INTRODUCTION

Melatonin is a chronobiotic, indoleamine neurohormone, primarily synthesized in the pineal gland, with a marked circadian rhythm that normally peaks in darkness. Most of the body’s circulating melatonin originates in the pineal gland. Melatonin’s major role is that of a photoneuroendocrine transducer, providing information about the length of daylight through its secretion profile. By virtue of its circadian rhythm, melatonin modulates a variety of biological rhythms in the eye. For example, ocular circadian rhythms play roles in postnatal ocular growth, axial elongation, and emmetropization. Under normal visual conditions, ocular elongation in chicks exhibits a pronounced growth rhythm, with maximum growth occurring during the
daytime and minimum growth occurring at night.\textsuperscript{2–4} Rearing chickens in either constant light or constant darkness produces excessive ocular elongation in otherwise normal eyes.\textsuperscript{5–8} These findings suggest that normal ocular growth requires a pattern of diurnal rhythm.

Given that myopia is a very common ocular condition, our knowledge of the mechanisms underlying its development is limited. In recent years, animal models for form deprivation myopia (FDM) and defocus myopia (DM) have been widely used in attempts to understand the underlying mechanisms of this condition. In investigations using different wavelengths of light, it has been demonstrated that the axial length of the developing eyeball is affected by illumination with colored light.\textsuperscript{9} The studies of Kroger et al.\textsuperscript{9} in fish and Chen et al.\textsuperscript{10} in guinea pigs showed that long wavelengths of light cause greater increases in axial length than do short wavelengths.

One study\textsuperscript{11} investigated the relationship between melatonin and myopia, and proposed that melatonin, which acts through specific melatonin receptors (Mel1a, Mel1b, and Mel1c) in retinal and extraretinal ocular tissues, plays a role in ocular growth and development. Several studies have described the distribution of these three melatonin receptors in ocular tissue in chick and Xenopus (frog).\textsuperscript{11–13} In guinea pig, only MT1 (Mel1a) has been shown to be expressed in the retina.\textsuperscript{14}

Melanopsin is a member of the opsin family of photopigments. In mammals, it is expressed in specialized photosensitive retinal ganglion cells involved in the regulation of circadian rhythms, the pupillary light reflex, and various non-visual light responses. There exists a relationship between irradiance and secretion of melatonin.\textsuperscript{1,15–17} Light of 446–480 nm has been shown to be the most potent wavelength for inhibiting production of melatonin.\textsuperscript{18} The peak sensitivity of melanopsin is also \( \sim 480 \text{ nm} \).\textsuperscript{19} Light of \( 480 \text{ nm} \) that is recognized by melanopsin-containing retinal ganglion cells is thus conveyed to the suprachiasmatic nucleus (SCN) to regulate biological rhythms by suppressing melatonin in the pineal gland.\textsuperscript{20–22}

The present study, therefore, was designed to investigate the effects of various wavelengths of light on refractive status, melatonin, MT1 receptor, and melanopsin in guinea pig eyes.

MATERIALS AND METHODS

Animal Housing and Light Conditions

All procedures were performed according to the ARVO statement on Use of Animals in Ophthalmic and Vision Research. Experimental guinea pigs were raised in specially designed cages that possessed sufficient light-emitting diode (LED) tubes in its walls, ceilings, and floors to ensure virtually homogeneous illumination, maximum light intensity, and evenly distributed lighting. The spectral composition of light sources was controlled using three different types of LED light tubes: blue (peak value, 480 nm; half bandwidth, 20 nm), green (peak value, 530 nm; half bandwidth, 30 nm), and white (color temperature, 5,000 K). The spectral sensitivity function of guinea pigs, as assessed by Jacobs and Deegan,\textsuperscript{23} was used to determine irradiance of illumination to ensure that brightness was similar for all guinea pigs. Irradiance in the cages was calibrated with an IL1700 Research Radiometer (International Light Inc., Newburyport, Massachusetts, USA), set to 0.46 W/cm\(^2\) for blue light, 1.05 W/cm\(^2\) for green light, and 0.80 W/cm\(^2\) for white light. Temperature and humidity were maintained at \( 22 \pm 2^\circ\text{C} \) and \( 60 \pm 10\% \), respectively.

