საქართველოს მეცნიერებათა ეროვნული აკადემიის მოამბე, ტ. 17, №3, 2023
BULLETIN OF THE GEORGIAN NATIONAL ACADEMY OF SCIENCES, vol. 17, no. 3, 2023

Human and Animal Physiology

Effect of Blue LED on Oxidative Stress Indices (Experimental Study on White Rats)

Levan Mikeladze[*], Nino Karanadze[*], Davit Delibashvili[*], Nino Tskhvediani[*], Nodar Mitagvaria[**]

[*] *Medicine Faculty, Tbilisi State Medical University, Tbilisi, Georgia*
[**] *Academy Member, Georgian National Academy of Sciences, Beritashvili Center for Experimental Biomedicine, Tbilisi, Georgia*

The light of blue electromagnetic waves has a negative effect on a living organism, which is manifested by the massive production of free radicals that accompany oxidative stress and destruction of the cellular structure, inflammation and a reduction in the lifespan of cells. High-energy blue electromagnetic waves, unlike rays, have the ability to penetrate deeper into the skin. The reason for this is to penetrate the normally well-protected layer of the skin and act on the collagen and elastin present there, which provides elasticity to the skin and keeps it looking youthful. If we add to this the effects of destroying the skin's natural protective barrier and slowing down skin regeneration, we can confirm that blue light accelerates aging. The number of smartphones around the world has increased in recent years, and many of them have a blue spectrum. Based on the foregoing, we considered it necessary to study the effect of irradiation with blue monochromatic light on the generation of free radicals in experiments carried out on rats. The aim of the study was the intensity of free radical formation in rats exposed to an incandescent lamp and a blue light-emitting diode. The study was conducted on 24 white rats, which were divided into three groups. The animals in the first series were under natural lighting conditions, during the second and third series under the influence of a 100-watt incandescent lamp and a blue monochromatic light-emitting diode. Oxidative stress indicators were recorded using the Italian computer system FRAS-5. It was measured (d-ROM-test) – hydroperoxidase concentration in the blood, i.e. reactive oxygen metabolites, through the PAT test – systemic antioxidant potential, the OBRI index determined the oxidative balance status, and the OSI index summarized the information obtained from the ROM test and the PAT test. The seventh edition of Stanton A. Glantz software – one-way analysis of variance – ANOVA and Bonteroni correction t-test were used for statistical processing of the obtained data. As a conclusion, we can use this very last change in the value of oxidative stress – irradiation with blue monochromatic light dramatically increases the index of oxidative stress. *© 2023 Bull. Georg. Natl. Acad. Sci.*

white rats, blue light, oxidative stress, oxygen metabolites, antioxidant test

A living organism has a unique feature: in response to many different natural external (and also produced in the body) factors (oxidative stress), it activates internal protective, protective-compensatory, including immune mechanisms and not only prevents harmful effects, but also develops and enhances the body's ability to fight the same

© 2023 Bull. Georg. Natl. Acad. Sci.

harmful factors when exposed to an increasing dose of factors, i.e. in response to oxidative stress.

The problem of oxidative stress has been intensively studied since the 60s of the last century. Its relevance has reached its peak today. It is probably difficult to find any pathological condition that is not accompanied by or involved in the development of this phenomenon – neurodegenerative disorders, cancer diseases, development of ischemic cascade, Parkinson's and Alzheimer's diseases, etc. [1-5].

The tests were carried out on the control and experimental groups of 24 white laboratory rats weighing 200-250 g. Each group consisted of healthy animals. After placing the animals on the device, they were irradiated both with a conventional lamp (100 W) in the control group and with a blue (IP 66 20 W) LED. Irradiation with both types of light was carried out daily (for 7 days), the material was taken on the 8th day. Oxidative stress indicators were recorded by the Italian computerized system FRAS-5 (Free Radical Analytical System, H@D company), which allows measuring the following free radical indicators:

1. d-ROM – This is a photometric test, it essentially determines the concentration of hydroperoxides in the blood, these substances belong to the broad class of reactive oxygen metabolites (ROM). According to the internationally accepted standard 1U.Carr=0.08 mg H202/di and 250-300 U.Carr or d-ROM is considered normal, 300 to 320 is the border with normal and elevated level, 321 and more is high oxidative stress.

