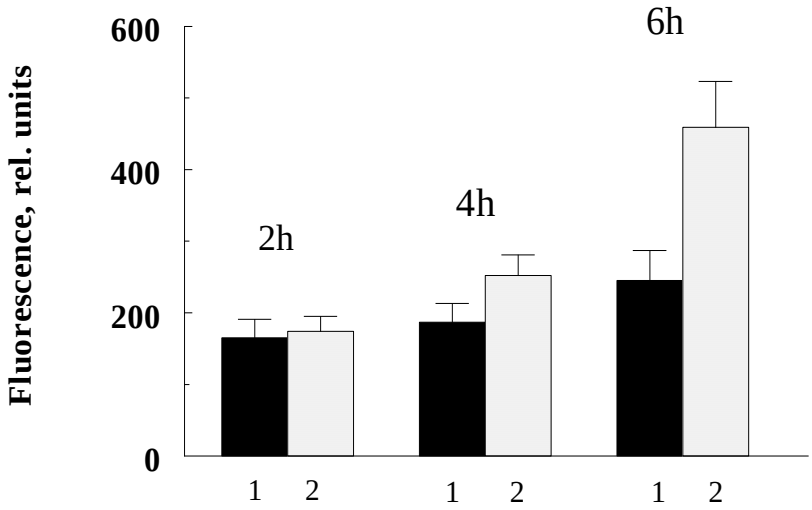


Figure1. *Stimulation of the metabolic activity of quail eye RPE cells after their low dose irradiation from the LED source. Restoration of resazurin by RPE cells after their irradiation with blue light at a dose of 0.3 J/cm^2. 1 - dark control, 2 - irradiated cells. Fluorescence was measured after 2, 4 and 6-hour incubation of cells with resazurin at 37 °C.*

From Figure1 can be seen that blue irradiation at a dose of 0.3 J/cm^2 (bars 2) leads to an enhanced recovery of resazurin compared to the control (bars 1), most pronounced after 6 hours of incubation. In none of the experiments has been registered a decrease in the intensity of the resazurin recovery process after irradiation of RPE cells with blue light.

Antioxidant Activity

The AOA activity was evaluated using a chemiluminescent system containing luminol and hydrogen peroxide. AOA activity in this system is characterized by the degree of decrease in the intensity of chemiluminescence emission and the increase in the latent period of reaching the maximum luminescence amplitude when a certain amount of the cell suspension is added to the system.

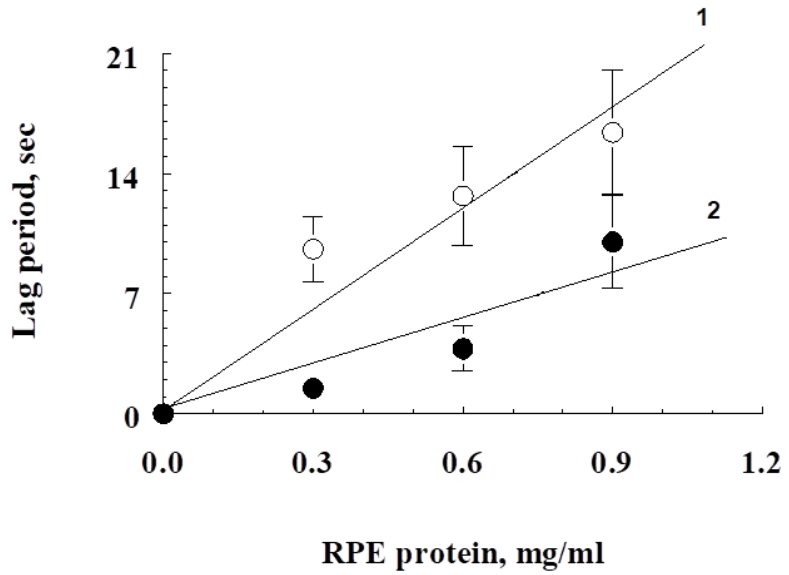
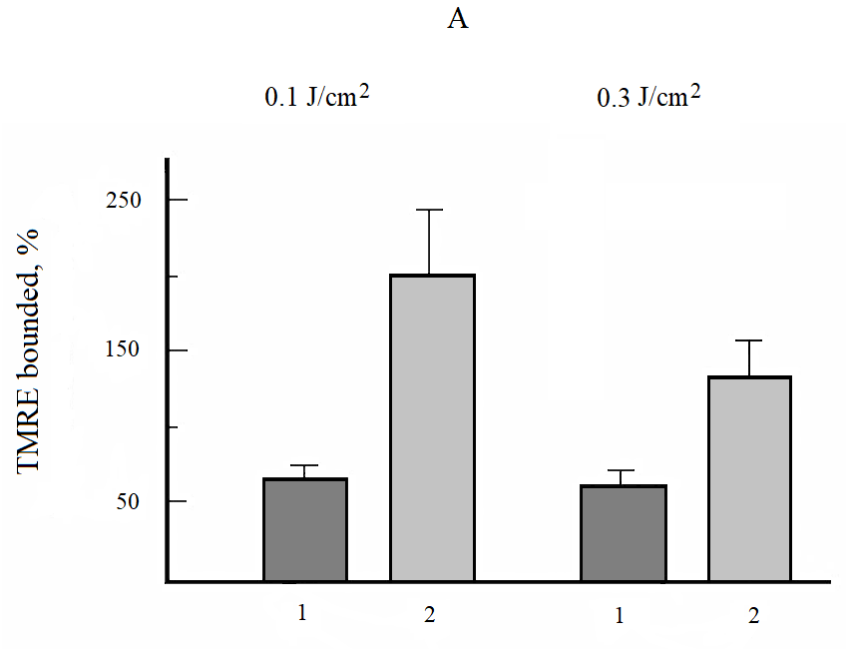


