A fully implanted programmable stimulator based on wireless communication for epidural spinal cord stimulation in rats

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Abstract

Clinical research indicates that the epidural spinal cord stimulation (ESCS) has shown potential in promoting locomotor recovery in patients with incomplete spinal cord injury (SCI). This paper presents the development of a fully implantable voltage-regulated stimulator with bi-directional wireless communication for investigating underlying neural mechanisms of ESCS facilitating motor function improvement. The stimulation system consists of a computer; an external controller, an implantable pulse generator (IPG), a magnet, the extension leads and a stimulation electrode. The telemetry transmission between the IPG and the external controller is achieved by a commercially available transceiver chip with a 2.4 GHz carrier band. The magnet is used to activate the IPG only when necessary to minimize the power consumption. The encapsulated IPG measures 33 mm × 24 mm × 8 mm, with a total mass of ~12.6 g. Feasibility experiments are conducted in three Sprague-Dawley rats to validate the function of the stimulator, and to investigate the relationship between lumbar-sacral ESCS and hindlimb electromyography (EMG) responses. The results show that the stimulation system provides an effective tool for investigation of ESCS application in motor function recovery in small animals.

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1. Introduction

Since 1967 (Shealy et al., 1967), the epidural spinal cord stimulation (ESCS) has become an established and widely adopted treatment for chronic pain. Recently, ESCS has shown positive effects in facilitating locomotor recovery in individuals with incomplete spinal cord injury (SCI) and multiple sclerosis (MS) (Cook and Weinstein, 1973; Davis et al., 1981; Tallis et al., 1983). Dimitrijevic and his colleagues reported that ESCS at 25–50 Hz elicited rhythmic lower limb flexion/extension movements, while at 5–15 Hz initiated lower limb extension movements in spinal cord injured (SCI) individuals in supine position (Dimitrijevic et al., 1998; Jilge et al., 2004; Minassian et al., 2007). In their experiments, an implantable system was used to provide epidural stimulation of spinal cord in patients. By combining ESCS with partial weight-bearing therapy, Herman and his team (Carhart et al., 2004; He and Herman, 2011; Herman et al., 2002) investigated a protocol for facilitating over-ground ambulation in wheelchair dependent individuals with chronic, incomplete spinal cord injury. In their clinical research, an externally powered ESCS system was used to provide high stimulation power and long pulse duration (up to 1000 μs). The system consisted of an implanted receiver (X-trel 3470), a pair of implanted quadrupole electrode leads (PISCES-Quad Plus, Model 3888), the dual implanted lead extensions, an external transmitter (X-trel, Model 3425), and an external antenna (Model 3440). The X-trel external transmitter powered the implanted receiver via transcutaneous RF telemetry. Their results demonstrated that ESCS with a frequency of 20–40 Hz, a pulse width of 800 μs, and the amplitude between the sensory and motor threshold had effective influence on the motor system (He and Herman, 2008; Huang et al., 2006).

Although these studies have indicated that ESCS can promote locomotor recovery in SCI patients, an optimal protocol and the underlying neural mechanisms of this therapy remain unclear. It is desirable to conduct further research to decipher the potential mechanisms for the exploration of optimal protocols and stimulating parameters to guide the future clinical application of this promising treatment for motor function recovery after severe spinal cord injury. It has been demonstrated that ESCS applied at the L2 segment in the rat spinal cord isolated from brain control can induce bilateral stepping patterns most readily when compared with stimulation at other spinal segments (Gerasimenko et al., 2008; Ichiyama et al., 2005). In their animal experiments, the stimulation electrodes were made of stainless steel microwire (Teflon-coated stainless wires, AS632, Cooner Wire, Chatsworth, California).
CA, USA) by removing a small portion (~1 mm notch) of the Teflon coating to expose the desired area of the wire and locate on the stimulating area of the spinal cord. The electrodes were connected to an amphenol connector cemented on the skull of the animal (Roy et al., 1991). The amphenol connector was then connected to a Grass S88 stimulator (Grass Instruments) through a stimulus isolation unit (Grass S11US, Grass Instruments). In order to implant the stimulation electrodes in the rat, several holes need to be drilled on the rat's skull to settle the connector on its head by using screws. The complicated surgery and cumbersome setup of the stimulation system hamper the research investigation and it becomes imperative to develop a low cost and easy to operate ESCS device for investigation of the neural mechanisms of ESCS on the motor system.

