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RESEARCH ARTICLES

Biological role of lutein in the light-induced retinal degeneration☆

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Abstract

Lutein, a xanthophyll of a carotenoid, is anticipated as a therapeutic product to prevent human eye diseases. However, its biological mechanism is still unclear. Here, we show the molecular mechanism of lutein's effect to reduce photodamage of the retina. We analyzed the light-exposed retinas of Balb/c mice given lutein-supplemented or normal diet. Visual function was measured by electroretinogram, and histological changes were observed. Immunohistochemical and immunoblot analyses were performed to analyze molecular mechanism. The reactive oxygen species induced in the retina was evaluated by fluorescent probes. In the mice after light exposure, reduction of a-wave and b-wave amplitudes in electroretinogram, indicating visual impairment, and thinning of the photoreceptor cell layer owing to apoptosis were both attenuated by lutein diet. Interestingly, γ-H2AX, a marker for double-strand breaks (DSBs) in DNA, was up-regulated in the photoreceptor cells after light exposure, but this increase was attenuated by lutein diet, suggesting that DSBs caused by photodamage contributed to the photoreceptor cell death and that this change was suppressed by lutein. Moreover, the expression of eyes absent (EYA), which promotes DNA repair and cell survival, was significantly up-regulated with lutein diet in the light-exposed retina. Therefore, lutein induced EYA for DNA repair, which could suppress DNA damage and photoreceptor cell apoptosis. Lutein reduced light-induced oxidative stress in the retina, which might contribute to promote DNA repair. The lutein-supplemented diet attenuated light-induced visual impairment by protecting the photoreceptor cells' DNA.

Keywords: Light exposure; Retina; Photoreceptor; Visual function; DNA damage; Oxidative stress

1. Introduction

Lutein has been anticipated to prevent age-related changes and/or diseases in various kinds of fields. Nutritional support, including supplemental intake of lutein, is recommended in response to the results from the epidemiological surveys. However, its biological effects *in vivo* remain to be elucidated.

Lutein is a xanthophyll, a kind of carotenoid. It can absorb blue light that has high energy in the visible light, and contains several double bonds that can quench reactive oxygen species (ROS) and reduce oxidative stress. It cannot be synthesized by mammals *in vivo* and is obtained from the diet. Once ingested, lutein is distributed to the retina, skin and so on. In the retina, it is delivered to outer segments of photoreceptor cells and, in humans, is concentrated in the macula. The long-term oral intake of lutein elevates its level in serum [1,2] and increases macular pigment density [1,3], suggesting that constant intake from diet causes the accumulation of lutein in the retina. Lutein is anticipated to prevent retinal diseases, i.e., retinitis pigmentosa, as well as skin diseases which can be accelerated by light exposure [4,5]. Moreover, a large-scale clinical study, the Age-Related Eye Disease Study 2 is now ongoing to test lutein's effects on age-related macular disease (AMD), most probably due to its antioxidative and anti-inflammatory effects. In fact, we have revealed that lutein can act on signal-transduction molecules, as shown in the endotoxin-induced uveitis model, by suppressing STAT3 activation and the subsequent degradation of rhodopsin [6–8] or, as shown in the laser-induced choroidal neovascularization model, by inhibiting NF-kB activation and following inflammatory neovascularization [9]. Lutein also inhibited ERK activation which can be activated by inflammatory factors such as angiotensin II [10], and prevented reduction of synaptic protein in diabetes [11]. Suppression of oxidative stress by lutein is involved in these effects. Thus, lutein can suppress pathogenic signals through reducing ROS. However, the underlying mechanism of lutein's effect in the light-induced retinal degeneration is still obscure.

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Light exposure has long been studied from several aspects. Previous studies have shown that the pathological effects of light stimuli are mediated by a variety of events, including the activation of AP-1 [12,13], excessive influx of calcium ion [14] and damage to mitochondria [13,14] and DNA [15,16], all of which can induce apoptosis.

In this study, we focused on DNA damage as a causative event of cell death after light exposure and investigated the effect of lutein intake. Previous reports showed DNA ladder formation after light exposure by biochemical methods. Here, to examine the DNA damage in photoreceptor cells, we analyzed Ser139-phosphorylated H2AX, γ-H2AX. γ-H2AX is recruited in response to DNA double-strand breaks (DSBs) and disappears after DNA repair [17,18]. It is becoming common to use γ-H2AX as a biomarker for gene instability associated with cancer development and tumor progression [19]. Recent studies have also shown that γ-H2AX can act as either a proapoptotic or antiapoptotic factor, depending on its phosphorylation state [20,21]. Tyr-142-phosphorylated γ-H2AX recruits JNK and other proapoptotic molecules to induce cell death, whereas Tyr-142-dephosphorylated γ-H2AX recruits a set of DNA repair factors to promote cell survival [20,21]. Since the tyrosine phosphatase responsible for γ-H2AX's Tyr-142 dephosphorylation was discovered to be eyes absent (EYA) [20,21], we also analyzed the expression of EYA. Eyes absent was originally reported as a transcription cofactor that determines eye formation during development, and it also has a phosphatase domain that interacts with other cofactors to fine-tune its transcription activity. But recently, knockdown of the *eya* gene was found to increase apoptosis in response to either hypoxia or ionizing radiation, because of the resulting deficiency in DNA repair [21]. Therefore, an interaction of EYA with γ-H2AX can influence cell death/survival. However, whether EYA is involved in the effects of lutein in light-induced photoreceptor cell death has not been reported.

