Proceedings volume 10858, Ophtalmic technologies XXIX, 1085813 (2019) ; Proceedings of SPIE BiOS, 2-7 February 2019, San Francisco, California, United States, which should be cited to refer to this work.
DOI: https://doi.org/10.1117/12.2511654

Device for silent substitution excitation of melanopsin for human eye

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Abstract

Research in neurobiology has identified a new ocular photoreceptor (melanopsin or ipRGC) which mediates a variety of light-based, non-visual effects on human physiology. One way to isolate the stimulation of ipRGCs is the silent substitution technique. We have built a Maxwellian view device capable of 85% ipRGCs contrast excitation with a large FOV (52°). Four modulated LED light sources, illuminate a diffusing sphere, which exit aperture is imaged into the pupil of the eye. A camera with a 900 nm illumination capture the pupil.

Without luminance changes (510 ± 2 lm/m^2), we increased ipRGC excitation from low to high level on three subjects. We observed a pupil constriction increasing with the ipRGC contrast. This suggests that we excite melanopsin silently. However, further experiments with electrophysiological and pupil recording needs to be done to completely validate our silent substitution device.

Introduction

Research in neurobiology has identified a new ocular photoreceptor with a unique photopigment, melanopsin. This photoreceptor is an intrinsically photosensitive retinal ganglion cell (ipRGC) which mediates a variety of light-based, non-visual effects on human physiology depending on the illumination environment.[1]

Isolated stimulation of the ipRGC on humans is difficult due to overlapping spectral sensitivity with other ocular photoreceptors (rods and cones). The silent substitution is a technique that permits the modulation of one photoreceptor type at a time, maintaining the rest at a steady state. By exploiting the metamers properties, it is possible to stimulate the melanopsin photopigment while maintaining a constant level of cone excitation.

Silent substitution technique

The basis of melanopsin light stimulation is the silent substitution technique[2] which is linked to *black metamers* only differentiated by melanopsine photodetectors. Considering that the eye has 5 specific types of photosensitive cells (L, M and S cones, rods and ipRGCs), the light stimulus can be considered as a 5-dimensional vector. Assuming that only the **photopic** range of illumination activates melanopsin[3] so that the rods are deactivated,[4] we can restrict the light stimulus vector to 4-dimension. In order to modulate the excitation of ipRGCs, at least four different light sources with different colors are required. This approach considers that any light stimulus is composed of a fundamental colored stimulus and a *metameric black* which excites only the melanopsin component of ipRGC photoreceptors.

With $S_p(\lambda)$ being the sensitivity of the photoreceptor p and $I_q(\lambda)$ the q^{th} light source intensity, the photoreceptor stimulation PS_{pq} is given by :

$$PS_{pq} = \sum_i [S_p(\lambda_i) \cdot K_p] \cdot [I_q(\lambda_i) \cdot LR_q] \tag{1}$$

where LR_q is the luminance ratio of the light source $I_q(\lambda)$ and K_p a factor for converting power into photometric units. K is well defined for the L and M (683) cones, but not for the S cone and ipRGCs, as only L and M contribute to the luminance.[5] Applying this to all photoreceptors for all illuminations, equation (1) becomes

$$PS = P2C \cdot LR \tag{2}$$

where $P2C$ is the matrix with the elements calculated as follows:

$$P2C_{pq} = \sum_i S_p(\lambda_i) \cdot I_q(\lambda_i) \tag{3}$$

This means that the 4-dimensional space of the light sources is projected through the $P2C$ matrix into the 4-dimensional space of the photoreceptors. Note that any pair (PS, LR) has a physical meaning only if all components of LR are positive. For the silent substitution, we set the photoreceptor stimulation vector PS and get the luminance ratio vector LR of the light source by the following equation

$$LR = P2C^{-1} \cdot PS \tag{4}$$

On the basis of these calculations, a sequence of *black metamers* is produced. This induces a signal whose maximum contrast for ipRGC excitation depends on the spectrum of the light sources.

Existing devices

Basically, such devices are based on two types of optical arrangements: Maxwellian view in which the pupil of the instrument (which could be an artificial pupil) defines the position of the eye and Newtonian or natural view in which only the eye and its own pupil is used. For Maxwellian view, the object plane is usually at a distance corresponding to the resting position of the lens of the eye. For Newtonian view, the object plane is at least 20 cm away from the corneal surface of the eye.