Although implantable stimulators for clinical application have been developed for spinal cord stimulation (SCS), these devices are not suitable in animal experiments due to high cost and large size. Some implantable neural electrical stimulators have been developed for animal experimental studies, but they have specific application for selected animal models on the treatment of diseases such as Parkinson's disease (Harnack et al., 2008), chronic pain (Ativanichayaphong et al., 2008), deafness (Millard and Shepherd, 2007), gastrointestinal motility-related disorders (Jalilian et al., 2007), muscle atrophy (Dennis et al., 2003; Thil et al., 2005), and focal cerebral ischemia (Zhou et al., 2010), as listed in Table 1. These devices have achieved several technological advancements such as data and power transfers. There are reports on the transcutaneous data transfer by using different communication modalities such as reed switch, inductive links, antenna coupled radio telemetry, and optical links. Some implantable stimulator is powered by an external pulsed magnetic field (Millard and Shepherd, 2007). Others are supplied by various primary batteries such as Li/SVO battery (Jalilian et al., 2007), button cell (Harnack et al., 2008; Lanmuller et al., 2005; Thil et al., 2005; Zhou et al., 2010), Li/SOCl2 battery (Lanmuller et al., 2005), etc. In general, the selection of battery in the implantable stimulator is a matter of its size and capacity.

In this paper, we present an implantable system for the epidural stimulation of spinal cord in small laboratory animals. The system is small in size, low in cost, easy to build and use with a radio frequency controlled stimulus waveform output. The performance of the stimulator is summarized in Table 1. We verified the function of ESCS system in vitro and vivo rat tests.

2. Materials and methods

The stimulation system shown in Fig. 1 consists of an implantable pulse generator (IPG), an external controller allowing the data communication with the IPG, a stimulation electrode, and the extension leads. After implanting IPG in the rat, the stimulation parameters are reset using a computer to send the control command to the external controller via USB port.

2.1. Implantable pulse generator: hardware overview

The schematic diagram of the IPG shown in Fig. 2 is composed of five major blocks: (1) the wireless link; (2) an embedded microcontroller unit (MCU); (3) DC–DC converter; (4) stimulation front-end circuits; (5) a primary battery.

2.1.1. The wireless link

The bidirectional wireless communication circuit is established by CC2500 (Texas Instruments, Dallas, USA), a single 2.4GHz ISM (Industrial, Scientific and Medical) band commercialized transceiver chip with very few external components. The CC2500 is chosen because of its small size and low current consumption. The CC2500 transceiver consumes 17 mA in receiving mode, and
11–20 mA in transmitting mode depending on the transmission power. In the power-down mode, only 0.5 μA current is drawn from the power supply. The external components around the CC2500 shown in Fig. 2 consist of an antenna, an antenna matching circuit, a 26 MHz crystal oscillator, and a digital interface with MCU. The ceramic chip antenna AN9520 (Rainsun group, Sindian City, Taipei), 9.5 mm × 2 mm × 1 mm in size, can be easily soldered on the printed circuit board (PCB). The digital interface with the MCU


Fig. 1. Illustration of the stimulation system. (a) The IPG contains a primary coin battery. The microcontroller is programmed by a five pole connector placed on the left side of the PCB, and the connector is removed before encapsulation; (b) the external controller is connected to a PC via a USB port. The user can send the control commands from PC to the external controller, and then the external controller transmits the stimulation parameters to the IPG via radio frequency telemetry; (c) the stimulation electrode and extension leads carry electrical pulse to the stimulated spinal segments; (d) the IPG and stimulating electrode in the stimulation system (revised from [15], with the permission of the author) are implanted in the rat. A USB cable connects the computer with the external controller. The experimenter can change the stimulation parameters by means of a user interface in the PC.

Fig. 2. The schematic diagram of the IPG. The MCU (C8051F411) is the central process unit of the IPG that modulates the transceiver (CC2500), the DC–DC converter (MAX686), and the operation amplifier (TLV27L2). The DC–DC converter permits a voltage transformation from 3 V to 10 V. The output of MAX686 is controlled by the MCU via EN pin. A magnet reed switch is connected to the MCU, which allows the user to initiate the IPG from sleeping mode. The 4-wire SPI serial interface (CSN, MOSI, SCLK, and MOSI) is used to configure CC2500 and access data buffer, while GDO2 can alert when the packet has been received/sent. C8051F411 includes two channel 12 bit current mode D/A converter. The MCU outputs two voltages from IDAC0 and IDAC1, and then the voltages are amplified in the TLV27L2.
includes a 4-wire SPI (CSN, SCLK, MOSI, MISO) interface and GDO2 pin. Via the SPI interface, the MCU enables the configuration of CC2500 and accesses the data buffer, while GDO2 provides a confirmation signal for transceiving a wireless data packet. Moreover, the CC2500 is configured to perform MSK (Minimum Shift Keying) modulation, with 500 kbps data rate. The transmission output power is programmed at 1 dBm to enable the reliable communication between the implant and the external controller separated by less than 1 m.

