TÜBİTAK

Turkish Journal of Veterinary and Animal Sciences

http://journals.tubitak.gov.tr/veterinary/

Research Article

Turk J Vet Anim Sci
(2013) 37: 404-407

doi:10.3906/vet-1109-19

The effect of flashed light on the ultrastructure of the retinal photoreceptor cells in rabbit: a transmission electron microscope study

Arash ESFANDIARI[1,*], Iraj POSTI[2], Asghar DEHGHAN[3], Nasser HADJIPOUR[4]

[1]Department of Anatomical Sciences, School of Veterinary Medicine, Kazerun Branch, Islamic Azad University, Kazerun, Iran
[2]Department of Anatomical Sciences, School of Veterinary Medicine, Science and Research Branch, Islamic Azad University, Tehran, Iran.
[3]Department of Clinical Sciences, School of Veterinary Medicine, Kazerun Branch, Islamic Azad University, Kazerun, Iran
[4]Department of Pathology, School of Veterinary Medicine, Kazerun Branch, Islamic Azad University, Kazerun, Iran

Received: 23.09.2011 • **Accepted:** 07.12.2012 • **Published Online:** 29.07.2013 • **Printed:** 26.08.2013

Abstract: The effects of light exposure on the retina have been carefully considered in recent years. The purpose of the current study was to investigate the effect of flashed light with low and high intensity on the photoreceptor layer of rabbit. Fifteen adult male New Zealand White rabbits were randomly divided into 3 groups: a control group, experimental group I (exposed to 60 W of flashed light 5 times), and experimental group II (exposed to 1000 W of flashed light 5 times). The retinas of the eyes were removed and studied by transmission electron microscope. The photoreceptor layer damage in experimental group II contained outer segment loss, vacuolated and disorganized mitochondria, vacuolization, cell swelling, and pyknotic, karyorrhexis, and karyolytic nuclei. The results suggested that high-intensity flashed light (1000 W) caused more photoreceptor layer damage than low-intensity light (60 W).

Key words: Retina, photoreceptor layer, transmission electron microscope, flashed light, rabbit

1. Introduction

It has been reported that exposure of rabbits to bright light with different intensities causes damage to the photoreceptor layers in the retina (1). In addition, earlier reports by Organisciak et al. (2) suggested that retinal light damage occurs in rats exposed to intermittent light. Further works have suggested that different animals might be injured by various continuous and intermittent light levels (3–9).

Retinal light damage in rats has been shown to be rhodopsin-mediated and a function of retinal irradiance, wave length, and duration of exposure (5,10). In addition, the sensitivity of the retina to light damage varies among species (4,11). The mechanism of the light damage is complicated and closely related to light history (12), nutritional state (10,13,14), genetic factors (7,15), age (16), and temperature (17,18).

The aim of the present study was to investigate the effect of flashed light with low and high intensity on the ultrastructure of the photoreceptor layer of the retina using a transmission electron microscope.

2. Materials and methods

Fifteen male New Zealand White rabbits were obtained from the animal house of Islamic Azad University, Kazerun Branch, Kazerun, Iran. The animals were allowed to adapted to the new environment for 2 weeks before the experiments commenced. The rabbits were randomly divided into 3 groups of 5 each: the control group (CON), experimental group I (EXP-I), and experimental group II (EXP-II).

The animals in the CON group were maintained in environmental conditions without exposure to flashed light, with 12 h/12 h light–dark cycles at an average illumination of 120 lx). The rabbits in the EXP-I and EXP-II groups were exposed to 60 W and 1000 W of flashed light 5 times, respectively. All animals in the experimental groups were allowed to adapt to the dark for 48 h before the flashed light treatment. The intensity of the light as measured by a power meter was 300 lux in EXP-I and 4500 lux in EXP-II. The bulbs were mounted vertically in each box (dimensions 1200 × 1200 × 1200 mm), which stood 700 mm above the floor.

The histological damage due to flashed light was evaluated using a transmission electron microscope. After enucleation of the eye, the cornea, lens, and vitreous body were removed, and the eyes were placed in 4% glutaraldehyde fixative for 4 h. The retina was separated near the optic disc and processed for viewing

* Correspondence: esfandiari.arash@gmail.com

under transmission electron microscope. Thin sections were obtained and examined under a Philips CM-10 transmission electron microscope (Philips, Eindhoven, the Netherlands).