Reference	FOV, pattern	λ [nm]	C
Maxwellian view			
[6–8]	30°, c. obsc. 10.5°	456, 487, 540, 592, 633	17%
[9]	27.5°, c. obsc. 5°	up to 56 λ	50%
Newtonian view			
[10]	2 rings	468, 524, 599, 633	4%
[5]	20°, homegeneous	470, 525, 500, 670	53%
[11]	homogeneous	466, 514, 590, 634	-
[12]	18.9°, homogeneous	468, 507, 593, 633	53%
[13]	95°	470, 510, 595, 635	11%
[14]	28.1°	468, 508, 593, 633	20%
[15]	25° × 12°	465, 500, 515, 595, 635	23%
[16]	1° or 20° homogeneous	447, 470, 505, 530, 590, 627	-
[17]	19°, homogeneous	460, 525, 635, 445/555	19%
[18]	homogeneous	447, 472, 502, 523, 594, 637, 656	21%

Table 1. Summary of the main instrumentation reference values for the excitation of ipRGC photoreceptors. FOV is the field of view and C the maximal ipRGC contrast which can be produced.

We found 14 articles (see table 1) in the peer-review scientific literature since 2010 dealing with silent substitution and melanopsin excitation. We also found more articles which are not aimed at melanopsin ganglion cell excitation but described interesting instrumentation for silent substitution technique, one describing a true retinal projection.[19]

Levels of illumination are not easy to compare because scholars report either lumen per square centimeter (lm/cm^2) or troland (Td), but do not give the necessary information to convert one to the other. Given values are either between 300 to 2500 lm/cm^2 or between 300 to 30000 Td but mostly around 3000 Td. Light stimuli

are usually presented homogeneously over a given area of the retina, either in an annulus or in a given circle centered on the fovea or outside of the fovea.[5–18]

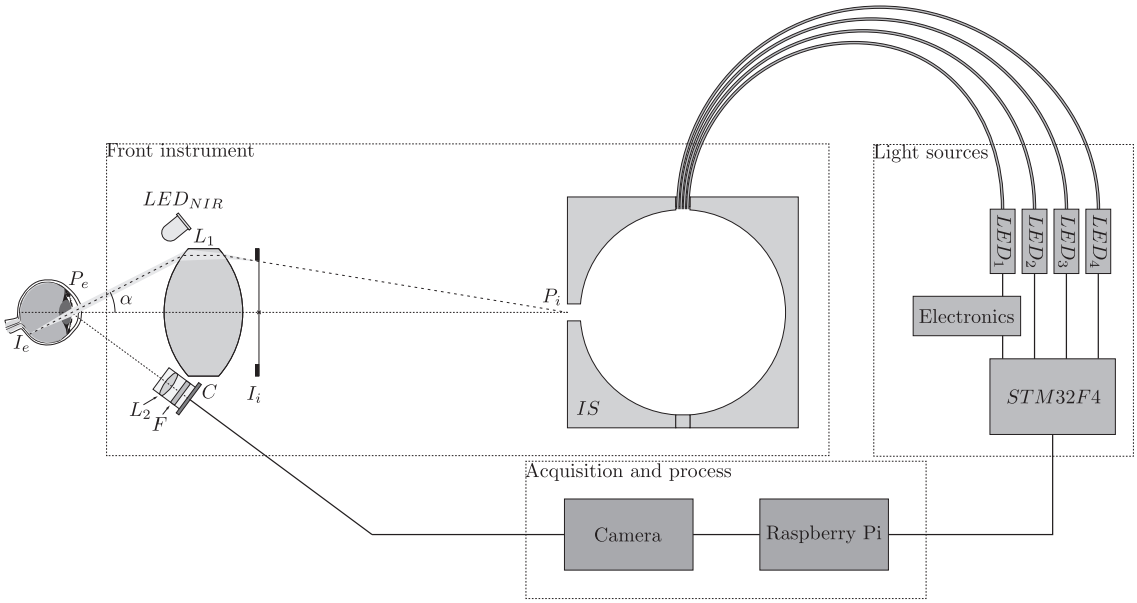
For silent substitution devices, instruments with Newtonian view are more common and are used in a larger number of studies than those with Maxwellian view. For the latter, out of five publications, three are based on the same instrumentation.[6–8] Another group of four publications proposes custom-built instruments accompanied by very vague explanation regarding the construction and working principle.[15,16,18] Spitschan[9] demonstrates 50% of contrast with a Maxwellian setup by using an artificial pupil and a digital spectral modulator. Only one commercially available solution for natural view could potentially be used for ipRGC excitation (ColorDome,Diagnosys,Cambridge,UK) because it includes 4 different light sources but are aimed at color stimulation only.[11,20,21]

Material and methods

Instrumentation

We propose a new device with a homogeneous 52° field of view (FOV) based on a Maxwellian view able to achieve a high contrast (theoretically 85%) for ipRGC excitation. The exit pupil diameter of the device was set to 2.5 mm but can easily be reduced to 1 mm, which requires a subject's pupil slightly larger than this value to accept slight lateral movements. In our system, the pupil diameter of the eye under light excitation is measured and not the contralateral eye which would be required if an artificial pupil is used.

