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Analytical study of optical component for optogenetic application

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ABSTRACT

Optogenetics is a combined technology of optics and genetics to achieve gain or loss functions of well defined events within specific cells of a living tissue. That is why in optogenetics there is need to choose specific optical components which cause no harm to cells or tissues or any kind alteration of cells. In optogenetics, stimulation of neurons strictly depends on optical power density received in the brain. In this paper, via simulation, we have shown dependency of optical power density for stimulation of neurons using three different wavelengths and for various parameters on which irradiance depends like, core radius, input light power and numerical aperture of the fiber.

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1. Introduction

In 1979 Francis Crick, taking note of the complexity of the mammalian brain and the fact that electrodes cannot readily distinguish different cell types, suggested that a major challenge in neuroscience was the requirement to precisely control activity in one cell type while leaving the others unaffected. Crick later suggested that light might be a relevant tool to control the different cells of brain, but he did not give the concept for how this could be done [1]. In years earlier, bacteriorhodopsin had been identified as a microbial single-component light-activated ion pump [2,3]. From here the term optogenetics was introduced. Reliable and targetable single-component tools (which encompass both light sensation and effect or function within a single protein) have enabled versatile new classes of investigation in the study of neural systems. Opsins are a group of light sensitive membrane bound G protein coupled receptors of the retinylidene family. These Opsins are planted surgically over brain, which get attached to the surface of neurons.

2. Sources used

Opsins responds to incident light on them and generate current same as the neurons generate when they get activated for any cellular activity. In optogenetics, main challenge is to design a tool for experimentation such that opsins generate desired current and stimulate neurons without damaging or burning the neurons. To provide opsins required optical power, various light sources can be used. These light sources are Lasers, LEDs and incandescent sources. Lasers are attractive choice for many optogenetic experiments. These have very narrow spectral linewidth (typically <1 nm), which can be matched closely to the peak activation wavelength of the optogenetic tool of interest; also, when many lasers can be directly modulated at kHz to MHz frequencies. Laser beams have a very low divergence, and can be focused in a very tiny area. The narrow width and low divergence of laser beams are very important in to couple and propagate light into optical fibers, which require light to be focused to a very small spot size [4,5]. Lasers with a power output on the order of mW are mostly used that are driven with a power supply that allows analog modulation of output power. This level is enough to generate high light power densities using small diameter optical fibers even after coupling and transmission losses, and after splitting into multiple fibers.

On the other hand, LEDs have the broad linewidth in comparison to Lasers, but have required narrow spectral tuning. These are readily modulated at the required frequencies, also have simple and stable operation, and do not require complex control electronics. But, when used near the tissue, substantial heat is generated, which can permanently damage the tissues, so for in vivo use caution should be taken. Because of heat generated by LEDs, it is safer to implement on the tissues outside their natural surroundings (in vitro) for experimenting. In vitro, LEDs can serve as the light source for optogenetic experiments, and LED arrays are available that permit focal stimulation of single cells, or even single neuritis [6,7] (Tables 1–3).

For in vivo applications (i.e. inside the tissue's or cell's natural surrounding), LEDs can be used to fill an optical fiber which is

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Table 1
Irradiance (mW/mm^2) for different wavelengths: 473 nm; 561 nm; 630 nm.

Depth (mm)	Irradiance (473 nm)	Irradiance (561 nm)	Irradiance (630 nm)
0	73.18	73.18	73.18
0.5	4.02	5.62	9.75
1	0.82	1.33	2.73
1.5	0.23	0.44	1.03
2	0.07	0.17	0.45
2.5	0.03	0.07	0.22
2.9	0.01	0.04	0.13

Table 2
Irradiance (mW/mm^2) for different wavelengths: 473 nm; 561 nm; 630 nm.

Depth (mm)	Irradiance (473 nm)	Irradiance (561 nm)	Irradiance (630 nm)
0	152.73	152.73	152.73
0.5	8.39	11.73	20.34
1	1.7	2.78	5.7
1.5	0.48	0.92	2.14
2	0.15	0.36	0.94
2.5	0.05	0.15	0.45
2.9	0.02	0.07	0.25

Table 3
Irradiance (mW/mm^2) for different wavelengths: 473 nm; 561 nm; 630 nm.

Depth (mm)	Irradiance (473 nm)	Irradiance (561 nm)	Irradiance (630 nm)
0	15.12	15.12	15.12
0.5	1.47	2.06	3.57
1	0.39	0.64	1.3
1.5	0.13	0.25	0.57
2	0.05	0.11	0.28
2.5	0.02	0.05	0.14
2.9	0.01	0.02	0.08

tethered to a behaving animal, but such applications are limited by the highly divergent beam pattern from LEDs, with high-power LEDs, this fraction of total power is sufficient to attain the required power density output [6,7]. Possible uses of LEDs include both direct implantation of small LEDs in or on tissue (with heating concerns requiring careful control as noted above), or permanently mounted to optical fiber waveguides carried on the subject [8].

Broadband incandescent microscopy light sources, such as arc lamp-based fluorescence illuminators, can also be used in optogenetics. One key advantage of the broadband light is the ability to select arbitrary wavelengths and spectral linewidth using bandpass filters, which was not possible with Lasers and LEDs. Even more flexible are monochromators, which output commanded wavelengths via positioning of a diffraction grating.

3. Observation and simulation

Brain tissue may be targeted with light through optical fiber which inserted in the head through skull, making hole in it. When light is incident on the brain tissues, optical losses in the form of geometric loss, scattering, attenuation, etc. occurred. These should be minimized. In optogenetics, irradiance is the key parameter for stimulation of neurons. Irradiance at a particular depth in the brain depends on many factors like numerical aperture, wavelength, input optical power and core radius of the optical fiber.

In this paper, we have studied the relationship between wavelength, numerical aperture, penetration depth in brain tissue and irradiance. Figs. 1–9 are logarithmic graphs for irradiance v/s depth of penetration in the brain tissue, for three wavelengths: 473 nm, 561 nm and 630 nm. In this study, we have used the simulation tool developed by Stanford University [9].

Case 1: NA = 0.22.

Light power from fiber tip = 2.3 mW.
Fiber core radius = 0.1 mm.

Case 2: NA = 0.22.

Light power from fiber tip = 4.8 mW.
Fiber core radius = 0.1 mm.

Case 3: NA = 0.22.

Light power from fiber tip = 2.3 mW.
Fiber core radius = 0.22 mm.

Figs. 1–3 of case 1 shows that with increase in depth of penetration, irradiance (mW/mm^2) decreases sharply and then becomes constant. It is clear that irradiance decreases less rapidly when wavelength increased. Decrease in irradiance is more sharp in 473 nm than 561 nm and curve is little smooth for 630 nm. Also there can be seen dependency of irradiance on parameters such as

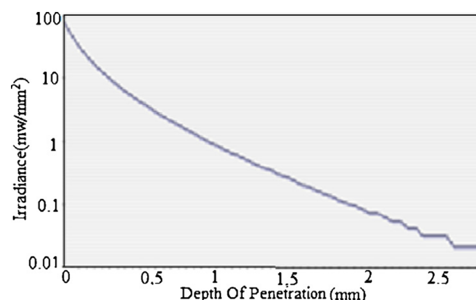

Fig. 1. Logarithmic graph of irradiance v/s depth of penetration in brain tissue (473 nm).

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