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ScienceDirect

Procedia Technology 20 (2015) 225 – 230

Procedia Technology

The International Design Technology Conference, DesTech2015, 29th of June – 1st of July 2015, Geelong, Australia

Wireless Optogenetics: An Exploration of Portable Microdevices for Small Animal Photostimulation

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Abstract

Preclinical research in optogenetic neuromodulation in small laboratory animals allows far greater control of neural circuitry. This precision provides an enhanced opportunity for understanding the neural basis of behavior. However, behavioral neuroscience research is limited by conventional benchtop optogenetic systems. By necessity, the animal is tethered to the light source external to the testing environment. Portable optogenetic microdevices enhance the potential for valid behavioral testing in naturalistic conditions by eliminating tethering and enabling free and unrestricted movement. This paper reviews recent advances in the development of portable optogenetic microdevices supported by wireless power transfer. Light sources and fiber coupling are common problems in optogenetic systems and are addressed. Device designs and parameters are summarized, along with advances in component technology for energy storage and distribution that make these devices possible.

Peer-review under responsibility of School of Engineering, Faculty of Science Engineering & Built Environment, Deakin University

Keywords: Neuronal cells; Optogenetics; Design; Microdevices;

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Peer-review under responsibility of School of Engineering, Faculty of Science Engineering & Built Environment, Deakin University
doi:10.1016/j.protcy.2015.07.036

1. Introduction

Optogenetic stimulation of neuronal cells has transformed our capacity for precise study of neural circuitry and emergent complex behavior. This research can help us understand how cognition and behavior are altered through selective manipulation of neuronal cell types and pathway projections. Optogenetic neuromodulation research can greatly enhance our understanding of the brains complexity. When used in combination with preclinical animal models of disease, our capacity to elucidate pathogenic and therapeutic mechanisms is greatly enhanced. Conventional benchtop optogenetic systems are costly, bulky, inefficient, and can restrict animal movement due to the tethering necessity for wired connections between the probe and the photostimulation system.

Tethering can cause the animal to become entangled or to experience torque if connected to a rotary device, preventing naturalistic observations that enhance the quality and validity of behavioral tests. In optogenetics, a solution to this issue would be to create a miniature photostimulation device that can be affixed to or implanted in the animal, such that behavior is minimally affected. This allows researchers to recreate naturalistic conditions so that tethering confounds are eliminated. This paper discusses the design and development of optogenetic devices that are small, low-cost, replaceable, and easy to use to allow for large-scale preclinical studies to occur in parallel. This is in contrast to using few, expensive benchtop optogenetic systems capable of stimulating only a few animals at a time. For this reason, fabrication of optogenetic microdevices is a rapidly developing area. We investigated the literature on the current portable devices and analyzed their technological direction, the studies that they are facilitating, the advanced components they utilize, as well as important parameters for their design.

2. Optogenetics

In brief, optogenetics offers the ability to modulate activity of transfected neurons that express opsins on their axonal surface. Activation of opsins through light allows them to transport specific ions by acting as channels. This affects the membrane potential of the cell, resulting in depolarization or hyperpolarization. Optogenetics therefore grants the ability to modulate neuronal activity through pulses of light. Each subfamily of opsins responds to different wavelengths of light, as well as controls which ions it gates. Table 1 lists common opsins used in optogenetics. An appropriate light source and delivery method is key to activating these opsins, as the penetration of light through the brain is relatively low.

Table 1. Common optogenetics opsins.