2. PAT-test – (Plasma Antioxidant Test). The PAT test allows us to measure the concentration of antioxidants in the blood, as agents that reduce iron from the Fe 3+ form to the Fe2+ form. This test is expressed as COR. A unit corresponding to 1.4 μmol/L Vit C. If the level of PAT is greater than 2800, it is considered a high level, 2200-2800 is normal, and less than 2000 is an indicator of deficiency status.

3. The OBRI index (Oxidative Balance Index) determines the status of oxidative balance based on cholesterol levels and is an interesting predictive index of cardiovascular risk. It is an indicator of the state of oxidative stress, the norm varies between 0.8-1.2.

4. The OSI Index (Oxidative Stress Index) summarizes the information obtained from the d-ROM test and the PAT test into a single value and makes the interpretation of the result easier and more immediate. < 40 is considered normal.

Thus, the use of FRAS 5 allows us to find out and get information about the state of oxidative stress and antioxidants in the blood of the patient (or animal, in our case).

Materials and Methods

Algorithm of conducted tests. In the tests presented by us, the following algorithm was selected:

The first series of tests (10 rats) is performed on intact animals, that is, under normal conditions. We take 1 ml of blood from the superior vena cava of the experimental rat. This blood is then separated into two eppendorfs, which are placed in the FRAS 5 centrifuges, and the instrument begins the measurements. The obtained results are automatically printed on the tool's printer.

Thus, the use of FRAS5 initially allows us to find out and obtain information about the state of the patient (or an intact animal, in our case) under normal conditions regarding oxidative stress and antioxidants in the blood (the first series of tests).

The second and third series of experiments are carried out both after irradiation with an incandescent lamp (6 rats – the second series of experiments) and using a blue LED lamp (8 rats – the third series of experiments). The corresponding device is presented.

The results obtained in the tests carried out are presented in the table below, the first column of which describes the effects in animal experiments, and the following columns show the results obtained with the parameters described above (d-

ROM, PAT, OBRI and OSI). Stanton A. Glantz "Primer of Biostatistics" software (7th edition) was used to statistically process the data obtained based on one-way analysis of variance and multiple comparison with the corrected Bonferroni t-test. The statistical significance level was taken as 0.05.

The Table of the obtained results and diagrams based on it are presented on the following pages.

The graphs below present the results of the analysis through FRAS-5, which allow us to make the following conclusions at the bottom of the article.

Results and Discussion

Analysis of diagrams. In all obtained diagrams, we have separated three experimental conditions – animals are in an intact (ie normal) condition, in the conditions of irradiation with incandescent lamp and irradiation with blue monochromatic light [1-3].

Table. The results obtained in experiments

		d-ROMs U.carr	PAT U. Cor	OBRI index	OSI index
I Control group		296.2±67.7	2573±305.7	0.91±0.42	54.5±70.2
II Incandescent lamp		254.5±42.5	2770±394.4	0.78±0.12	23.8±15.7
III Blue LED		326.4±109.7	3515±435	0.8±0.19	54.5±50.2
Between groups	SS	17710	3990000	0.081	3688
Between groups	DF	2	2	2	2
Between groups	MS	8857	1995000	0.0405	1844
Whithin group	DF	21	19	21	20
	p	0.273	0	0.642	0.566
	F	1.38	14.07	0.45	0.59
t	I-II	1.403	1.32	0	1.38
t	I-III	1.118	7.273*	0	0
t	II-III	2.323	4.908*	0	1.326

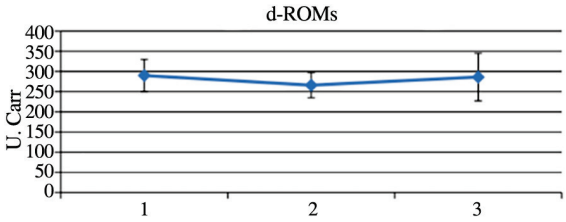

Fig. 1. Number of free radicals: 1 – intact animals, 2 – irradiation with a conventional lamp (100W), 3 – irradiation with a blue monochromatic lamp.