We first evaluated the effect of lutein intake by analyzing visual function and photoreceptor cell apoptosis after light exposure. The analyses of γ-H2AX and EYA elucidate the role of lutein in DNA damage in light-induced retinal degeneration. The effects of lutein in these analyses support the idea that lutein has a potentiality as a useful agent in neuroprotective therapy.

2. Methods and materials

2.1. Animals and laboratory chow

Six-week-old BALB/c male mice were purchased (Clea Japan, Tokyo, Japan) and kept under dim cyclic light (5 lux, 12 h on/off) for 2 weeks before exposure to light at experimental level (described below). Beginning 10 days prior to the light exposure, three groups of mice were established. Two groups were fed standard laboratory powder chow, and one was given the same chow supplemented with 0.1% lutein (provided by Wakasa Seikatsu Co., Ltd., Kyoto, Japan), and the mice were maintained on these diets throughout the study. The dose of lutein taken by a mouse per day was estimated at 170 mg/kg body weight.

All animal experiments were conducted in accordance with the ARVO Statement for the Use of Animals in Ophthalmic and Vision Research.

2.2. Light exposure

Before the light exposure, the mice were dark-adapted by keeping them in complete darkness for 12 h. The pupils were dilated with a mixed solution of 0.5% tropicamide and 0.5% phenylephrine (Mydrin-P; Santen, Osaka, Japan) just before the light exposure. Then, two groups of mice, one fed the control diet and one fed the lutein-supplemented diet, were exposed to 5000 lux of a white fluorescence lamp (FHD100ECW, Panasonic, Osaka, Japan) for 3 h, starting at 9 a.m., in a dedicated exposure box with stainless-steel mirrors on each side of wall and floor. The temperature of the box was maintained with an air conditioner. After the light exposure, the mice were returned to their cages and maintained under dim cyclic light (5 lux, 12 h on/off).

2.3. Electroretinogram

Electroretinogram (ERG) was performed as previously described [7,8]. Responses were differentially amplified and filtered through a digital bandpass filter ranging from 0.313 to 1000 Hz to yield a-waves and b-waves. Light pulses of 800 cd s/m^2 and 4-ms duration were delivered via a commercial Ganzfeld stimulator (Ganzfeld System SG-2002, LKC Technologies, Inc.). The amplitude of the a-wave was measured from the baseline to the trough, and the amplitude of the b-wave was determined from the trough of the a-wave to the peak of the b-wave. The implicit time of the a- and b-waves was measured from the onset of stimulus to the peak of each wave.

2.4. Histology

Eyes were enucleated and fixed in 4% paraformaldehyde overnight at 4°C and embedded in paraffin. The eyes were marked with a 7-0 nylon suture (MANI Inc., Tochigi, Japan) to mark the vertical line. Eyes were sectioned (6 μm) through the vertical meridian including the optic nerve head and stained with hematoxylin and eosin. The outer nuclear layer (ONL) thickness was measured every 300 μm from the optic nerve head to the most peripheral region of the retina using ImageJ software [National Institutes of Health (NIH), Bethesda, MD, USA]. The retinas of 300 μm apart from the disc were shown in the figure.

2.5. Immunohistochemistry

Eyes were fixed with 4% paraformaldehyde and prepared for cryosectioning (8 μm) through the vertical meridian including the optic nerve head. Apoptotic cells were detected using a TdT-dUTP terminal nick-end labeling (TUNEL) kit (Chemicon-Millipore, Billerica, MA, USA). The sections were also incubated with a rabbit antiphosphorylated Histone H2AX antibody (anti-γ-H2AX, 1:200; Trevigen, Gaithersburg, MD, USA) followed by Alexa 568-conjugated goat anti-rabbit IgG, or with a goat anti-EYA3 antibody (1:200; Santa Cruz Biotechnology, Santa Cruz, CA, USA) followed by Alexa 488-conjugated rabbit anti-goat IgG. Nuclei were counterstained with bisbenzimide at a 1:1000 dilution of a 10-mg/ml stock solution (Hoechst 33258; Sigma). All the sections were examined using a microscope equipped with a digital camera (Carl Zeiss, Jena, Germany).