Opsin Type (subfamily)	*Gating Type*	*Wavelength (nm)*	*Reference*
Channelrhodopsin (ChR-2)	Cation Channel	460	[1]
Halorhodopsin (NpHR)	Chloride Pump	580	[2]
Archaerhodopsin (Arch)	Proton Pump	566	[3]

3. Light Source

Several photostimulation sources have been introduced to the neuroscientist’s arsenal including halogen lights, liquid crystal displays (LCDs) light-emitting diodes (LEDs), and arc lamps. Recent devices incorporate LEDs as the light source for photostimulation. They have the benefits over laser system due to beam stability, price, size, and precision. The system by Clements et al. [3] for example uses an LED light source. Despite a typical laser’s ability to produce high-power irradiance, optical power levels sufficient to activate opsins remain modest, about 0.1-1 mW/mm^2. Clements et al. used their tethered LED system to deliver photostimulation to ChR-2 transfected neurons in mice. This caused the animals to perform a freezing response, demonstrating a behavioral change by optogenetic stimulation, Although their system used a tethered fiber cord to deliver photostimulation, they introduce the important concept of fiber coupling to optimally transmit the LED light into the fiber.

4. Fiber Coupling

An important consideration in developing an optogenetic microdevice is deciding how to transport the light produced from the light source to the neural cells. In cases of portable devices, where a limited power source constricts the amount of energy that can be used, transmission efficiencies becomes an important aspect at every junction that the light transfers between. Optimizing the coupling efficiency of light transfer is challenging because of the size discrepancies between the optical fiber and the light source. A smaller optical fiber core allows for less incident light to enter the fiber. Multi-modal fibers however offer better light transmission because of their larger core diameters. The numerical aperture (NA) should be maximized for optimal coupling efficiencies as this allows less light to escape from the cladding during light propogation through the fiber. The NA can be defined as:

$$NA = \sqrt{n_{core}^2 - n_{clad}^2} \tag{1}$$

where n_{core} is the refractive index of the core, and n_{clad} is the refractive index of the peripheral cladding. This value helps define the capacity of the optical fiber to gather and transmit light. It is therefore better to choose fibers with a higher NA value in most cases.

5. Devices

There have been many stepwise improvements in technology since the inception of optogenetics, however a critical hurdle has been the portability of such devices. Previous optogenetics research consisted of only stimulating while a stereotaxed animal lay motionless under anesthesia, and the system required connection to an external power supply. Portability becomes increasingly necessary when performing preclinical behavioral research. Due to the inherently large size and weight of most battery-based power sources, bulky devices can interfere with the behavior of animals. Therefore, smaller wireless systems have greater promise for ensuring animals behave in environments that are more naturalistic during stimulation. Importantly, smaller devices give the animal less material to grab onto during self-grooming, which prevents the animal from damaging or removing the system and causing unintentional pain and stress.

One issue with many animals being removed from a study is their tendency to grab a device or electrode and pull it off their skull. This leads to a high loss of resources from costs of each animal's treatment as well as time lost performing surgeries and other procedures. It also results in extreme pain and stress to the animal, which requires termination of the animal from the study and thus increases the likelihood of loss of life associated with research. Therefore, the use of a chip with dimensions of only 9 mm^2 chip is an advantage in the system by Yeh et al. [4] (see Fig. 1). This small chip can be implanted under the skin, and prevents the animal from easily removing it. They harness power through an electromagnetic midfield, where the device gathers energy from a coil outside of the head to charge a capacitor. This capacitor in turn is used to power the LED. The electromagnetic midfield produced by the source's circuit board transmits half a Watt of power, sufficient to power the device. This is also well within a safe range for organic tissue, as it is less powerful than a cellular phone. Higher efficiencies of power transfer can be achieved when the receiver is much smaller than the source [5], an important consideration when designing a portable optogenetic device.

Wireless and battery-less devices take advantage of power transmission for energy harvesting. However, this harvested energy still needs to be stored in the device in sufficient quantities until needed for the light-pulse. This energy storage is typically done using a smaller battery or a capacitor. Unfortunately, there are issues with using any battery system including having a considerably slow charge rate, a finite number of charge and discharge cycles, and being a hazardous substance if the internal chemicals were to leak [6]. Therefore, simply using the energy received from a transmitter to charge a battery will not only be bulky, but would also not be able to efficiently power the device for sustained periods of time. On the other hand, delivering energy to the light source would require several capacitors to supply the required energy. While batteries have sufficiently high energy densities and low power densities, capacitors have much higher power densities but lack in energy density [6]. Both are required for an ideal microdevice.