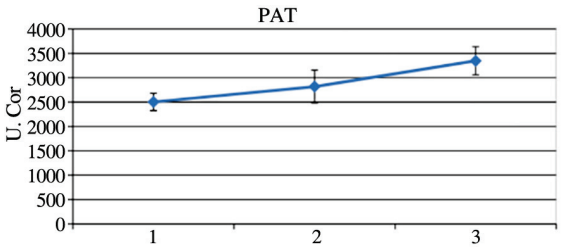

Fig. 2. Number of antioxidants: 1 – intact animals 2 – irradiation with a conventional lamp (100W), 3 – irradiation with IP66 LED blue monochromatic light flow.

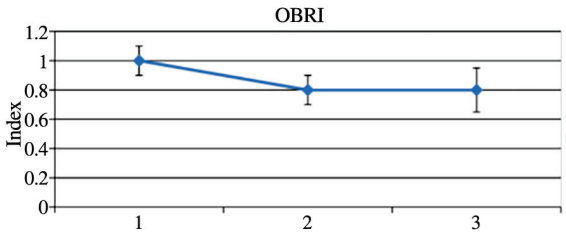

Fig. 3. Oxidative balance index: 1 – intact animals, 2 – irradiation with incandescent lamp (100W), 3 – irradiation with IP 66 LED blue (monochromatic) light flow.

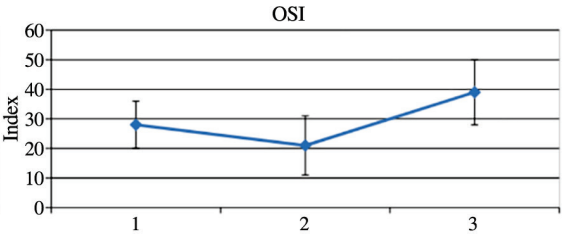

Fig. 4. Oxidative stress index: 1 – intact animals, 2 – irradiation with incandescent lamp (100W), 3 – irradiation with IP 66 LED blue (monochromatic) light flow.

As we can see in Fig. 1, irradiation with a incandescent (100 watt) lamp led to a slight decrease in free radicals, and when irradiated with blue monochromatic light, the amount of free radicals practically returned to the initial state.

Fig. 2 shows the change in free radicals (PAT test). It was statistically significantly increased by 100% when exposed to blue monochromatic light.

Fig. 3 shows the Oxidative Balance Index (OBRI), which, as previously mentioned, determines the status of oxidative balance based on cholesterol levels and is a predictive index of cardiovascular risk. This indicator is sufficiently reduced when irradiated with a incandescent lamp and, in fact, remained at the same level under the conditions of irradiated with blue monochromatic light.

And finally, the Oxidative Stress Index (OSI), as we can see in Fig. 4, is dramatically increased under blue monochromatic light irradiation conditions. It summarizes the information obtained from the d-ROM test and the PAT test into one value.

Conclusion

As a conclusion, we can use this very last change in the value of oxidative stress – irradiation with blue monochromatic light dramatically increases the index of oxidative stress.

ადამიანისა და ცხოველთა ფიზიოლოგია

ლურჯი ფერის შუქ-დიოდის გავლენა ოქსიდაციური სტრესის მაჩვენებლებზე (ექსპერიმენტული კვლევა თეთრ ვირთაგვებზე)

ლ. მიქელაძე*, ნ. კარანაძე*, დ. დელიბაშვილი*, ნ. ცხვედიანი*, ნ. მითაგვარია**

** თბილისის სახელმწიფო სამედიცინო უნივერსიტეტი, მედიცინის ფაკულტეტი, თბილისი, საქართველო*

*** აკადემიის წევრი, საქართველოს მეცნიერებათა ეროვნული აკადემია, ბერიტაშვილის ექსპერიმენტული ბიომედიცინის ცენტრი, თბილისი, საქართველო*