Figure2. *The increase of antioxidant activity of the quail's eye RPE cells after their irradiation with blue light. The y-axis indicates the latent period for reaching the chemiluminescence maximum in seconds, and the abscissa is the protein concentration of the suspension of the RPE cells. Curve 1 - irradiated (1 J/cm^2) cells, curve 2 - dark control (non-irradiated cells)*

Figure 2 shows a true increase in the latency period for reaching the maximum of fluorescence caused by the addition of a suspension of the RPE quail cells after their irradiation with blue light at a dose of 1 J/cm^2 (curve 1) compared to the suspension of cells in the darkness (curve 2). This indicates a higher antioxidant activity of irradiated cells.

Mitochondrial Membrane Potential

To evaluate the effect of blue irradiation on MMP, a fluorescent potential-dependent TMRE dye was used. Evaluation of the change in MMP under the action of light was carried out on a suspension of the RPE cells of the Japanese quail eye and on the culture of human monocyte cells. It should be noted that the cells of the quail RPE have a high content of black pigment melanin, which makes it difficult to measure the fluorescence intensity of TMRE. Therefore, in experiments on quail RPE cell suspensions, to estimate the amount of dye absorbed by the cells as a result of mitochondrial activation, the fluorescence was measured in extracts containing no melanin pigment.



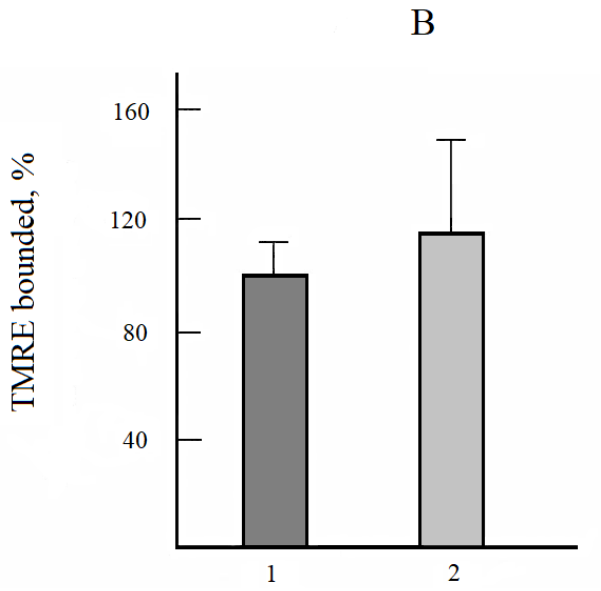


Figure3. *Accumulation of mitochondrial cationic dye TMRE by the quail's eye RPE cells after blue irradiation in low doses*

A. Increase in fluorescence upon irradiation of RPE cells preliminarily loaded with the mitochondrial TMRE dye. Irradiation with blue light with doses of 0.1 J/cm^2 and 0.3 J/cm^2. 1 - dark control, 2 - irradiated cells. The concentration of the dye absorbed at the zero point (before irradiation) is assumed as 100%.