2.6. Immunoblot analyses

Isolated retinas were placed in lysis buffer [10 mM Tris-HCl (pH 7.6), 100 mM NaCl, 1 mM EDTA, 1% Triton X-100, protease inhibitors]. Each sample was separated by sodium dodecyl sulfate polyacrylamide gel electrophoresis and electroblotted onto a polyvinylidene fluoride membrane (Millipore, Bedford, MA, USA). After being blocked in 4% skim milk, the membrane was incubated at 4°C overnight with goat anti-EYA3 (1:200; Santa Cruz) or mouse anti-α-tubulin (1:2,000; Sigma-Aldrich, St. Louis, MO, USA) antibodies. The membrane was then incubated with a biotinylated secondary antibody against goat immunoglobulins followed by avidin–biotin horseradish peroxidase complex (Vectastain Elite ABC Kit; Vector, Burlingame, CA, USA) or a horseradish peroxidase-conjugated antibody against mouse immunoglobulins. The signals were developed using a chemiluminescence kit (ECL Blotting Analysis System; Amersham, Arlington Heights, IL, USA), measured by ImageJ software (NIH), and normalized to α-tubulin.

2.7. Measurement of ROS

Eyes were enucleated and immediately frozen in OCT compound (Sakura Finetek, Torrance, CA, USA). Unfixed cryosections (8 μm) were incubated with BODIPY$^{581/591}$ C11 probe (BODIPY-C11, Invitrogen-Molecular Probes, Eugene, OR, USA) (10 μM) for 30 min at room temperature or with dihydroethidium (DHE; Invitrogen-Molecular Probes, Eugene, OR, USA) (5 μM) for 20 min at 37°C, as previously described [8]. Light emission from BODIPY-C11 converts irreversibly from red to green when a hydroxyradical reacts with polyunsaturated fatty acids. Dihydroethidium specifically reacts with intracellular superoxide anions and is converted to the red fluorescent compound ethidium in nuclei. The sections were examined using a microscope equipped with a digital camera (Carl Zeiss, Jena, Germany), and the intensity of the staining was measured in the retina 300 μm apart from the disc using the ImageJ program (version 1.37; NIH, Bethesda, MD, USA).

2.8. Statistical analysis

Data were expressed as the mean value±standard deviation. Statistical significance was tested by one-way analysis of variance with Tukey's post hoc test or (for ERG data) with Bonferroni's procedure. Differences were considered statistically significant at $P<.05$.

3. Results

3.1. The impairment of visual function induced by light exposure was attenuated by lutein

To determine lutein's effect on visual function, we recorded ERGs (Fig. 1 A–C). In the retinas of light-exposed mice, the amplitudes of the a-wave, which corresponds to photoreceptor function, and of the b-wave, which reflects the electrical activity in the inner retina subsequent to photoreceptor stimulation, were reduced 4 days after

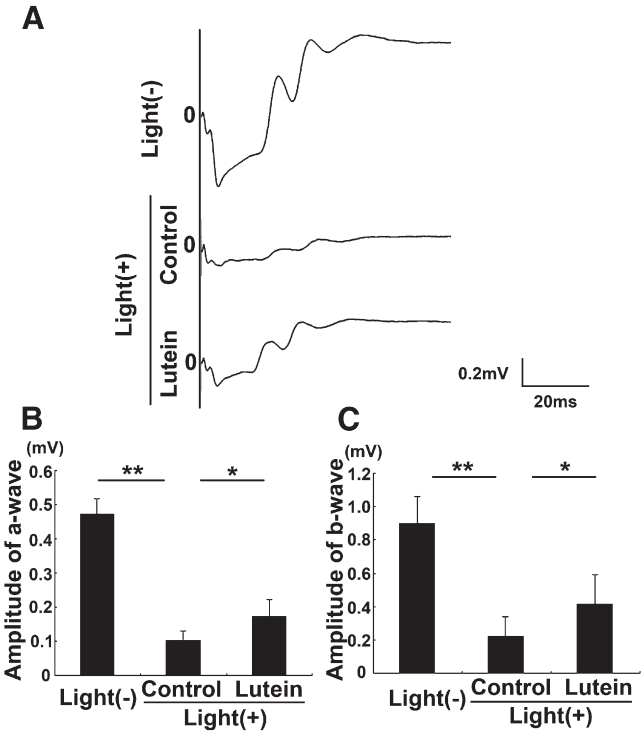

Fig. 1. Light-induced visual function impairment was attenuated by lutein. Electroretinogram (A–C). The reduction in a- and b-wave amplitudes recorded 4 days after light exposure was attenuated by lutein intake. n=5 [light(−), control diet]; n=6 [light(+), control diet]; n=6 [light(+), lutein diet]. *P<.05; **P<.01.

light exposure. However, importantly, the amplitudes of the a- and b-waves after light exposure remained significantly larger in the mice who had been given the lutein-rich diet compared with those given the control diet (P<.05). Light exposure had no effect on the implicit time of either wave, regardless of lutein intake.