კვლევის მიზანი იყო ვირთაგვების თავისუფალი რადიკალების წარმოქმნის ინტენსივობა ვარვარა ნათურისა და ლურჯი ფერის შუქდიოდის ზემოქმედების დროს. კვლევა ჩატარდა 24 თეთრ ვირთაგვაზე, რომელიც დაყოფილი იყო სამ ჯგუფად. პირველი სერიის ცხოველები იყვნენ ბუნებრივი განათების პირობებში, მეორე და მესამე სერიის დროს 100-ვატიანი ვარვარა ნათურისა და ლურჯი მონოქრომატული შუქდიოდის ზემოქმედების ქვეშ. ოქსიდაციური სტრესის მაჩვენებლების აღრიცხვა ხდებოდა იტალიური კომპიუტერული სისტემით FRAS-5 მეშვეობით. იზომებოდა (d-ROM-ტესტი) – ჰიდროპეროქსიდაზას კონცენტრაცია სისხლში,

ანუ, რეაქტიული ჟანგბადის მეტაბოლიტები; PAT ტესტის მეშვეობით – სისტემური ანტიოქსიდანტური პოტენციალი, OBRI ინდექსით განისაზღვრებოდა ჟანგვითი ბალანსის სტატუსი, ხოლო OSI ინდექსი აჯამებდა ROM ტესტი და PAT ტესტებიდან მიღებულ ინფორმაციას. მიღებული მონაცემების სტატისტიკური დამუშავებისათვის გამოყენებულ იქნა Stanton A.Glantz-ის პროგრამული უზრუნველყოფის მეშვიდე გამოცემა – ცალმხრივდისპერსიული ანალიზი – ANOVA და Bonteron-ის კორექტირებული t ტესტი. შეიძლება დავასკვნათ, რომ ოქსიდაციური სტრესის მნიშვნელობის ცვლილება – ლურჯი მონოქრომატული სინათლით დასხივება მკვეთრად ზრდის ოქსიდაციური სტრესის ინდექსს.

REFERENCES

1. Duteil L., Cardot-Leccia N., Queille-Roussel C., Maubert Y., Harmelin Y., Boukari F., Ambrosetti D., Lacour J-P., Passeron T. (2014) Differences in visible light-induced pigmentation according to wavelengths: a clinical and histological study in comparison with UVB exposure. *Pigment Cell Melanoma Res.*, 27(5):822-826. doi:10.1111/pcmr.12273. Epub. Jul 25. https://pubmed.ncbi. nlm.nih.gov /24888214/#full-view-affiliation-1.
2. Zastrow L., Groth N., Klein F., Kockott D., Lademann J., Ferrero L. (2008) Detection and identification of free radicals generated by UV and visible light in ex vivo human skin. *Physics / International Journal of Cosmetic Science Citation*: IFSCC Magazine, **11**(3): 207–215 DOI:10.1111/J.1468-2494.2009.00511 _2.X / Corpus ID: 95415196. https://www.semanticscholar.org/paper/Detection-and-Identification-of-Free-Radicals-by-UV-Zastrow-Groth/b94825ffec81960c30bdd423d9f986de10d0ab26.
3. Falcone D., Natallia E Uzunbajakava, Frank van Abeelen, Gerrit versluizen, Peppelman M., Piet E J van Erp, Peter C M van de Kerkhof (2018) Effects of blue light on inflammation and skin barrier recovery following acute perturbation. Pilot study results in healthy human subjects. *Photodermatol Photoimmunol Photomed,* **34**(3): 184-193. doi:10.1111/phpp.12367. Epub 2017 Dec 22. https://pubmed.ncbi.nlm.nih.gov/ 29150968.
4. Hoidal J. R. (2001) Reactive oxygen species and cell signaling. *Am J Respir Cell Mol Biol.,* **25**(6): 661-663. doi:10.1165/ajrcmb.25.6.f213. https://pubmed.ncbi.nlm.nih.gov/11726 388.
5. Russel J. Reiter (2000) Melatonin: lowering the high price of free radicals. *News Physiol Sci.,* 15: 246-250. doi:10.1152/physiologyonline.2000.15.5.246 https://pubmed. ncbi.nlm.nih.gov/11390919.

Received February, 2023