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Design of headstaged behavior experiment system based on wireless program control technology

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Abstract. In view of such problems as high cost and large volume of laser output optogenetics experimental instrument and limited range of activity of the experiment object in the experiment of optogenetic behavior of small animals,aheadstaged wireless programmed instrument for optogenetic experiment for small animals is designed.The instrument is made up of wireless transmitter, main controller and optrode based on the principle of low power consumption and radio frequency. Compared with traditional laser,the optical pulse power,frequency,pulse width and time parameter are all wirelessly adjustable.The control distance can be up to 10 meters,which overcomes the disadvantage that the traditional laser is dependent on the output of optical fiber to limit the range of the object's range of activity.At the same time,it supports extended channel output,and the optrode can be biocompatible and long time implanted in the body.The experimental results are in agreement with the theoretical analysis through experiment.The size of the instrument is 2.01.20.6cm^3.The weight is only 2.5g.The electrode shape can be customized according to experimental needs.Therefore,the instrument meets the needs of optogenetics and behavioural experiments of small animals,and promotes the application of optogenetic technology in the research of neural circuits and disease models exploration.

Key words. Optogenetics, behaviouralexperiment, headstaged, wireless program control.

1. Introduction

With the continuous development of optics and genetics, optogenetics technology is being applied more and more to basic science, neurophysiology and other fields[1,2].Selectively expressing optical sensitive channels on specific types of cells by genetic means, the technology irradiates the specific wavelength of the laser to specific area through optical fiber, so as to achieve the stimulation or inhibition of the neuronal cluster[3]. In recent years, the technology has been gradually applied to clinical study of diseases, such as narcolepsy, depression, anxiety, pain and

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blindness[4,5,6,7]. Basic behavioral observation is an important criterion to judge whether the optogenetic experiment is effective.

The mainstream optogenetic experiment device mostly uses the output of laser mounted optical fiber.But the wired connection limits the range of the object to be measured, and it can not be the best choice for the experiment of basic behavior [8,9]. In view of this, a new type of headstaged micro-load wireless programmed instrument for optogenetic experiment is presented and designed.The instrument overcomes the defects of the traditional wired connection restrictive behavior experiment, and solves the existing problems in the application of the existing wireless devices.It also has far control distance, light load and low loss, and meets the requirement of optogenetics technology in small animals in-vivo experiments, represented by mice.

2. System Design

This system mainly consists optrode,controller,emitter and software.Under the premise of low load and low loss, the parameters of output optical pulse are adjustable. Figure 1 is a schematic diagram of the working process of this instrument.

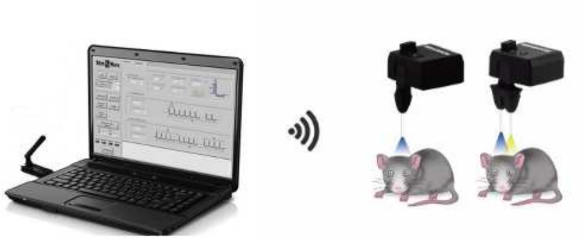

Fig. 1. Schematic Diagram of Wireless Programmable Optogenetic Experiment Instrument

2.1. Optrode design

The overall structure of the optrode that can be designed with a customizable wavelength and can be implanted into the designated area of the experiment is shown in Figure 2.