Laboratory care of the animals used in this study was performed in accordance with the Guide for the Care and Use of Laboratory Animals (19).

3. Results

The photoreceptor layer of an eye contains the rod and cone cells. The rod and cone outer segments consist of stacks of discs surrounded by the cell membrane. The inner segments contain several round to oval mitochondria, rough endoplasmic reticulum, and glycogen particles. The outer nuclear layer consists of the perikarya of the rods and cones. Cone nuclei are located nearest to the outer limiting membrane (Figure 1). In our examination, the outer limiting membrane appeared normal and was formed by zonula adherens. The nuclei were round to oval-shaped.

The high-intensity flashed light clearly damaged the photoreceptor layer. The light-damaged photoreceptor layer was identified by loss of outer segments, vacuolated and disorganized mitochondrial crista, cell swelling in the inner segment, and pyknotic, karyorrhexis, and karyolytic nuclei in the outer nuclear layer.

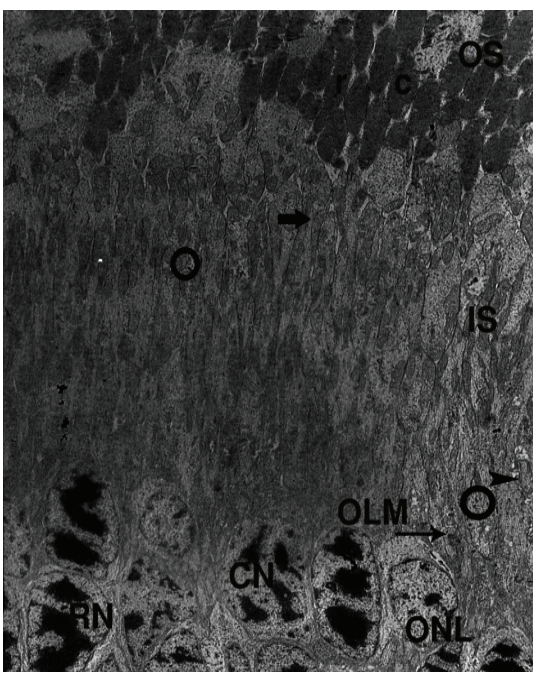

Figure 1. Micrograph of the photoreceptor layer in the control group. c = cone outer segment, r = rod outer segment, OS = outer segment, IS = inner segment, mitochondrium (arrow), glycogen (circles), rough endoplasmic reticulum (arrowhead), OLM = outer limiting membrane (thin arrow), ONL = outer nuclear layer, CN = cone nuclei, and RN = rod nuclei (2950×).

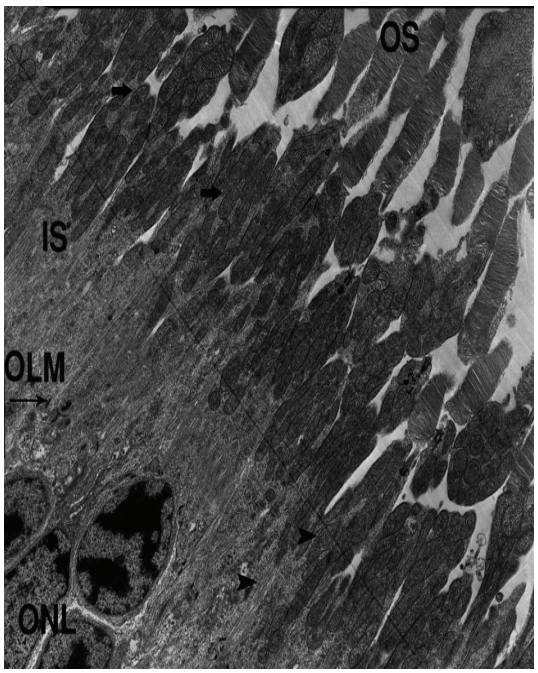

Figure 2. Micrograph of the photoreceptor layer in the low-intensity flashed light group. OS = outer segment, IS = inner segment, mitochondria (arrows), rough endoplasmic reticulum (arrowheads), OLM = outer limiting membrane (thin arrow), and ONL = outer nuclear layer (2950×).