Thus, the lutein-supplemented diet attenuated the impairment of photoreceptor cell function after light exposure.

3.2. The reduction in ONL thickness induced by light exposure was suppressed by the lutein

We next compared the residual thickness of the ONL, the photoreceptor cell layer, from the center to the periphery of the retina. Interestingly, the mice who were given the lutein-rich diet had a significantly thicker ONL 7 days after light exposure than light-exposed mice fed a control diet in most regions of the retina (Fig. 2A and B, *P<.05).

Thus, lutein suppressed the light-induced thinning of the ONL, which implied that lutein protected the photoreceptor cells from light-induced neurodegeneration.

3.3. Photoreceptor cell apoptosis was suppressed by lutein

To confirm the protection of photoreceptor cells by dietary lutein supplementation, we counted the apoptotic cells in the ONL, which we identified using the TUNEL assay. The TUNEL-positive cells appeared in the ONL after light exposure in mice fed the control or the lutein-supplemented diet. However, the number of apoptotic photoreceptor cells was significantly lower in the lutein-fed mice 2 days after light exposure (Fig. 3A and B, *P<.05, **P<.01). Thus, the consistent intake of lutein before, during and after light exposure attenuated the extent of light-induced photoreceptor cell apoptosis.

3.4. DNA DSBs preceding apoptosis were reduced by lutein

To analyze whether DNA damage was induced by light exposure, we examined the γ-H2AX expression in the retina by immunohistochemistry. γ-H2AX expression was observed only in the ONL of light-exposed mice 1 day after light exposure. However, its expression was obviously lower in the mice fed the lutein-supplemented diet (Fig. 4A and B, *P<.05, **P<.01). The increase in γ-H2AX expression peaked at 1 day after light exposure and was somewhat reduced by day 2 (data not shown). These results suggested that the constant intake of lutein protected the DNA from DSBs that were detectable within 1 day after light exposure.

3.5. Light-induced EYA expression in the retina was increased by lutein

Next, we investigated whether dietary lutein intake would affect the level of EYA, a critical protein for the DSB repair system, after light exposure. We analyzed EYA3 among the EYA family proteins because previous reports have most clearly shown the contribution to DNA repair of EYA3 [20,21]. Strikingly, EYA3 was expressed only in the ONL after light exposure, and the number of cells expressing EYA3 1 day after light exposure was significantly greater in the mice fed the lutein-rich diet compared with those fed the control diet (Fig. 5A and B, *P<.05, **P<.01). Consistent with these findings, the level of EYA3 was significantly up-regulated in the retina of light-exposed mice fed the lutein-supplemented diet, as shown in the immunoblot analysis of whole retinal lysates (Fig. 5C and D, P<.01).

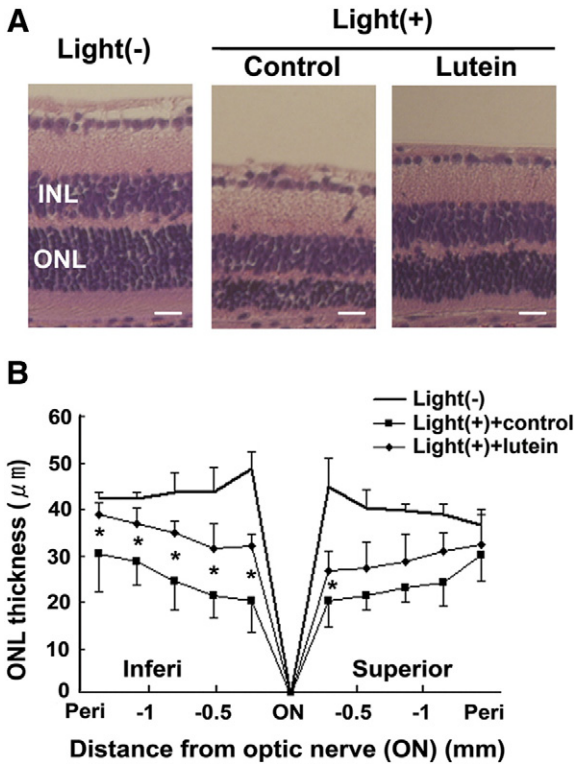

Fig. 2. Light-induced reduction in ONL thickness was suppressed by lutein. Histology (A, B). The reduction in ONL thickness measured 7 days after light exposure was attenuated by the intake of dietary lutein. INL, inner nuclear layer. n=5 [light(−), control diet]; n=7 [light(+), control diet]; n=7 [light(+), lutein diet]. *P<.05. Original magnification, ×200; scale bar, 30 μm.