Experiment 1: Effects of Blue and Green Light on Light-Induced Inhibition of Melatonin during the Daytime

Forty-eight 10-day-old guinea pigs, obtained from the Animal Breeding Unit of Shanghai Medical College of Fudan University (Shanghai, China), were exposed to blue light, green light, or white light. All guinea pigs were subjected to alternating 12-hr periods of dark and light (lights-on from 8:00 a.m. to 10:00 p.m.). No light filtered into the animals’ rooms during the dark phase. After 10 days, animals were anesthetized with ketamine (50 mg/kg ip) and euthanized with an overdose of ketamine. Pineal glands were removed rapidly, and stored frozen at \(-80^\circ\text{C} \) until used for high-performance liquid chromatography (HPLC) assay for melatonin concentrations. Samples were collected under a dim (5 lux) red light in a completely darkened room during the dark phase. Care was taken to ensure that there were no light leaks.

HPLC

The melatonin used to construct the calibration curve was purchased from Sigma Chemical Co. (St Louis, Missouri, USA). The calibration curve was constructed by analyzing various concentrations of melatonin solution in the range of 50–500 pg/mL. Melatonin concentrations in samples were then measured.

Tissues were homogenized by vortexing with 4.0 mL methylene bichloride. Homogenates were centrifuged at \( 240 \times g \) at \( 4^\circ\text{C} \) for 10 min, after which the supernatants were removed. Residues were evaporated under vacuum, and 0.2 mL of the mobile phase was added to...
the dried residues. The mobile phase consisted of 50\% acetonitrile (v/v) and was pumped at the low rate of 0.5 mL/min at 30°C. Samples with mobile phase were injected into an HPLC system equipped with an Agilent Zorbax Oligo 5 μm column (80 × 6.2 mm i.d.) and fluorometric detector (Waters, New York, New York, USA), the latter of which utilized excitation and emission wavelengths of 285 and 350 nm, respectively.

Experiment 2: Changes in Expression of Melanopsin and MT1 Receptor during Induction of Myopia by 530 nm Monochromatic Light

Thirty 10-day old guinea pigs, each weighing 100–150 g, were obtained from the Animal Breeding Unit at Shanghai Medical College of Fudan University (Shanghai, China) and were randomly assigned to one of three groups: 530 nm green-light (n = 10), 480 nm blue-light (n = 10), or white-light (n = 10) groups. Animals in each group were exposed to green, blue, or white light for 8 weeks. All guinea pigs were raised according to the standards and illumination times described earlier.

Observation of Ocular and Refractive Development

Biometric measurements were made by a research optometrist during the illumination cycle (daytime). The optometrist was masked with regard to the identity of the treatment group. Specific parameters (retinoscopy and ultrasonography) were recorded before and after experimentation.

Retinoscopy. Retinoscopy was performed three times in all animals by the same optometrist—accuracy: 0.50 diopters (D)—in a dark room using a streak retinoscope and trial lenses. One drop of topical compound tropicamide eye drops (Santen Pharmaceutical Co., Osaka, Japan) was administered four times at 10-min intervals to completely dilate the pupil. Refraction was recorded as the mean value of the horizontal and vertical meridians.

Ultrasonography. A-scan ultrasonography (Opticon Hiscan A/B, OPTICON, Rome, Italy) was used to measure the vitreous and axial lengths of each eye. Topical anesthetic was administered along with 0.4% oxybuprocaine hydrochloride (Santen Pharmaceutical Co.) prior to ultrasound. Each of the axial components recorded was the mean of five repeated measurements (accuracy: 0.01 mm).

There were no significant differences among the three groups for any of the biometric measurements made before light exposure (p > 0.05). Refractive status, ocular length, and vitreous depth were compared at different time points, both within the same group and among the three groups, using one-way analysis of variance (ANOVA) with Bonferroni correction and SPSS Version 11.5 software (SPSS Inc., Chicago, Illinois, USA). Both intra- and inter-group differences were defined as significant at p < 0.05 and highly significant at p < 0.01.