B. Accumulation of TMRE by RPE cells after their irradiation with blue light in the absence of a dye. 1 - dark control, 2 - irradiation with blue light at a dose of 0.3 J/cm^2. The concentration of the dye absorbed at the zero point (before irradiation) is assumed as 100%.

Figure 3A shows a significant increase of fluorescence in RPE cells irradiated with blue light at a dose of 0.1 J/cm^2 (Figure 3A) and a reliable but smaller increase in the fluorescence intensity of cells irradiated at a dose of 0.3 J/cm^2. Figure 3B demonstrates mitochondrial stimulation during irradiation by the blue light of the RPE cells originally containing no fluorescent dye. The dye was added to the RPE cells after their irradiation and then the process of fluorescence distribution between the cells and the incubation medium was recorded. These results indicate that the activated cells retain this state for a certain time.

Figure 4 shows the distribution of the population of human THP-1 monocyte cells colored with TMRE, without irradiation (A) and after irradiation of the cells with blue light (B) in doses of (1 J/cm^2).

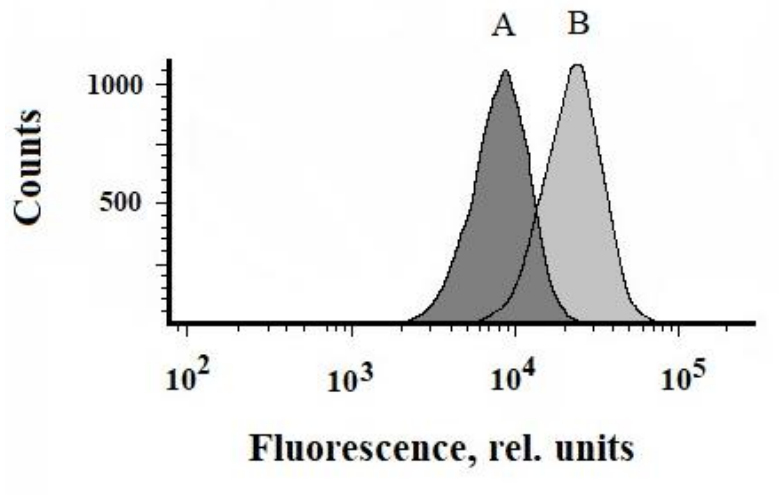


Figure4. *Influence of low-dose blue LED irradiation on the mitochondrial membrane potential of human monocyte cell culture. A - control without irradiation; B - irradiation with blue light in a dose of 1 J/cm^2.*

All data were normalized in relation to control - THP-1 cells colored with TMRE and not subjected to other actions. It can be seen that irradiated cells had a much higher fluorescence intensity compared to non-irradiated ones. A comparison of the mean and median fluorescence intensities of THP-1 cells colored with TMRE without irradiation and with blue light irradiation is presented in the table.

Table1. *The increase of the mitochondrial membrane potential in THP-1 cells when irradiated with blue (450 nm) light from a LED lamp*

Conditions for illumination of culture cells THP-1	**The fluorescence intensity of TMRE-stained cells, Rel. Units**		
	Average intensity	Median intensity	Standard deviation (SD)
Control,without irradiation	7961	7660	2938
Irradiation with blue light, 1 J/cm^2	17371	16680	3042

The data show that the median intensity of cell fluorescence after blue light irradiation increases more than twice.

Increase in the Mitochondria Amount in Response to Irradiation

A single short-term (15 min) exposure to low-dose blue irradiation on the eyes of 78-week-old birds caused a small but a true increase in the amount and specific volume of all mitochondria. If in control unirradiated cells the mitochondrial amount was 17.8 ± 0.7 and their specific volume was 11.8 ± 0.7, then after irradiation the number of mitochondria increased to 20.6 ± 0.9 and their specific volume to 15,3 ± 0.7. The total number of modified mitochondria did not change (Figure 5).