Figure 1. System layout with the optical parts, the light sources and the control system.



The four light sources: LED_1 (A42182, Seoul Semiconductor), LED_2, LED_3 and LED_4 (M405F1,M505F1 and M625F2, ThorLabs) modulated with pulse width modulation signal, are homogenized through an integrating sphere IS. The LEDs are controlled with a microprocessor (STM32F4). The exit port P_i of the sphere is imaged onto the pupil of the eye P_e with lens L_1 with a 5:2 magnification. The surface I_i, with a central dot for eye fixation purpose, is projected onto the retina of the subject. Four NIR led at 900 nm illuminate the pupil which is observed with a camera fixed below the lens L_1. The camera includes a lens L_2 and a high pass filter F to avoid visible light from the integrating sphere. During light experiments, a video at 24 frames per second is acquired. The red channel is processed on a Raspberry Pi with the library opencv in python. A Gaussian filter is applied in order to remove noise and each pixel are then reduced to one bit. Pupil contour is extracted and an ellipse is fitted to this contour which gives the size and the position of the pupil. If no contour is found, image is considered as part of eye blinking.

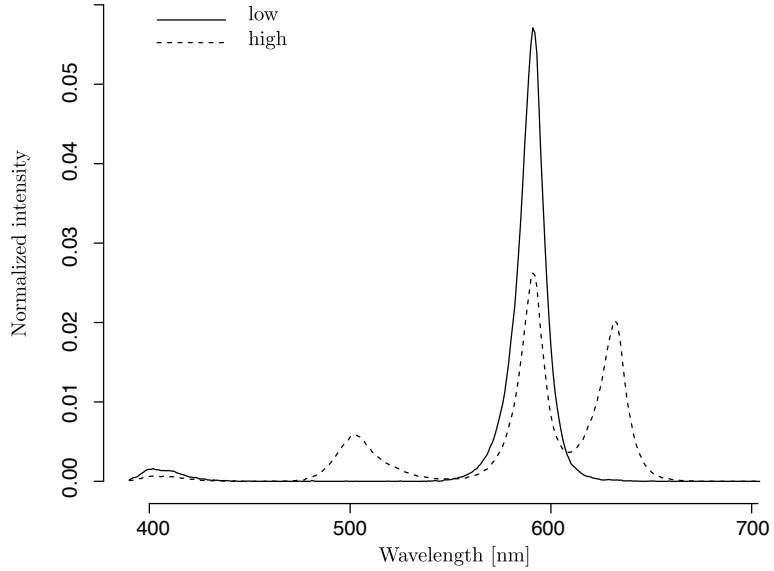
Luminance

The luminance L_v is the result of L and M cones excitation. In case that the cones are silently stimulated, then the luminance remains constant. Its was computed as following

$$L_v = \sum_{\lambda} \left(\frac{I_{555} \cdot S(\lambda) \cdot V(\lambda)}{R(\lambda) \cdot \Omega \cdot A} \cdot 683 \right) \left[\frac{\text{cd}}{\text{m}^2} \right] \tag{5}$$

where I_{555} is the optical power (measured with the powermeter PM100D, ThorLabs, set at 555 nm), $S(\lambda)$ the power spectrum, $V(\lambda)$ the light efficiency function of the eye normalized to 683, $R(\lambda)$ the responsiveness function of the powermeter, Ω the solid angle sustended by the illumination ($\Omega = 0.636$ for a FOV of $\pm 26°$) and A the area of the exit pupil of the instrument with 2.5 mm in diameter.

Figure 2. Illumination spectrum for the low and high level of ipRGC excitation with identical luminance and color.



The calculated luminance L_v for different excitation levels of ipRGCs varies little ($\pm 0.44\%$) up to a contrast level C of 70%. Table 2 shows the measurement of L_v at the contrast levels used for the evaluation of the instrument.

Contrast C [%]	0	20	50	70	from 0 to 70
L_v [cd/m^2]	511.4	511.6	509.6	506.7	509.8 ± 2.3

Table 2. Measured L_v values for different ipRCGs contrast.

Optical validations were performed by measuring the spectrum variation for different excitation steps (see example in Figure 2).

Experiments

We performed two experiments:

1. L-cone: We exposed three eyes to a bluish light background (50 cd/m^2). At 7-second intervals, we added, during one second, red light with logarithmic increasing luminosity L_v from 1 cd/m^2 to 316 cd/m^2.