Immunohistochemistry

Immunohistochemistry was used to identify the locations of melanopsin and MT1 receptor. At the end of the treatment period, eyes were removed during the daytime, punctured at the corneal limbus, and soaked overnight at 4°C in paraformaldehyde. After removal of the anterior pole, the remainder of the eye was first soaked overnight at 4°C in 20\% sucrose solution and then overnight in 30\% sucrose solution. To observe melanopsin and MT1 receptor immunocytochemically, tissues were embedded in optimum cutting temperature compound, after which 15 μm serial sections were cut on a cryostat at –20°C. After drying for 2 hr at room temperature, sections were rinsed three times (5 min each) in 0.01 M phosphate-buffered saline (PBS) and then incubated in 0.4\% Triton X-100 (Sigma, St. Louis, Missouri, USA) for 10 min. After sections had again been rinsed three times (5 min each) in 0.01 M PBS, they were incubated in 10\% normal goat serum (Sigma-Aldrich, St Louis, Missouri, USA) for 30 min at 37°C. Primary antibody—1:500 goat anti-mouse melanopsin (SC26959, Santa Cruz Biotechnology, Santa Cruz, California, USA) or 1:500 rabbit anti-mouse MT1 receptor (AbMax Biotechnology Co., Beijing, China)—was placed on the sections, which were then incubated overnight at room temperature in a humidified chamber. Sections were then rinsed three times (10 min each) in PBS to remove any unbound primary antibody. Following this, secondary antibody—rhodamine-conjugated 1:100 goat anti-rabbit IgG (AP132R, Chemicon International Inc., Temecula, California, USA)—was applied for 2 hr at 37°C in a light-protected setting. Immunolabeled sections were rinsed three times (10 min each) in PBS in a dark chamber, mounted on slides with phosphoglycerol, and viewed on a fluorescence microscope (Leica DMR, Leica Microsystems Inc., Wetzlar, Germany).

Real-Time Quantitative Reverse Transcription–Polymerase Chain Reaction (RT–PCR)

Total RNA was isolated during the daytime from each retina and sclera using trizol reagent (Invitrogen Life Technologies, Carlsbad, California, USA), following
sonication. RNA was treated with DNase1 to remove any traces of genomic DNA. First-strand cDNA was synthesized from 2 µg of each RNA sample using oligo(dT) and MMLV reverse transcriptase (Promega, Madison, Wisconsin, USA), according to the manufacturer’s protocol. Each set of samples was processed simultaneously for RNA extraction, DNase1 treatment, cDNA synthesis, and PCR. RT–PCR were performed, and results were analyzed using the 7000 RT-Time PCR System (Applied Biosystems, Carlsbad, CA, USA). β-Actin was used as the internal standard. Briefly, the primers used were as follows: for guinea pig melanopsin, forward 5’-TGGCAGTCAGCGACTTCTT-3’ and reverse 5’-TGTAGAGGCTGCTGCGAA-3’; for guinea pig β-actin, forward 5’-CTCCCTATGCCAACACAGTG-3’ and reverse 5’-GTAACCTCTGGCTCTTGATCC-3’; and for guinea pig MT1 receptor, forward 5’-CAACCCTGCAAACCGGAAC-3’ and reverse 5’-GACGGACTGGTGAAAGGTACA-3’. Reaction mixtures (50 µL) consisted of serially diluted samples containing 5 µL of MgCl₂, 5 µL of 10× conventional PCR buffer, 50 pM of each primer, 1 µL of each dNTP (10 mM) (TaKaRa Biotechnology Co., Dalian, China), and three units of Taq DNA polymerase (Promega). Cycling parameters were 95°C for 2 min to activate Taq DNA polymerase, then 40 cycles at 94°C for 20 sec, 60°C for 20 sec, and 72°C for 30 sec. After each cycle, fluorescence of the accumulated product was assessed for melanopsin, β-actin, and MT1 receptor. To confirm the specificity of the PCR products, melting curves were determined using ABI Prism 7000 SDS v1.1 software (ABI, Carlsbad, CA, USA), and samples were run on an agarose gel. ABI Prism 7000 SDS v1.1 software was then used to detect and quantify concentrations of melanopsin and MT1 receptor in samples.