2.1.2. The microcontroller unit

The MCU is the central processing unit of the IPG. In the stimulation system, C8051F411 (Silicon Laboratories, Boston, USA) is chosen to build the IPG because of its small size (5 mm × 5 mm, QFN package, 28 pins), relatively low power consumption, and two embedded 12-bit current-mode digital-to-analog converters (DACs). In working mode, the chosen system clock of the MCU is 0.76 MHz (24.5 MHz/32) with the current consumption of ~0.3 mA. In order to minimize the power consumption of the implant, the MCU of the implant is placed in sleep mode when it is not activated. In sleep mode, the 3.2768 kHz external crystal oscillator is used, and C8051F411 draws only about 13 μA current from the power supply.

A normally off magnetic reed switch connects the active-low external interrupt pin and the ground. Most of the time, the MCU is in sleep mode at 32 kHz and the CC2500 is in power down mode, which minimize the power consumption of the IPG. When a magnet is held close to the IPG, an external interruption is generated in the MCU. To avoid the misoperation, the internal timer of the MCU is enabled to calculate the times of the external interruption caused by the reed switch within 6 s. If the external interruption occurs more than three times, the MCU will switch into its working mode.

2.1.3. DC–DC converter

MAX686 (Maxim, Sunnyvale, CA, USA) is an inductor-based step-up DC–DC converter. MAX686 is selected due to its space saving size (6 mm × 5 mm, 16 pins), with the efficiency up to 90%, and its maximum 27.5 V output voltage. In our application, the MAX686 provides 10 V to power the stimulation front-end circuits. Pin EN controls MAX686 mode (disable/enable). In disable mode, MAX686 only consumes 1.5 μA current.

2.1.4. Stimulation front-end circuits

The key block in the pulse generator architecture is the stimulation front-end (SFE) circuits, which is the interface between the MCU and ESCS electrode. In Fig. 2, each channel of the two-channel SFE circuits contains an output current from a current-mode DAC in the MCU, i.e. IDA0 or IDA1, a voltage amplifier, and a switch (Q1 or Q2). The current output of the IDACs can be adjusted to four different settings: 0.25 mA, 0.5 mA, 1 mA and 2 mA, with the maximal output voltage of 2 V. To provide the voltage-controlled stimulation up to 10 V, a low voltage rail to rail operation amplifier TLV272L (Texas Instruments, Dallas, USA) is used to amplify DAC outputs to the desired stimulus voltage. The TLV272L is chosen because of its low quiescent current (7 μA/channel), high slew rate (0.1 V/μs) and suitable supply voltage range (2.7–16 V). The small signal MOSFET NTA14153N (ON Semiconductor, Phoenix, USA) is selected as a switch (Q1 or Q2) in the circuits.

The SFE circuits of the IPG select a pair of active electrodes ( bipolar), and control the stimulus voltage between them. Since the unbalanced stimulus pulses result in net ion flow, tissue damage and corrosion of the anode electrode surface (Dennis et al., 2003), a charge balanced waveform is important between the bipolar electrodes. As illustrated in Fig. 3, one channel of the SFE circuits outputs a square pulse ($V_A$), another channel ($V_B$) outputs a delayed square pulse with a quarter of amplitude and four times pulse duration of $V_B$. Then a charge balanced biphase stimulation waveform ($V_{stim}$) obtained from $V_A$ and $V_B$ has a cathodic phase followed by an anodic phase with an inter-pulse delay. In the cathodic phase, the stimulus current depolarizes nearby axons and initiates the action potential. The succeeding anodic phase cancels the charge accumulated in the cathodic phase on the electrodes. An inter-phase delay separates the two stimulus pulse slightly, so that the anodic phase does not block action potential propagation initiated by the cathodic phase (Liu et al., 2007).

2.1.5. A primary battery

A 3 V, 270 mAh lithium coin battery with legs CR2430 is utilized in the IPG. The size of the battery is Ø23.0 mm × 3 mm with a weight of ~4.3 g.