A solution to this issue lies in using electric double-layer capacitors or supercapacitors, which store the most energy per unit mass among capacitor designs. They represent a fundamental middle-ground between batteries and capacitors. Supercapacitors are capable of maintaining power densities much higher than batteries. Meng et al. [6] fabricates these supercapacitors to be solid-state and flexible. They measure only 1 mm^2 yet produce an incredibly high area of capacitance (1.3 mF/mm^2). It is worth noting that excessive current could cause permanent damage to the supercapacitor.

Combining the components of a supercapacitor and a RF power harvester can substantially aid in animal research on a mass scale, while the animals perform their behavioral activities in specialized apparatus or in their homecage. The device developed by Wentz et al. [7] (see Fig. 2) induced rotational movements in animals through stimulation of cortical motor neurons. The entire device was affixed to the rat's head using a headstage, but the rat maintained electrical and physical insulation from the device components. This device harnessed energy through a low-strength oscillating magnetic field of 300 A/m. They powered an LED using a supercapacitor at 4.3 W in burst mode.

A further upgrade to these devices would couple the discussed photostimulation components with electrical recording. Ameli at al. [8] (see Fig. 3) have more recently developed such a device. This device's size is $15x25x17mm^2$, and weighs about 7.4 grams. It is able to harness power through a wireless signal rectifier at 7 cm, yet can send back information to a computer 2 m in distance. This makes such a device suitable for large animal studies in parallel, making high throughput neurostimulation studies for rodents possible.

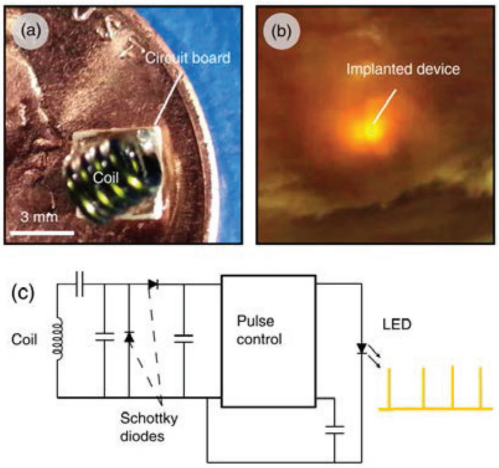

Fig. 1. Yeh et al.'s device [4]. (a) Stimulator. (b) Implanted stimulator. (c) Circuit diagram.

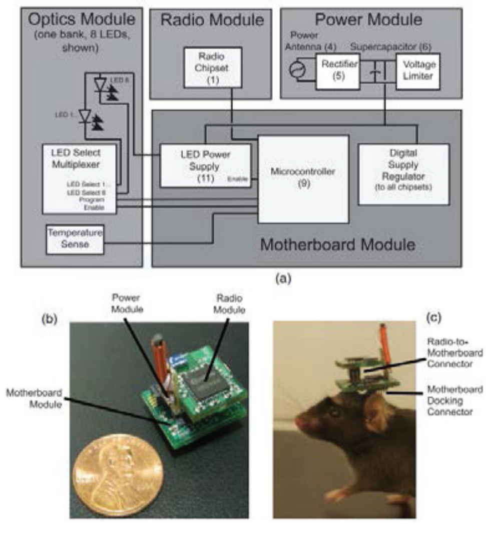

Fig. 2. Wentz et al.'s device [7]. (a) Block diagram of the device. (b) The device. (c) The device mounted on an animal.

6. Discussions

The diversity in parameters needed for optogenetics is indicative of the considerable research potential it holds. During its infancy, optogenetics relied on opsins that were readily available in nature (see Table 1). They come from the green algae *Chlamydomonas reinhardtii* [1] and *Volvox carteri* [9], and the archea *Natronobacterium pharaonis* [2] and *Holorubrum sodomense* [10]. Each is unique in its excitation wavelength and the method through which it modulates the cell. Newer genetically engineered forms of opsins are being introduced. There are now a variety of opsins that are tailored to be excited at unique wavelengths and with unique modes of action.