**Western Blot Analysis**

After tissue samples were divided into several portions, they were frozen and lysed in 500 µL of phenylmethylsulfonyl fluoride-containing PBS. Lysates were sonicated and centrifuged at 14,000 × g for 15 min at 4°C. Supernatants were collected, and the protein concentration of each lysate determined using the Bradford assay (Bio-Rad, Hercules, California, USA). Total protein (30 µL) was applied to each lane of 12% sodium dodecyl sulfate polyacrylamide gels. After electrophoresis and immunoblotting, the polyvinylidene difluoride membranes (Millipore, Billerica, Massachusetts, USA) were washed in Tris-buffered saline containing 0.1% Tween 20 and then incubated with one of the following primary antibodies: anti-actin, anti-melanopsin, or anti-melatonin receptor. Membranes were then incubated with secondary antibody: goat anti-rabbit IgG or rabbit anti-goat IgG antibody. Immunoreactive proteins were detected using a chemiluminescent substrate (Tanon GIS-2008, Tanon Science & Technology Co., Shanghai, China). β-Actin was used as the internal standard. Melanopsin, MTR-1A, and β-actin protein were distinguished in the different membranes.

**RESULTS**

**Experiment 1: Effects of Blue and Green Light on Light-Induced Inhibition of Melatonin during Daytime**

Quantification of Melatonin in the Pineal Gland

The concentration of melatonin in the pineal gland was higher in the green-light group than in either the blue- or white-light groups, regardless of the time of day or night. Concentrations of melatonin in the blue-light group were the lowest among the three groups. Concentrations of melatonin clearly fluctuated between day and night in each group (Table 1).

**Experiment 2: Changes in Expression of Melanopsin and MT1 Receptor with Induction of Myopia by 530 nm Monochromatic Light**

Refractive Changes

After 8 weeks of illumination, eyes in the green-light group were ~2 D more myopic (1.37 ± 0.75 D [mean ± SD]) than eyes in either the blue- (3.25 ± 0.78 D) or white-light groups (3.10 ± 0.84 D; F = 17.46, p = 0.0032). The refractive change in the green-light group was the greatest of any group, while the refractive changes in the blue- and white-light groups were relatively small. Guinea pigs in the green-light group had a relatively myopic refractive status.

**Axial Dimensions**

After light treatment, eyes in the green-light group had an axial length of 8.46 ± 0.13 mm, which was ~0.9 mm greater than that of eyes in either the blue-light (7.56 ± 0.18 mm) or white-light (7.62 ± 0.17 mm) groups (F = 20.86, p = 0.0020). The increase in axial length of eyes in the green-light group was the most pronounced.

**TABLE 1** Pineal gland concentrations* of melatonin in the three treatment groups

<table>
<thead>
<tr>
<th>Time of day</th>
<th>Green-light group</th>
<th>Blue-light group</th>
<th>White-light group</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>10 a.m.</td>
<td>85.2 ± 2.79</td>
<td>45.92 ± 1.74</td>
<td>53.32 ± 2.51</td>
<td>0.0000</td>
</tr>
<tr>
<td>10 p.m.</td>
<td>118.15 ± 4.23</td>
<td>63.27 ± 1.43</td>
<td>73.12 ± 2.72</td>
<td>0.0000</td>
</tr>
<tr>
<td>p-value</td>
<td>0.0000</td>
<td>0.0000</td>
<td>0.0000</td>
<td>—</td>
</tr>
</tbody>
</table>

*Concentrations are reported in pg/ml ± SD.
of any group, as was the vitreous depth: $3.70 \pm 0.16$ mm (green-light group), $3.24 \pm 0.21$ mm (white-light group), and $3.21 \pm 0.19$ mm (blue-light group) ($F = 11.04$, $p = 0.0098$) (Figure 1).

**Immunohistochemistry**
Melanopsin and MT1 receptor were detected immunohistochemically in all cases. Melanopsin-immunolabeled cells were found in the retinal ganglion cell layer of all three groups (Figure 2). Labeled cells were more densely distributed in the retinal ganglion cell layer of eyes in the blue-light group, relative to eyes of either the green- or white-light groups (Figure 2C). The numbers of stained cells were fewest in the eyes of the green-light group, which also had the lowest density and fluorescence of cells among the three groups after 8 weeks of treatment (Figure 2B).

MT1 receptor-positive cells were found in the retinal ganglion cell and inner nuclear layers of eyes in all three groups (Figure 3). The distribution of stained cells in the blue-light group (Figure 3C) was similar to that of the white-light group (Figure 3A). Also, in the blue-light group, the fluorescent intensity of stained cells in the ganglion cell and inner nuclear layers was slightly reduced compared to that of the green- or white-light groups. Labeled cells were more densely distributed and more intensely fluorescent in the ganglion cell and inner nuclear layers of eyes in the green-light group (Figure 3B) compared to eyes of either the blue- or white-light groups.