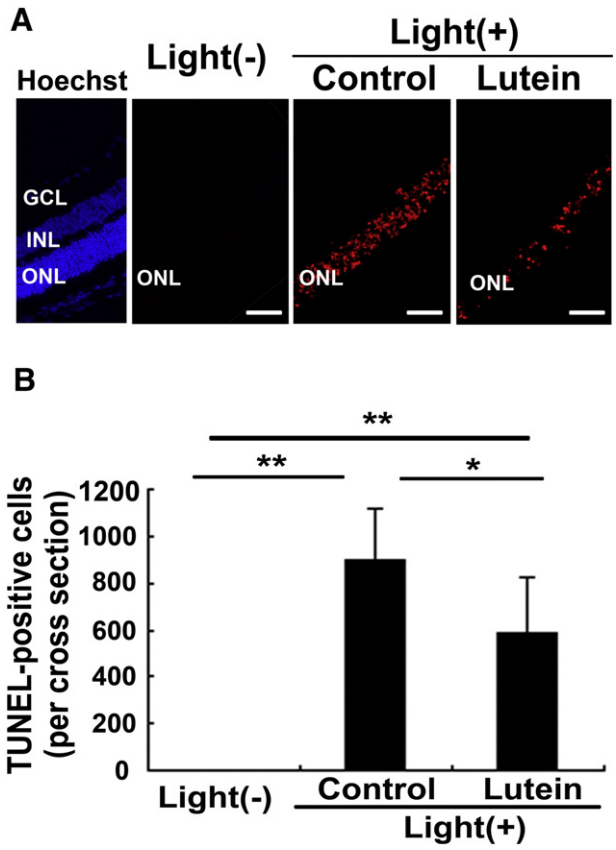

Fig. 3. Light-induced photoreceptor cell death was reduced by lutein. TUNEL staining (A, B). The increase in TUNEL-positive cells 2 days after light exposure was significantly reduced by lutein intake. Nuclear staining was shown as a guide to the retinal layers. GCL, ganglion cell layer; INL, inner nuclear layer. $n=6$ [light(−), control diet]; $n=7$ [light(+), control diet]; $n=8$ [light(+), lutein diet]. *$P<.05$; **$P<.01$. Original magnification, ×200; scale bar, 50 μm.

In mice fed the control diet, increase in EYA3 after light exposure was not clear by this method, most likely because the whole retinal lysate, which includes cells besides photoreceptor cells, was analyzed. As regards EYA3 expression in lutein-fed mice that were not exposed to light, no EYA3 increase was detected in photoreceptor cells by immunohistochemical analysis or by the immunoblot analysis of whole retina lysates (data not shown).

Therefore, lutein intake promoted EYA expression in photoreceptor cells after light exposure.

3.6. Light-induced ROS generation in the retina was suppressed by dietary lutein

We measured the ROS level in retinas 1 h after light exposure using the probe BODIPY-C11, which emits green fluorescence after undergoing lipid peroxidation, a reaction between hydroxyradicals and polyunsaturated fatty acids. The oxidized BODIPY-C11 signal was up-regulated in the outer segments of photoreceptor cells in light-exposed mice fed the control diet compared with the non-light-exposed mice, but this increase was significantly suppressed in the retinas of light-exposed mice fed the lutein-supplemented diet (Fig. 6A and B, *$P<.05$, **$P<.01$). We also measured superoxide anions using DHE which emits red fluorescence when oxidized, and found that the signal from this probe increased in all the retinal layers after light exposure, but that it clearly decreased in mice fed the lutein-supplemented diet (Fig. 6C and D, $P<.05$).

Thus, ROS generation was already up-regulated in the retina by 1 h after light exposure, and the lutein-supplemented diet suppressed this increase.

4. Discussion

We demonstrated the effect of lutein in the light-exposed retina, administered as a daily intake of a lutein-supplemented diet. We first showed that light-induced impairment of visual function and photoreceptor cell loss due to apoptosis were attenuated by lutein. Interestingly, lutein decreased the expression of γ-H2AX which reflects DSBs, and increased the expression of EYA which functions in DNA repair and cell survival. In the retina, elevated ROS level after light exposure was attenuated by dietary intake of lutein.