Similarly, each microdevice made for optogenetics contains unique engineering parameters optimized for the system, such as for wireless power transmission. Preliminary validation of new devices can be accomplished by replicating previously described neuromodulation studies. The devices are then subsequently refined to be able to handle experiments that are more diverse. Therefore, some aspects of each device (see Table 2) may seem limiting, such as its relative size and weight [8] or the low transmission ranges of power [7], [8]; these limitations are progressively minimized as subsequent iterations of the device are fabricated. Animal trials are important in validating a device's utility in order to determine if the device is ready for biological testing. Yeh et al.'s [4] and Wentz et al.'s devices [7] performed such testing and yielded results supporting their device. Although Yeh et al.'s device [4] demonstrated the effectiveness of the device in freely-moving rats for different positional orientations, Wentz et al.'s device [7] actually manipulated the behavior of the rat by increasing the number of rotations performed by the rat when stimulating the motor cortex. This demonstrates the necessity of behavioral research in animal models through optogenetic stimulation.

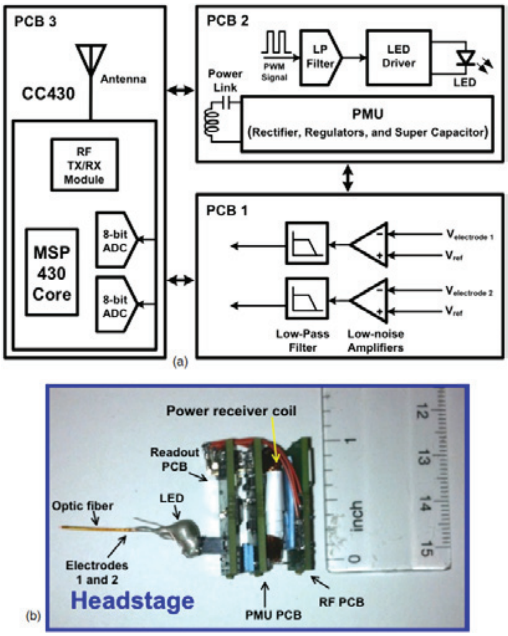

Fig.3. Ameli at al.'s device [8]. (a) Block diagram of the device. (b) The device.

Table 2. Portable device parameters.

Parameter	Device [4]	Device [7]	Device [8]
LED Wavelength	590/630 nm	470 nm	-
Device Dimensions	9 mm^2	<1 cm^2	15 x 25 x 17 mm
Weight	-	2 g	7.4 g
Receiver Frequency	1.6 GHz	2.4 – 2.485 GHz	868 MHz
Transmission Range	15 cm	"Several cm"	<7 cm
Data Rate	-	1 Mbit/sec	320 Kbit/sec
Animal Trial	Tested device efficacy at different orientations in freely-moving animal	Stimulated motor cortex and counted rotational turns made by rat	No animal trial performed

7. Conclusions

Advances in medical technology and microdevice fabrication are capable of substantially advancing brain research and neuromodulation technologies. Neuromodulation devices are constantly being developed and refined for use by researchers for both basic and translational research. Understanding the needs of preclinical research paradigms and adapting the trends and advances in the field of optogenetics is necessary for continued innovation. This way, animal behavior studies of critical clinical implications can be performed simultaneously in multiple subjects in large scale to better understand treatments for wide-ranging neural syndromes such as depression, epilepsy, Alzheimer's, Parkinson's Disease, and many more. By constantly improving and adapting new technologies, researchers can tap into the hidden physiology of the brain to better understand the underpinning biological processes, and work towards developing rational therapies for these ailments.

Acknowledgements

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Wentz, C.T., et al., *A wirelessly powered and controlled device for optical neural control of freely-behaving animals.* Journal of neural engineering, 2011. **8**(4): p. 046021.
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