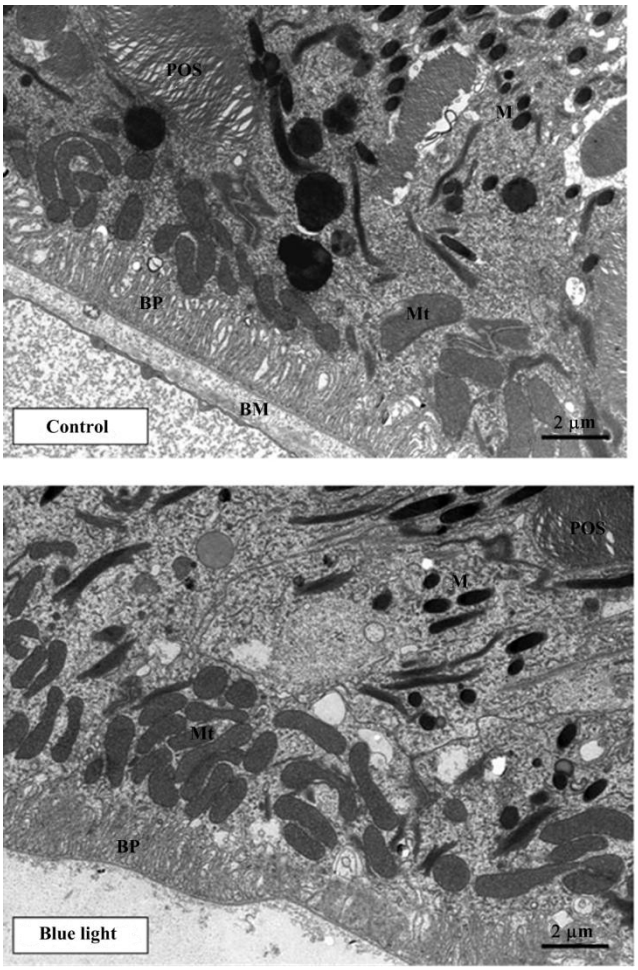


Figure5. *Electron microscopy images of the cross-section of RPE cells of the Japanese quail before (control) and after irradiation with blue light. Mt - mitochondria, MB - Bruch's membrane, M - melanin granule, BP - basal processes, POS – photoreceptor outer segment*

DISCUSSION

In this study we present and study a model system in vitro of quail RPE cells and human monocyte cell line THP-1 in which irradiation with blue light from an LED source does not induce stress reactions and cell apoptosis. Blue light at 450 nm in low doses, used in this model, has a positive effect on cellular functions. Using the resazurin test, we have found an increase in the metabolic activity of RPE cells in response to this irradiation. Moreover, irradiation of a suspension of RPE cells even in relatively high doses (1J/cm^2) also resulted in a significant increase in the resazurin reduction rate (not shown in the figure). The increase in the metabolism of RPE cells can explain the observed increase in the antioxidant activity of RPE cells irradiated with blue light. It is known that irradiation of RPE cells in sublethal doses (3 J/cm^2) leads to an increase in the production of reactive oxygen species [9]. This in turn leads to activation of the antioxidant defense system [15], which is facilitated by an increase in the metabolic activity of cells in response to irradiation. This effect can explain the fact that we discovered the increase in the antioxidant activity of RPE cells in response to irradiation with blue light. It is known that blue light can interact with mitochondrial cytochrome-c-oxidase [16,6]. In this case blue light irradiation in high doses can lead to damage to the mitochondrial apparatus and impairment of cellular functions [17]. On the other hand, irradiation of mitochondria with blue light in relatively low doses may have a positive effect [6].

In our model systems in vitro irradiation with the blue light of the RPE cells of the Japanese quail and human monocytes led to an increase in the absorption of the mitochondrial potential-dependent TMRE dye, which indicates an increase in MMP of irradiated cells. It should be noted that irradiation of the cells with blue light with doses up to 1 J/cm^2 did not lead to a decrease in MMP and, consequently, to a decrease in energy metabolism in mitochondria in any of the performed experiments.

The increase of the metabolic activity of Japanese quail RPE cells can also be associated with an increase in the number of mitochondria, accompanied by an increase in their specific volume.

This growth can be caused by the division of mitochondria. These results are confirmed by the data of other authors [18], that showed an increase in the number of mitochondria after low-dose laser and LED irradiation with red light.

Thus, the obtained results indicate that the action of blue (450 nm) LED irradiation at low doses can have a positive effect on cellular functions and can be used in certain cases as a photobiomodulating agent.