**Expression of Melanopsin mRNA in Retina**
Relative melanopsin mRNA levels in retinas of the white-, blue-, and green-light groups after 8 weeks were $0.3257 \pm 0.0228$, $0.3273 \pm 0.0142$, and $0.2521 \pm 0.0251$, respectively. Expression of melanopsin mRNA in the green-light group was clearly lower than that of the white-light group (Figure 4A). After 8 weeks, relative MT1 receptor mRNA levels in retinas of the white-, blue-, and green-light groups were $0.0839 \pm 0.005$, $0.0821 \pm 0.004$, and $0.0979 \pm 0.003$, respectively. Expression of retinal MT1 receptor mRNA was clearly higher in the green-light group than in the white-light group ($F = 0.7945$, $p = 0.000$). Expression of retinal MT1 receptor mRNA was slightly lower in the blue-light group than in the white-light group, but the difference was not statistically significant ($F = 2.1637$, $p = 0.092$) (Figure 4C).

Expression of **MT1 Receptor mRNA in Retina and Sclera**
After 8 weeks, relative MT1 receptor mRNA levels in scleras of the white-, blue-, and green-light groups were $0.1607 \pm 0.0013$, $0.1543 \pm 0.0139$, and $0.5529 \pm 0.0024$, respectively. Expression of scleral MT1 receptor mRNA was clearly higher in the green-light group than in the white-light group ($F = 5.3987$, $p = 0.000$). Expression of scleral MT1 receptor mRNA in the blue-light group was slightly lower than that of the white-light group, but the difference was not statistically significant ($F = 7.0065$, $p = 0.649$) (Figure 4B).

![Comparison of refraction, axial length, and vitreous chamber depth](image)

**FIGURE 1** Refraction, axial length, and vitreous length in guinea pigs irradiated by white, green, or blue light for 8 weeks. Representative sections from each group are shown for (A) eyes illuminated with white light, (B) eyes illuminated with 530 nm green light, and (C) eyes illuminated with 480 nm blue light. Melanopsin-positive cells are stained red. (D) Eye treated without primary antibody. Stained cells were more densely distributed and more intensely fluorescent in the retinal ganglion cell and inner nuclear layers of eyes in the blue-light group (Figure 2B) than those in the green-light (Figure 2C) or white-light (Figure 2A) groups.

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Levels of Melanopsin Protein in Retina

Western blot analysis showed that changes in the levels of melanopsin protein in the retina differed according to illumination at different wavelengths of light. The level of melanopsin in green-light-irradiated retinas was only 59.67 ± 13.72% that of white-light-irradiated retinas, while the level of melanopsin in blue-light-irradiated retinas was 107.07 ± 3.77% that of white-light-irradiated retinas ($p = 0.004$) (Figure 5).

Levels of MT1 Receptor Protein in Retina and Sclera

Changes in the levels of MT1 receptor in both the retina and sclera differed according to illumination at different wavelengths of light. The level of MT1 receptor in green-light-irradiated retinas was 164.84 ± 3.23% that of white-light-irradiated retinas, while the level of MT1 receptor in blue-light-irradiated retinas was only 79.36 ± 1.2% that of white-light-irradiated retinas ($p < 0.001$) (Figure 6A). Expression of MT1 receptor in sclera was similar to that in retina. The level of MT1 receptor in green-light-irradiated scleras was 181.53 ± 15.31% that of white-light-irradiated scleras, while the level of MT1 receptor in blue-light-irradiated scleras was only 83.23 ± 4.34% that of white-light-irradiated scleras ($p = 0.012$) (Figure 6B).

DISCUSSION

In almost all species studied, melatonin levels peak during nighttime and are lowest during daytime. A previous study also demonstrated a dose–response relationship between light irradiance or light intensity, and suppression of melatonin. Therefore, melatonin indirectly acts as a marker of bioclock sensitivity. The experiments reported in the present study were designed to investigate the effects of different wavelengths of light on secretion of melatonin in the pineal gland.
In recent years, myopia, in terms of FDM and DM, has been studied in animal models. In several animal species, experiments have demonstrated that both FDM and DM in neonates produce a breakdown of the emmetropization process, resulting in axial elongation and myopia. In our study, eyes exposed to 530 nm green light for 8 weeks were rendered more myopic than those of the other groups. The myopization induced by green light results from increased axial elongation of the vitreous chamber, since both the vitreous depth and ocular length of eyes in the green-light group increased at a significantly greater rate than eyes in the white- or blue-light groups (Figure 1, Table 1). It is clear that axial myopia can be induced by long-wavelength green light. Although a relationship between color sense and myopia is not generally accepted, our study demonstrates that long-wavelength green light induces greater axial myopia than does short-wavelength blue light.

