

SHORT COMMUNICATION

Effect of high-intensity light on the retinal pigment epithelium of zebrafish – An induced retinal damage study model

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ABSTRACT


Background: Natural light sources emit light with two physical properties namely, spectrum and intensity. However, the electronic light sources these days were adjusted in their parameters of color and the intensity without the consumer being aware of these. These lights become potential harm to its users both in household and handheld digital devices. **Aims and Objectives:** In this study, we have tried to estimate the changes in the morphology of the retinal pigment epithelium of zebrafish when exposed to high-intensity light. **Materials and Methods:** Institutional Animal Ethics Committee clearance was obtained. Two groups of zebrafish with an average bodyweight (690 ± 50 mg) and length (26.75 ± 1.39 mm) were taken for the study. Group I ($n = 6$) control group: Housing was in a glass tank in the common working area exposed to the sunlight for the light source. Group II ($n = 6$) study group: Housing was done in a dark room with the custom-built electrical light source with an adjustable setting for a uniform light intensity of 15,000 lux. Experimental study period was for 15 days with maintained 12 hrs light and 12 hrs dark cycle. Fishes were euthanized by exposing them to cold water with ice and the eyeballs removed and stored in 4% formalin. Whole eye histological staining was done with hematoxylin and eosin stain. The histological slides were photographed in a digital microscope and the distribution of neurons in the selected layer of the retinal epithelium was estimated and compared among the groups using a software. **Results:** The study results are conclusive with zebrafish animal models and further suggest that even under the water when exposed to high intensity of light there is a potential risk of damaging the retinal epithelium. **Conclusion:** An experimental damage of the retinal epithelium of zebrafish was established to be used for further research.

KEY WORDS: High-Intensity Light; Retinal Pigment Epithelium; Zebrafish

INTRODUCTION

Visible light is the primary source for degenerations in the retina as well as macular regions of the human eye.^[1] The same light can be used as an experimental tool to damage the retina with control on the intensity of exposure against

the required extent of retinal degeneration. Validated exposure protocols exist, such as (a) short-term exposure to white light, (b) long-term exposure to low-intensity white light, (c) exposure to broadband blue green-yellow light, and (d) exposure to monochromatic light of definite wavelength.^[1] These exposures take different molecular pathways to exhibit the damage caused due to the exposure to light waves. These days there is a shift in the use of light-emitting diode (LED) lights against the common compact fluorescent light. The advantage is reduction in the power consumption and thereby the consumers money. These LEDs enriched with blue radiations are known to be a hazard for human eyes.^[2] However, across the nations there is massive movement of conversion of conventional

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incandescent lights to LED incorporated lighting devices is happening, which is even enforced by the governmental agencies in different nations.^[2] LED toxicity has been studied in rat models as evident through many studies.^[3] However, in this study, we followed a different animal model, the zebrafish to establish a protocol that could be employed to explore the ill effects of high-intensity LED lights on the retinal epithelium of these animals which are capable of regenerating their damaged retina. Our main objective in this study is to establish the changes in the morphology of the retinal pigment epithelium of zebrafish, when exposed to high-intensity LED lights.

MATERIALS AND METHODS

Institutional Animal Ethics Committee clearance was obtained. Two groups of zebrafish (5 months old) with an average bodyweight (690 ± 50 mg) and length (26.75 ± 1.39 mm) were taken for the study. Group I ($n = 6$) control, fishes were housed in a glass tank (5 L) in the common working area exposed to the sunlight for light source whereas Group II ($n = 6$) was the group which was kept in a dark room with the custom-built electrical light source.^[4] These fishes in the study group were expose to a set intensity of light, 15000 ± 500 lux from an array of LED lights. Intensity of the light was measured using a lux meter^[4-6] and was assured of uniform light intensity in the different corners of the experimental tank. Experimental study period was for 15 days with maintained 12 hrs dark and 12 hrs light cycle. Fishes were euthanized by exposing them to ice-cold water and the eyeballs dissected and stored in 4% formalin. Whole eye histological staining was done with hematoxylin and eosin stain. Histological slides were photographed in a digital microscope under 40X. In each histopathology slide, we randomly selected two segments in the whole of the eye as the sample for further image processing and analysis. Distribution of neurons in the selected layer of the retinal epithelium was estimated and compared among the groups using special image processing software, Tissue quadrant V2.0.

RESULTS

The findings of the present study are depicted in Figure 1 and Table 1.

DISCUSSION

Statistically significant damage is seen in the ganglion cell layer of the retina. Damage to the retina shuts down the source of light inputs to the circadian pathways and therefore disturbances in the circadian rhythm. A similar approach to study the circadian rhythm was done by Vatine *et al.* (2011), which can also be done with this setup and the methodology.^[7] More and more studies wherein induced retinal damages and then its chronobiological effects can be studied with this

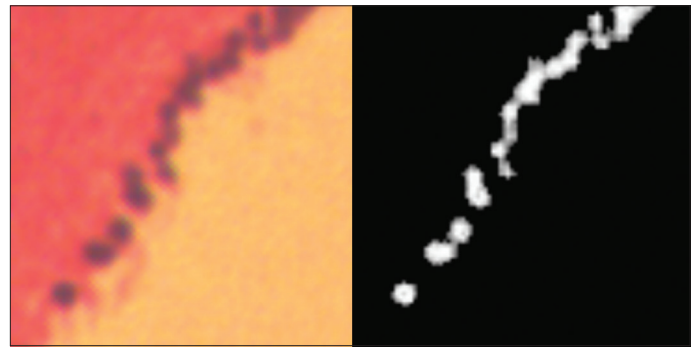


Figure 1: Color intensity quantification used to evaluate the amount of biological specimen in an image (software used: Tissue Quant v 2.0)

Table 1: Differences in the total pixel area occupied in the 1×1 cm of the retinal cross-sectional image of the zebrafish

Retinal image samples (1×1 cm)	Total area (pixel)	P-value
Control ($n=12$)	536.5 ± 46.24	$<0.0001^*$
Group ($n=12$)	243.8 ± 26.29	

*Statistically significant

experimental animal model set up. Behavioral changes in zebrafish animal models with respect to the disturbances brought about by the continuous exposure to bright lights can also be explored.

CONCLUSION

The prototype experimental model and the results obtained were promising in producing extensive damage in the retinal layer of the zebrafish and hence can be considered as an animal model for research that involves induced damage of retina or the light-dark cycle.

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