2. ipRGC: We exposed 3 eyes to a contrast change of resp. 20%, 50% and 70% at a constant luminosity of 510 cd/m^2 in the following order (see upper-left figure 4): 12s baseline at $PS = (S, L, M, ipRGC) = (28, 224, 137, 12)$, 12s at resp. $ipRGC = 18$, 36 and 57 then back to baseline value. Duration of the transition was 0.5s and change was linear on $ipRGC$. We did not record the pupil diameter from darkness to light and from light to darkness before and after the measurement (no long recovery).

Results

1. L-cone: As shown in the literature[22] we found a linear relationship between the normalized relative constriction of the pupil nC_p and luminance of the red excitation L_v, $nC_p = 0.5 - 12.5 \cdot \log(L_v)$, $R^2 = 0.9758$ (figure 3).

2. ipRGC: For all subjects, the pupil diameter was 4.55 ± 1.63 mm and the minimum was 2.97 mm which is above the exit pupil diameter of our experimental set-up. The changes of the pupil diameter increase with the increase of the contrast of the ipRGC excitation (see figure 4 right).

Figure 3. Experiment a) L-cone. Mean results for the L cone excitation. Upper-right: one single recording at $log(L_v) = 2.5$.

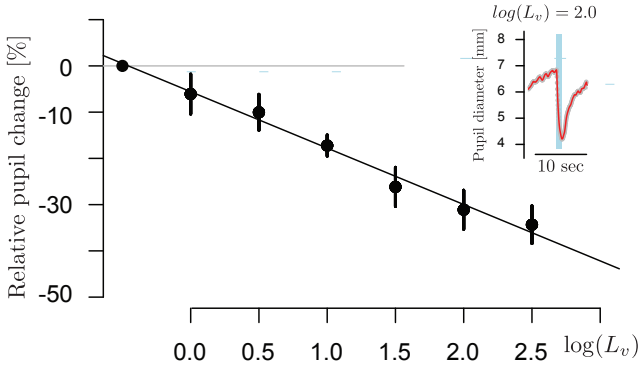
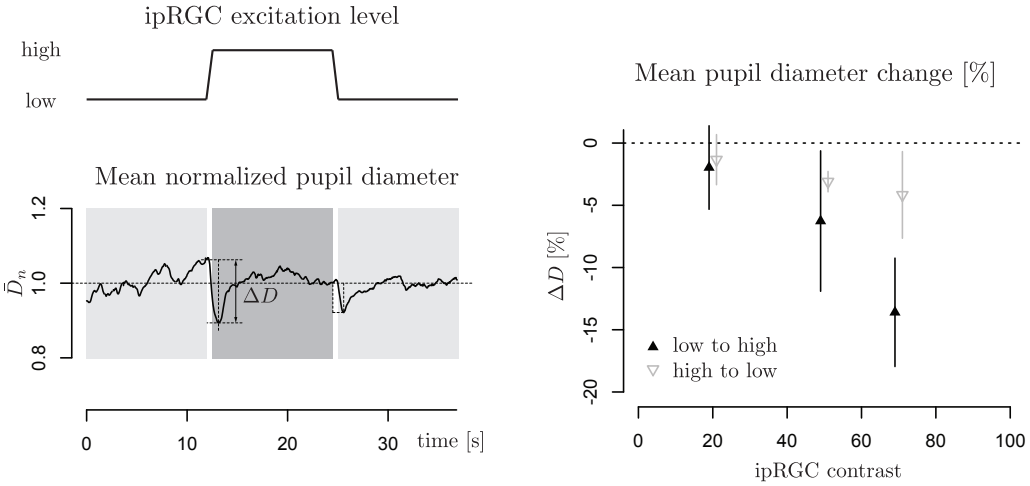


Figure 4. Experiment b) ipRGC. Top-left: time change of the ipRGC excitation. Bottom-left: example of the mean of normalized pupil diameter of 5 recordings on one subject at 70% ipRGC contrast. Right: mean and standard deviation of the pupil diameter change from low to high and from high to low ipRGC excitation for different contrast.



Conclusion

We demonstrate a working light stimulus device which appear to excite melanopsin ipRGC independently of simultaneous cone excitation. A detailed quantification of the time variation of the pupil waveform parameters

is the next step and will help to better differentiate cones excitation from ipRGC. Simultaneous psychophysical and electrophysiological recording with the pupil recording will also help to validate the silent substitution. Compared to instrumentation reported in the literature, our device exhibits higher FOV and potentially higher ipRGC contrast. The optical system has a precise exit pupil where the eye's pupil is located. The instrument can be aligned in the same way as a fundus camera.

Thanks

We are grateful to Prof. Sei-ichi Tsujimura for the many helpful and constructive discussions.

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