The ERG findings indicated that visual function after light exposure was impaired, consistent with previous reports [22]. The reduction in a-wave amplitude after light exposure, which reflects photoreceptor cell damage (Fig. 1), was consistent with the finding of photoreceptor cell loss (Fig. 2) through apoptosis (Fig. 3). The reduction in b-wave amplitude, which reflects the neural transduction from the photoreceptor cells, most likely corresponded with the

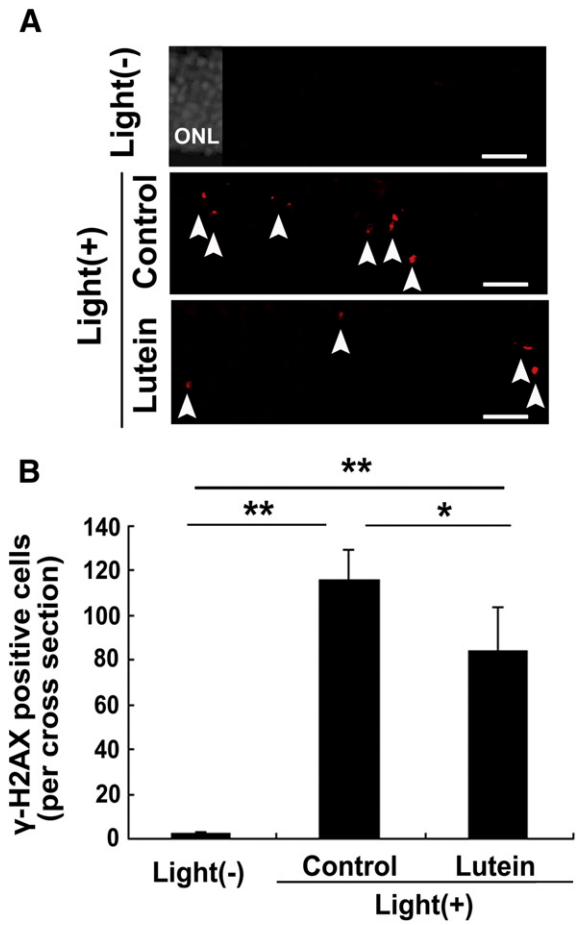

Fig. 4. γ-H2AX expression (and DSBs) was induced in the ONL after light exposure, but was suppressed by lutein. Immunostaining of γ-H2AX in the ONL (A, B). γ-H2AX was up-regulated 1 day after light exposure only in the ONL. The up-regulation was significantly suppressed by dietary lutein intake. Nuclear staining of part of the area was shown at the top panel as a guide to the ONL. $n=5$ for each group: [light(−), control diet]; [light(+), control diet]; [light(+), lutein diet]. *$P<.05$; **$P<.01$. Original magnification, ×400; scale bar, 20 μm.