The photoreceptor layers of the rabbits exposed to low-intensity flashed light with a 60 W bulb appeared normal (Figure 2). However, the animals exposed to high-intensity flashed light with a 1000 W bulb had major signs of damage. In the EXP-II group, a loss of outer segments and vacuolated and disorganized outer segments were obvious (Figure 3). Disorganization and loss of mitochondrial crista, extreme vacuolization, and cell swelling were evident in the inner segment (Figure 3). The outer limiting membrane appeared normal. In this group, pyknotic, karyorrhexis, and karyolytic nuclei were seen in the outer nuclear layer (Figure 3).

4. Discussion

In the EXP-II group, the results showed that the outer segments of the photoreceptor layer were disorganized and lost. Coordinated phagocytosis of outer segment shedding by the retinal pigmented epithelium is necessary for maintenance of normal retinal function. New discs are constantly generated at the basal end of the outer segment and old discs are shed from the apical end of the outer segment, where they are digested by the phagocytic retinal pigmented epithelium. Outer segment shedding is impaired and may be spawned by photogenerated radicals due to oxidative modifications (20,21), suggesting that light damage triggers outer segment phagocytosis by the retinal

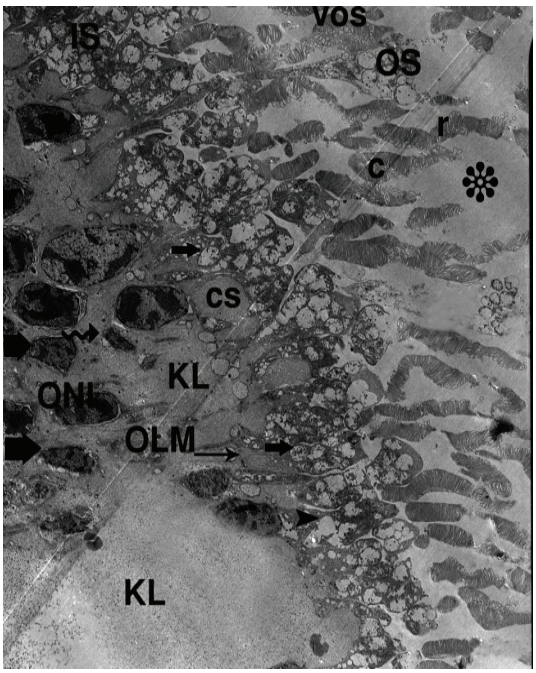

Figure 3. Micrograph of the photoreceptor layer in the high-intensity flashed light group. c = cone outer segment, r = rod outer segment, OS = outer segment, VOS = vacuolated outer segment, loss of outer segment (asterisk), IS = inner segment, CS = cell swelling, abnormal mitochondria which lost crista (arrows), vacuole (arrowhead), OLM = outer limiting membrane (thin arrow), ONL = outer nuclear layer, KL = karyolytic, pyknotic nuclei (thick arrows), and karyorrhexis (waved arrow) (2200×).

pigmented epithelium (21). Therefore, it is reasonable to hypothesize that disorganization and loss of the outer segment may occur in the retina, and that phospholipid peroxidation products may serve as signaling molecules for retinal pigmented epithelium phagocytosis (22). These findings agree with those of Organisciak et al. (2) and Esfandiari et al. (1). Our observation in the EXP-II group indicated that damage including vacuolization, loss of mitochondrial crista, and cell swelling had occurred in the inner segment. In fact, vacuoles are a common response to damage in cells. Furthermore, cell swelling is recognized as vacuolization and decreased specific gravity. This suggests that mitochondrial crista were lost and decreased oxidative phosphorylation and ATP occurred. In addition, ATP deficiency impairs Na–K pumps; therefore, Na and H_2O move into the cells and cell swelling may occur. Pyknotic and karyolytic nuclei resulted in damage to photoreceptor cells. These results are consistent with those of Esfandiari et al. (1) and White and Fisher (7).

In conclusion, the present results demonstrate that high-intensity flashed light not only has a notable effect on the retinal photoreceptor, but also damages this layer.

Acknowledgments

This study was conducted under the sponsorship of the Kazerun Branch of Islamic Azad University, Kazerun, Iran.