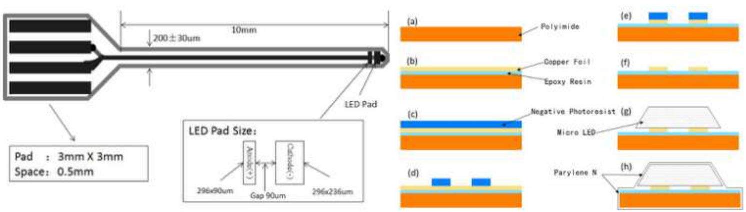

Fig. 2. Optrode specification and fabrication flow

In this paper, the innovation of optrode design combines the flexible printed circuit (FPC) and micro electro mechanical system (MEMS), including four parts: flexible substrate, optical source, gold finger and covering material. The optical source is fixed on the substrate's designated pads by coating the conductive paste, and the biocompatible material is modified on the line and the surface of the optical source, so as to improve the reliability and stability of the optrodeimplantation.The substrate is made of Polyimide (PI) which has good mechanical and electrical properties. It can be used as an optrode carrier to support the optical source and act on the experimental target location. The CREE 460nm wavelength blue optical LED is used as the optical source. Its volume is only 350x470x155μm^3, and the luminous intensity is up to 82mW/mm^2. It can meet the needs of all kinds of optogenetics experiment in a large range. In order to ensure excellent electrical insulation, heat resistance and biocompatibility, the Parylene-C is used as the optrode protection material. The process flow of the optrode is (a)prepare the PI substrate;(b)paste the copper foil;(c)coat the photoresist;(d)pattern transfer process;(e)etch the unexposed copper foil;(f)peel off the residual photoresist;(g)solder the μLED;(h)parylene coating layer.

2.2. Hardware and software design

As shown in Figure 3, the Nordic nRF51822 is selected as the main control chip in the emitter and main controller plan of the instrument. The chip integrates 2.4G private protocol while ensuring the size. Under the requirement of expansion, the constant current driver selects the MBI5036 model chip that is driven by 16-channel current. Under the requirement of miniaturization, the main controller uses a high integrated impedance matching element 2450BM14E0003 to replace the matching circuit of the inductance and capacitance combination. At the same time, a ceramic antenna with a size of only 5.1×2.0×1.2mm^3 is used to ensure the quality of wireless transmission and reduce the size and weight of the load unit effectively. The instrument is equipped with 60mAh micro lithium battery, which can be recharged in circulation.

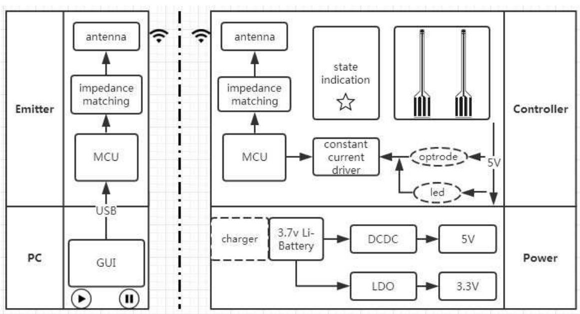

Fig. 3. Schematic of the headstaged wireless programmed instrument

The design process of embedded software is shown in Figure 4.

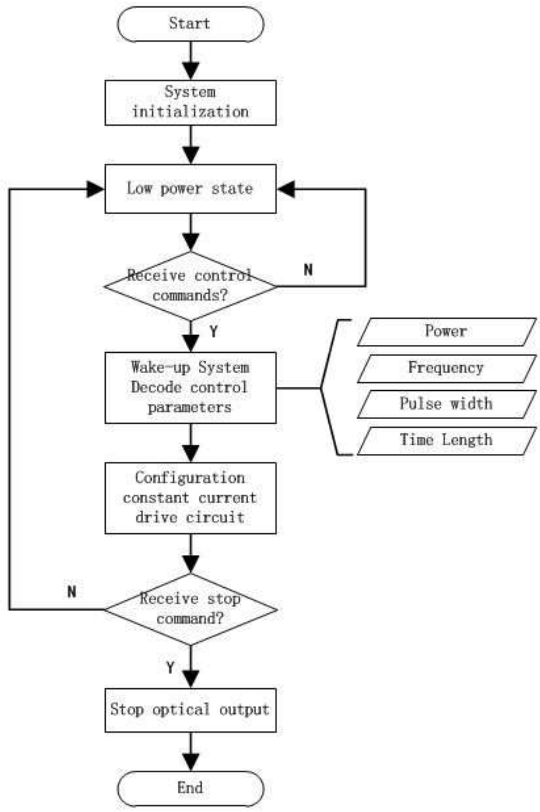

Fig. 4. Software Design Process

3. Experimental Test

Calibrating and testing the output optical power of the instrument is an important parameter for testing the optogenetic experimental instrument. In this paper, the LP1 portable optical power meter produced by SANWA company of Japan is used for measurement. The direct reading correction wavelength of the instrument is 633nm, according to the 460nm wavelength sensitivity correction coefficient 3.00 and the conversion formula:

Wavelength conversion value (W) = Display value (W) Correction coefficient

When configuring output of 5, 10, 15, 20 and 25mW by software, the mean value is obtained by the three measurements and the actual optical power is converted as shown in Table 1. The maximum of the average error is not more than 5% through the test data, which can meet the optical intensity requirements of the optogenetic experimental paradigm for the activated ion channel.