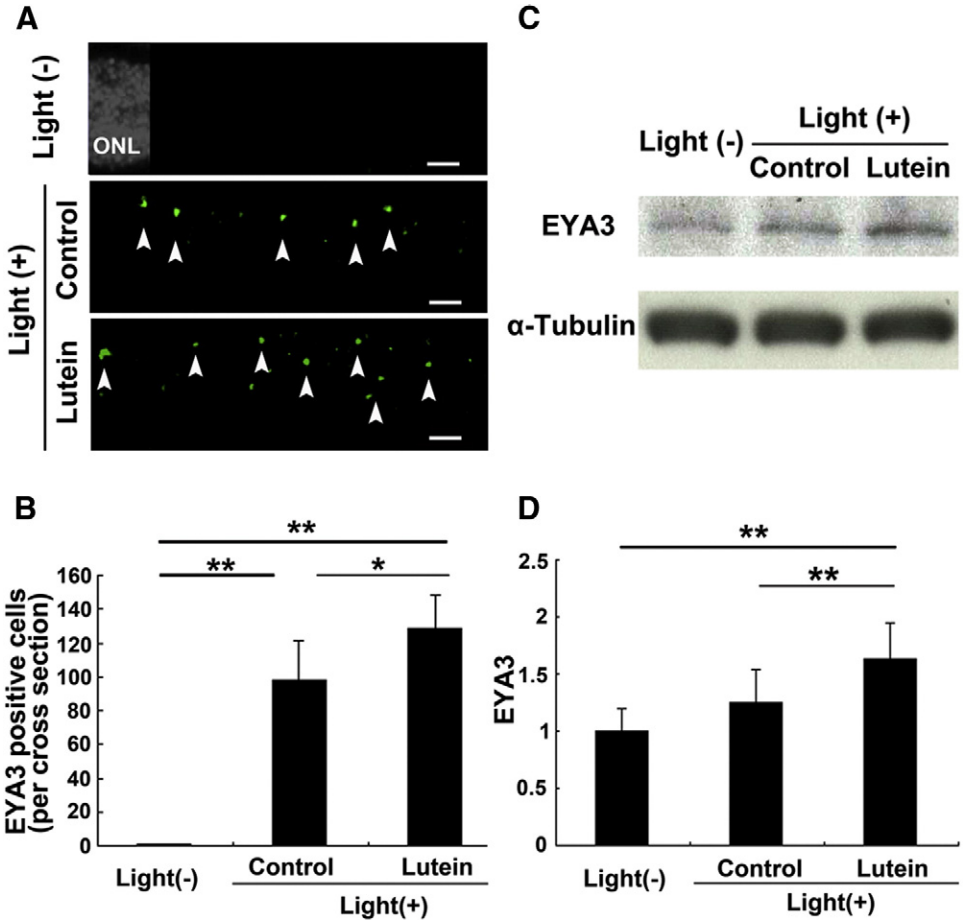

Fig. 5. EYA was expressed in the ONL after light exposure, which was enhanced by lutein. Immunostaining of EYA3 in the ONL (A, B). EYA3 was detected 1 day after light exposure only in the ONL. The EYA3 level was significantly enhanced in mice fed the lutein-rich diet. Nuclear staining of part of the area was shown at the top panel as a guide to the ONL. Immunoblot analysis of EYA3 in whole retina lysates (C, D). The level of EYA3 1 day after light exposure was significantly up-regulated in mice fed the lutein-rich diet compared with mice fed the control diet, with or without light exposure. $n=7$ [light(−), control diet]; $n=7$ [light(+), control diet]; $n=8$ [light(+), lutein diet]. *$P<.05$; **$P<.01$. Original magnification, ×400; scale bar, 20 μm.

degeneration of photoreceptor cells. These changes were attenuated by the intake of lutein. Therefore, these data demonstrated that lutein has a neuroprotective effect in the photoreceptor cells against light-induced retinal degeneration.

Irreversible DNA damage generally results in cell death. Among the patterns of DNA damage, such as DNA single-strand breaks and base damages, DSBs are the hardest to repair since both alleles are damaged simultaneously, and no complete template remains for DNA repair. To detect DSBs after light exposure, we evaluated the expression of γ-H2AX, which is recruited to DSBs immediately after the DNA damage (Fig. 4). In this study, the number of γ-H2AX-positive cells had increased by 1 day after the light exposure, and the number dropped by 2 days after light exposure, when photoreceptor cell apoptosis was at its peak. Thus, the light-induced generation of DSBs resulted in photoreceptor apoptosis by 2 days after light exposure. However, the daily intake of a lutein-supplemented diet suppressed the expression of γ-H2AX, suggesting that lutein attenuated the induction of DSBs after light exposure.

Although γ-H2AX is utilized as a good indicator of DSBs, recent researches reveal that it also contributes to DNA repair when Tyr-142 is dephosphorylated by EYA [20,21]. Interestingly, we found that consistent dietary lutein intake positively regulated EYA expression after light exposure (Fig. 5). Mice supplemented with lutein diet did not show EYA induction without light exposure (data not shown), and EYA was induced only by light exposure. But the level was further increased by lutein which reduced ROS in the retina, suggesting that EYA induced by light stimuli might have been suppressed by ROS after light exposure and did not reach the sufficient level to repair DNA for survival, in the absence of lutein administration. This association between EYA induction and ROS reduction may be an attractive research target in the future.

Light-induced photoreceptor cell degeneration occurs in two phases: an immediate phase and a subsequent, long-lasting phase [15,23,24]. The latter is at least partly explained by the cumulative changes in enzymatic activity that are triggered by light exposure. Previous reports showed that the activation of AP-1, which occurs in the second phase, is involved in light-induced photoreceptor cell death, using *c-fos*-deficient mice. AP-1 is activated by enzymes such as JNK. Since γ-H2AX recruits JNK, AP-1 can be activated by the light-induced DSBs and up-regulation of γ-H2AX. When light-induced DSBs exceed the level of EYA induction after light exposure, Tyr-142 phosphorylation of γ-H2AX and the second-phase reaction may be enhanced, leading to the increase in apoptosis. In contrast, the lutein-rich diet positively regulated the induction of EYA in the first phase. Eyes absent should dephosphorylate γ-H2AX and may suppress AP-1 activation and further cell death in the second phase.

Previous investigations demonstrated that a homozygous knockout of *eya* function in *Drosophila* results in severe embryonic defects and absence of compound eyes. This defect turned out to be through excessive cell death during retinal development [25,26].