In previous experiments of FDM and DM, several neurotransmitters, e.g., dopamine and retinoic acid, and growth factors, e.g., basic fibroblast growth factor (bFGF) and transforming growth factor (TGF)-β, were considered to be associated with the development of myopia.\(^{27-30}\) In our study, we observed that 530 nm of green light was able not only to induce greater axial myopia, but also to increase secretion of melatonin. Evidence from human and animal model studies indicates that ocular circadian rhythms play roles in postnatal ocular growth, axial elongation, and emmetropization. This evidence also indicates that alterations in these rhythms, or in the phase relationships between them, affect development of and recovery from myopia.\(^{2,8,31}\) Rada’s study\(^{31}\) indicated that systemic administration of melatonin causes significant elongation of the vitreous chamber in healthy and form-deprived chicken eyes, and reduces the thickness of the choroid in form-deprived chicken eyes; all of these anatomic changes can lead to myopia. It has been suggested that melatonin, acting through specific melatonin receptors in retinal and extra-retinal ocular tissues, plays a role in ocular growth and development.

A previous study\(^{14}\) of guinea pig retina showed that the most intense immunoreactivity for the MT1 receptor was in ganglion cell bodies. Subpopulations of amacrine cells, the inner plexiform layer, and the outer plexiform layer also exhibited moderate-to-extreme immunolabeling. Results of the present study indicate that the MT1 receptor subtype is expressed mainly in the retina ganglion cell and inner nuclear layers of guinea pig retina. Moreover, the extent of the MT1 receptor mRNA transcription and protein expression changed according to different concentrations of melatonin. In the green-light group, MT1 receptor mRNA transcription and protein expression clearly increased with increasing concentrations of melatonin. In the blue-light group, mRNA transcription and protein expression decreased along with a slightly decreased concentration of melatonin. Expression of membrane MT1 receptor suggests that it mediates
the effects of melatonin on ocular growth according to the wavelength of light. However, other pathways through which melatonin might act should also be considered. For example, there is a class of nuclear melatonin receptors (ROR/RZR), associated with melatonin signaling, that appear to act as transcription factors directly regulating gene expression. Such widely distributed nuclear melatonin receptors may be primary targets for membrane melatonin receptors or, perhaps, melatonin may bind to them directly to influence gene expression.

As described above, melanopsin, which plays a role in the regulation of melatonin, is sensitive to blue light and insensitive to long-wavelength light. In our study, we also observed changes in retinal melanopsin according to exposure to different wavelengths of light. Levels of melanopsin in green-light-irradiated retinas clearly decreased, while levels of melanopsin in blue-light-irradiated retinas increased. A previous study revealed that ~74% of the retinal ganglion cells that project into the SCN express melanopsin. In mammals, a number of diurnal rhythms, such as the sleep–wake state, movement, feeding, drinking, and endocrine expression, are regulated by the SCN in the hypothalamus. Rhythms of the SCN are affected by several zeitgebers, the strongest of which is light. Blue-light signals are perceived by melanopsin-containing retinal ganglion cells. This information is conveyed to the SCN through the retino-hypothalamic tract to regulate biological rhythms by suppressing pineal gland melatonin. With exposure to monochromatic green light, such suppression is weakened, and the level of pineal gland melatonin increases as a result of this lack of an appropriate light stimulus to melanopsin-containing retinal ganglion cells.

The present study describes changes in refractive status, melatonin, melanopsin, and MT1 receptor in guinea pigs irradiated by various wavelengths of light. We found that 530 nm of green-light illumination induced a significant myopic shift, as well as vitreous chamber and axial elongation. These changes are associated with increased melatonin secretion in the pineal gland, increased MT1 receptor expression, and decreased melanopsin expression in retina. The mechanism(s) underlying these changes is not yet clear. Moreover, further investigations are needed to clarify the mechanism(s) of long-wavelength light-induced myopia.

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REFERENCES