Table 1 Optical Power Test and Conversion

Set value	Measured Value1	Measured Value2	Measured Value3	Mean Value	Error Rate
5.0mW	1.68	1.56	1.64	4.88 mW	2.4%
10.0 mW	3.32	3.35	3.15	9.82 mW	0.18%
15.0 mW	5.12	4.95	4.80	14.87 mW	0.87%
20.0 mW	7.18	6.64	6.69	20.52 mW	2.6%
25.0 mW	8.91	8.50	8.78	26.19 mW	4.76%

The power of the instrument is an important factor in determining the service time of the instrument. The ammeter mode of 18B type multimeter produced by FLUKE company is used in this paper to est the mean value of power driven current under normal working condition.The test results, as shown in Figure 5, show that the optical output power of the optrode at different driving current is basically linear.

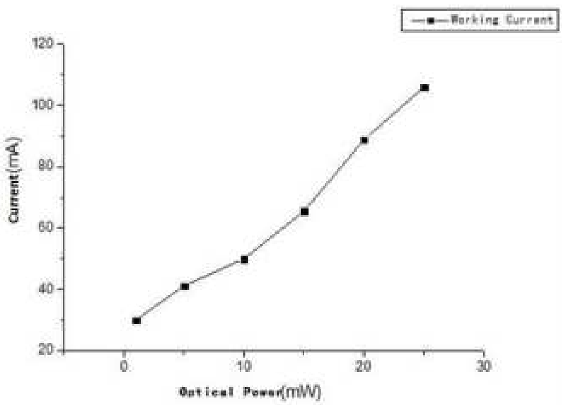

Fig. 5. The Relationship Between Optical Power and System Power

Wireless communication distance is an important indicator of the performance of instrument control. The test scheme is that the upper computer sends the data packets of 10,000 bytes, the lower computer detects the number of receiving at different distances, and turns it into the packet loss rate. The result of the test is shown in Table 2, it shows that the system can guarantee the effective communication distance within 10m, and the distance increases while the packet loss rate of communication increases. The distance of 10 m completely meets the requirements of basic behavioral experiment paradigm.

Table 2. Test of Effective Wireless Communication Distance

Distance	Send bytes number	Receive bytes number	Packet loss
0m	10000	10000	0
5.0m	10000	10000	0
10.0m	10000	10000	0
15.0m	10000	9960	0.4%
20.0m	10000	9920	0.8%

In-vivo experiment is a verifying experiment for this instrument. In this paper, the effects of optical stimulation on the mating behavior of mice pre-transfected with photoreceptor genes in a sober state are observed to verify its usability in the behavior experiment of free active mice.

Fig. 6. System Prototype for Optogenetic Experiment

There was a significant change in the behavior of the mice before and after the stimulation. In view of this, the headstaged instrument for optogenetic experiment designed in this paper can be effectively applied to behavioristics and other optogenetic experiments. The system prototype is as shown in Figure 6. The size of master controller of the system is 2.0x1.2x0.6cm^3, and the weight in together with the adapter and electrode is about 2.5g. The output pulse parameters of the system are as follows: the adjustable range of optical power is 1.0-25.0mW, and the adjustable precision is 0.5mW; the frequency adjustable range is 1-200Hz, and the adjustable precision is 1Hz. The pulse width can be configured to be 10%˜100% of the cycle period, with a step of 10%. The time length range is 1-255s, and the precision is 1s, and it can also be arranged into an infinite time length. In addition, the shape of the optrode can be designed according to the requirements of the experiment. Both the performance test and the animal experiment proved that the system fully meets the requirements of the optogenetic experiment for the instrument.

4. Conclusion

In view of the fact that lasers and fibers can not be effectively applied to the behaviouraloptogenetic experiment of small animals, a headstaged wireless programmed instrument for optogenetics experiment is designed. Moreover, combined with the new technology, a set of mating implanted optrode for optogenetic experiment is also designed, which solves the problems of restricting free movement distance, overloading and biocompatibility. The final experimental results of this design prove that it can be used in the experiment of optogenetic regulation of small animal behavior. At the same time, it can also provide a new instrument for exploring the working mechanism of the neural loop, revealing and developing new methods for the treatment of diseases under the optogenetic technology.

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