ACKNOWLEDGEMENTS

The work was supported by the Russian Foundation for Basic Research (grant № 15-29-03865).

REFERENCES

[1] Begum R, Powner MB, Hudson N, Hogg C, Jeffery G Treatment with 670 nm light upregulates cytochrome C oxidase expression and reduces inflammation in an age-related macular degeneration model. PLoS ONE 2013; 8: e57828

[2] Tang J, Du Y, Lee CA, Talahalli R, Eells JT, Kern TS Low-intensity far-red light inhibits early lesions that contribute to diabetic retinopathy: in vivo and in vitro. Invest Ophthalmol Vis Sci 2013; 54: 3681-3690

[3] Yeager RL, Oleske DA, Sanders RA, Eells JT, Henshel DS Melatonin as a principal component of red light therapy. Med Hypotheses 2007; 69: 372-376

[4] Fuma S, Murase H, Kuse Y, Tsuruma K, Shimazawa M, Hara H Photobiomodulation with 670 nm light increased phagocytosis in human retinal pigment epithelial cells. Molecular Vision 2015; 21: 883-892

[5] Karu T Primary and secondary mechanisms of action of visible to near-IR radiation on cells. J. Photochem Photobiol B Biol 1999; 49: 1-17

[6] Buravlev E, Zhidkova T, Osipov A, Vladimirov Y Are the mitochondrial respiratory complexes blocked by NO the targets for the laser and LED therapy? Laser Med Sci 2015; 30: 173-180

[7] Karu T Universal cellular mechanism of laser biostimulation: photoactivation of the respiratory chain enzymecytochrome c oxidase. Digest Scientific Works IPLIT RAS 2003; 131-143 [in Russian].

[8] Wu J, Seregard S, Algvere PV Photochemical damage of the retina. Surv Ophthalmol 2006; 51: 461-481

[9] Roehlecke C, Schaller A, Knels L, Funk RHW The influence of sublethal blue light exposure on human RPE cells. Molecular Vision 2009; 15: 1929-1938

[10] Teselkin Y, Babenkova I, Lyubitsky O, Klebanov G, VladimirovY Measurement of antioxidant activity of blood serum using the hemoglobin-hydrogen peroxide-luminol system. Voprosy Med Chemistry 1998; 44: 70-76 [in Russian].

[11] Gan Z, Audi SH, Bongar RD, Gauthier KM, Merker MP Quantifying mitochondrial and plasma membrane potentials in intact pulmonary arterial endothelial cells based on extracellular deposition of rhodamine dyes. Am J Lung Cell Mol Physiol 2011; 300: 762-772

[12] Scaduto RCJ, Grotyohann LW Measurement of mitochondrial membrane potential using fluorescent rhodamine derivatives. Biophys J 1999; 76: 469–477

[13] O'Brien J, Wilson I, Orton T, Pognan F Investigation of the Alamar blue (resazurin) fluorescent dye for the assessment of mammalian cell cytotoxicity. Eur J Biochem 2000; 267: 5421-5426

[14] Gonzalez RJ, Tarloff JB Evaluation of hepatic subcellular fractions for Alamar blue and MTT reductase activity. ToxicolIn Vitro 2001; 15: 257-259

[15] Roehlecke C, Schumann U, Ader M, Brunssen C, Bramke S, Morawietz H, Funk RHW Stress reaction in outer segments of photoreceptors after blue light irradiation. PLOS ONE 2013; 8: 1-12

[16] Lascaratos G, Ji D, Wood JP, Osborne NN Visible light affects mitochondrial function and induces neuronal death in retinal cell cultures. Vision Res 2007; 47: 1191–1201

[17] Kroemer G Mitochondrial control of apoptosis: an introduction. BiochemBiophys ResCommu n 2003; 304: 433–435

[18] Passarella S, KaruT Absorption of monochromatic and narrowband radiation in the visible and near IR by both mitochondrial and non-mitochondrial photoacceptors results in photobiomodulation. J Photochem Photo biolB 2014; 140: 344–358

Citation: *Natalia Seryoznikova et.al, "The Stimulating Action of Low-Dose Blue Light Irradiation on Functional Activity of Retinal Pigment Epithelium Cells". International Journal of Research Studies in Medical and Health Sciences. 2018; 3(11):01-09.*

Copyright: *© 2018 Natalia Seryoznikova et.al, This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.*