References

1. Esfandiari, A., Yousofi, A.R., Dehghan, A., Safavi, A.: Effect of intermittent light on the photoreceptor cells of the retina in rabbits. Jpn. J. Ophthalmol., 2009; 53: 635–639.
2. Organisciak, D.T., Jiang, Y.L., Wang, H.M., Pickford, M., Blanks, J.C.: Retinal light damage in rats exposed to intermittent light. Comparison with continuous light exposure. Invest. Ophthalmol. Vis. Sci., 1989; 30: 795–805.
3. Hansson, H.A.: A histochemical study of cellular reactions in rat retina transiently damaged by visible light. Exp. Eye. Res., 1971; 12: 270–274.
4. Noell, W.K.: Possible mechanisms of photoreceptor damage by light in mammalian eyes. Vision Res., 1980; 20: 1163–1171.
5. Williams, T.P., Howell, W.L.: Action spectrum of retinal light damage in albino rats. Invest. Ophthalmol. Vis. Sci., 1983; 24: 285–287.
6. La Vail, M.M., Gorrin, G.M., Repaci, M.A., Thomas, L.A., Ginsberg, H.M.: Genetic regulation of light damage to photoreceptors. Invest. Ophthalmol. Vis. Sci., 1987; 28: 1043 –1048.
7. White, M.P., Fisher, L.J.: Degree of light damage to the retina varies with time of day of bright light exposure. Physiol. Behav., 1987; 39: 607–613.
8. Braekevelt, C.R.: Photoreceptor fine structure in light- and dark-adaptation in the butterfly fish (*Pantodon buchholzi*). Anat. Anz., 1990; 171: 351–358.
9. Braekevelt, C.R., Smith, S.A., Smith, B.J.: Photoreceptor fine structure in oreochromis niloticus in light- and dark-adaptation. Anat. Rec., 1998; 252: 453–461.
10. Noell, W.K., Albrecht, R.: Irreversible effects of visual light on the retina, role of vitamin A. Science, 1971; 172: 76–79.
11. Lanum, J.: The damaging effects of light on the retina. Empirical findings, theoretical and practical implications. Surv. Ophthalmol., 1978; 22: 221–249.
12. Penn, J.S., Anderson, R.E.: Effect of light history on the rat retina. In: Osborne, N., Chader, G.J., Eds. Progress in Retinal Research. Pergamon Press, Oxford. 1990; 75–98.
13. Organisciak, D.T., Wang, H.M., Noell, W.K.: Aspects of the ascorbate protective mechanism in retinal light damage of rats with normal and reduced ROS docosahexaenoic acid. Prog. Clin. Biol. Res., 1987; 247: 455–468.
14. Koutz, C.A., Wiegand, R.D., Rapp, L.M., Anderson, R.E.: Effect of dietary fat on the response of the rat retina to chronic and acute light stress. Exp. Eye Res., 1995; 60: 307–316.

15. Borges, J.M., Edward, D.P., Tso, M.O.: A comparative study of photic injury in four inbred strains of albino rats. Curr. Eye Res., 1990; 9: 799–803.

16. Osteen, W.K., Anderson, K.V., Shear, C.R.: Photoreceptor degeneration in albino rats: dependency on age. Invest. Ophthalmol., 1974; 13: 334–339.

17. De Lint, PJ., Van Norren, D., Toebosch, A.M.: Effect of body temperature on threshold for retinal light damage. Invest. Ophthalmol. Vis. Sci. 1992; 33: 2382–2387.

18. Organisciak, D.T., Darrow, R.M., Noell, W.K., Blanks, J.C.: Hyperthermia accelerates retinal light damage in rats. Invest. Ophthalmol. Vis. Sci., 1995; 36: 997–1008.

19. Olfert, E.D., Cross, B.M., McWilliam, A.A.: Guide to the Care and Use of Experimental Animals, Canadian Council on Animal Care, 2nd ed., Vol. 1. 1993. Available at: http://www.ccac.ca/Documents/Standards/Guidelines/Experimental_Animals_Vol1.pdf. Accessed 8 September 2011.

20. Young, R.W., Bok, D.: Participation of the retinal pigment epithelium in the rod outer segment renewal process. J. Cell Biol., 1969; 42: 392–403.

21. La Vali, M.M:. Rod outer segment disc shedding in rat retina: relationship to cyclic lighting. Science, 1976; 194: 1071–1074.

22. Watson, C.L., Gold, M.R.: Lysophosphatidylcholine modulates cardiac I (Na) via multiple protein kinase pathways. Circ. Res., 1997; 81: 387–395.