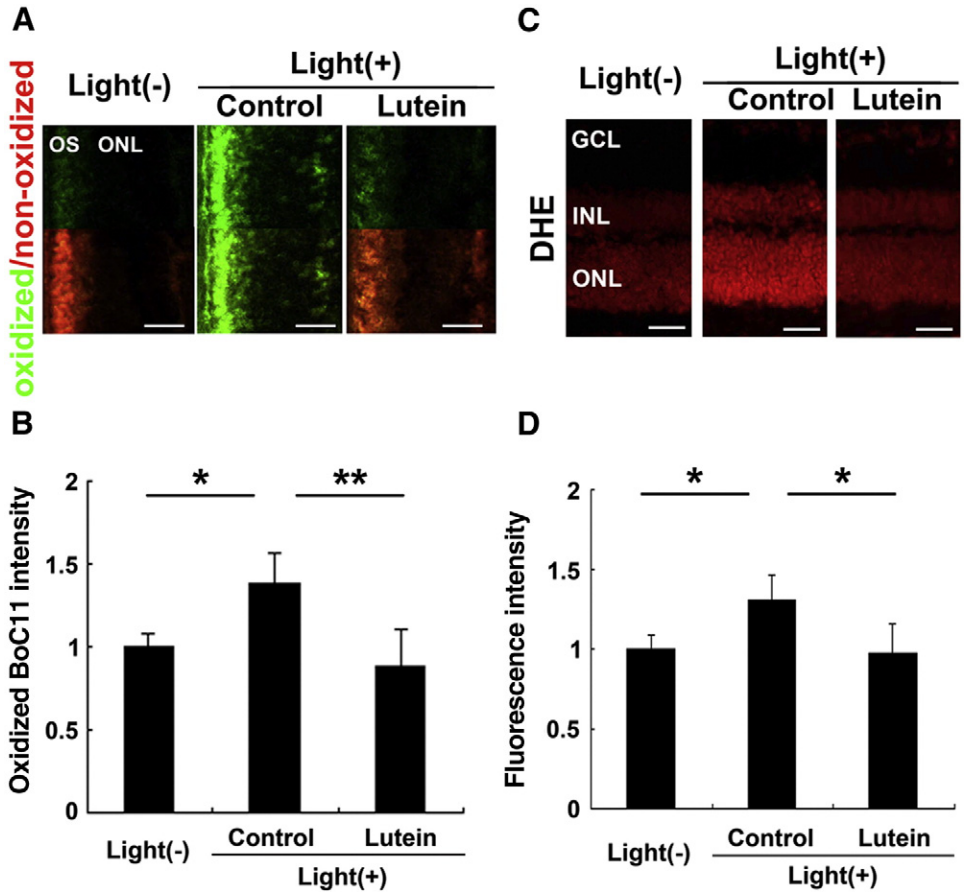

Fig. 6. Light-induced oxidative stress in the retina was suppressed by lutein. The BODIPY-C11 reaction that changes the emission from red to green in the presence of hydroxy-radicals (A, B). Oxidized BODIPY-C11 was up-regulated 1 h after light exposure, mainly in the photoreceptor outer segments (OS) of mice fed the control diet, but this increase was largely suppressed in animals maintained on a lutein-supplemented diet. The DHE reaction showing the generation of the superoxide anion (C, D). Dihydroethidium fluorescence was up-regulated in the whole retina after light exposure of mice fed a control diet, which was mostly prevented by the daily intake of lutein. OS, outer segment; GCL, ganglion cell layer; INL, inner nuclear layer. $n=4$, for each group: non-light-exposed mice fed the control diet [light(−), control diet], light-exposed mice fed the control diet [light(+), control diet] and light-exposed mice fed the lutein-supplemented diet [light(+), lutein diet]. *$P<.05$; **$P<.01$. Original magnification, ×200; scale bar, 30 μm.

These data support the idea of EYA3′s role to protect DNA of the retinal cells.

Lutein accumulates physiologically in the retina, although its level decreases with age in humans [27]. Thus, the age-related decrease in retinal lutein may make the retina more vulnerable to light-induced DNA damage, exacerbating the degeneration of photoreceptor cells with advancing age. The concept of photoaging, now on focus in the field of skin biology [28], can be also proposed in the field of retinal cell biology in terms of DNA instability and reduction of DNA repair ability [29]. This can be, at least in part, relieved by lutein administration. On the other hand, light damage may also occur during ophthalmic surgery, such as cataract surgery and pars plana vitrectomy. Further study on the mechanism of lutein's protective effect may help in the development of a preventive therapy for light-exposure-induced retinal degeneration. Although lutein has been applied as a dietary supplement for chronic diseases, such as AMD, it may have a chance to be involved as a preventive medicine for acute diseases in the future. Moreover, elucidating the molecular mechanism of lutein's effect on light-induced photoreceptor cell apoptosis might also be helpful for analyzing lutein's effect on the photodamage in other organs. In the skin, lutein is believed to protect against edema and hyperplasia after UV exposure [30]. The present study will help understand its molecular mechanism.

In summary, we found that lutein suppressed DSBs and increased EYA after light exposure, suggesting that photodamage increased DSBs by both promoting their occurrence and decreasing their repair. Lutein functioned in the neuroprotection of photoreceptor cells from light-induced DNA damage.

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