2.2. Implantable pulse generator: software overview

The software is written in C and compiled by Keil C51 Compiler. Upon power-up, the program configures I/O pins, disables DC–DC converter MAX686, and enters sleep mode to conserve power. Thereafter, upon detection of an external magnetic field that is close to the reed switch, an external interruption is generated. If the reed switch triggers external interruptions more than three times in 6 s, the MCU exits sleep mode. Then the MCU activates CC2500, and sets it to receive mode. After receiving the parameter, Cyclic Redundancy Check (CRC) is performed on the received data to ensure that the data integrity is preserved during the wireless communication. Following CRC check, a confirmation message is transmitted to the external controller to inform the operator of the status of the IPG. Thereafter, the IPG interprets the received data, and makes decisions for three different functions: (1) protocol stimulation function, (2) custom stimulation function, and (3) stimulation shutdown function. In protocol stimulation, the stimulation parameters are preprogrammed and the stimulation session lasts 20 min; while in custom stimulation mode, the operator needs to specify the stimulation parameters and ends the stimulation by activating the stimulation shutdown function.

2.3. Stimulation electrodes and leads

As shown in Fig. 2(c), a stimulation electrode manufactured with a flexible circuit board technique can be placed into the dorsal epidural space. There are three round golden contacts with the 1 mm diameter separated at 8 mm center-to-center distance on the electrode, while eight small holes at the middle are used to fix the electrode on the nearby tendon. At the proximal end of the electrode, the Teflon coated stainless steel wires (Cat No. 793500, A-M System Inc, http://www.a-msystems.com/) are soldered to connect the electrode to the IPG. Moreover, the medical silicon tube (inside diameter 0.5 mm, outside diameter 0.9 mm) is used to cover the stainless steel wires to avoid the mechanical fracture after...
implantation. The impedance of the electrode and the extension leads is measured at different frequencies (100, 120 Hz, 1k, 10 kHz) with a LCR meter (Agilent 4263B) and Agilent 16089E Kelvin Clip Leads. In vitro test, the electrode-lead system is immersed in 0.9% saline. During in vivo test, the stimulation electrode is placed in the epidural space of spinal cord in rats during the surgical procedure.

2.4. User interface and external controller

The block diagram of the external controller is illustrated in Fig. 4. The external controller consists of a USB to UART convert chip (FT232R, FTDI, UK), a microcontroller unit (C8051F410, Silicon Laboratories, Boston, USA), a RF transceiver chip CC2500, and a whip antenna. The MCU is the central processing unit of the external controller, which controls the operation of FT232R and the transceiver chip CC2500. The SComAssistant (serial port testing assistant) is used as a user interface for the operator to send commands to the implant. The returned confirmation message from the IPG is also displayed in the user interface to inform the operator of the status of the implant.

2.5. Package and test

The IPG is fabricated using surface-mount devices (SMD) assembled on a very small four layer printed circuit board (PCB). The assembled PCB is cleaned with ethanol. Once it is dry, the PCB is coated with conformal coating (Electrolube DCA SCC3) to protect the circuit from the acidic acid in the sealant. After conformal coating was cured overnight at 80 °C (Millard and Shepherd, 2007), the assembled board is then coated with a silicone elastomer (Permatex 65AR, www.permatex.com). After the silicone elastomer coating dries, the button battery is assembled and the IPG is coated with a silicone elastomer. After the silicone elastomer coating on the IPG is dried at 40 °C, another five layers of elastomer encapsulant are applied and allowed to dry overnight (Dennis, 1998).

After encapsulation, the functionality of the IPG is verified by the following procedure: (1) send stimulation parameters to the PC; (2) sweep the magnet near the IPG; and (3) observe the voltage waveform between the two output wires in the electrode with an oscilloscope. During this functional test, a resistive load of 10 kΩ between the two output wires is used to simulate the impedance of the electrode-tissue-interface.

In the heat failure test, the IPG is immersed in 0.9% saline for 24 h at 50 °C (Dennis et al., 2003). Afterwards, the IPG is removed from the saline, rinsed in tap water, and functionally tested again by using an oscilloscope. Then the IPG is coated with a silicone elastomer again. The silicon devices in the vitro test are not implanted in rats. Finally, a patch of prolene mesh (Jiangsu sempoll pharmaceutical Co., Ltd., China) is affixed to the encapsulated stimulator with silicone adhesive to prevent the stimulator migration after implantation.

2.6. Experimental subjects and surgical procedures

The experiments were conducted in three adult Sprague-Dawley rats (1 male and 2 female, 250–310 g) to evaluate the function of the IPG and investigate the relationship between ESCS and the electromyography (EMG) of the hindlimb muscles. The rats were housed in a well ventilated room during a 12 h light/dark cycle with access to food and water ad libitum. The experimental protocol was approved by the Ethics Committee for Animal Research, Huazhong University of Science and Technology, China. During surgical procedures, the rats were anesthetized with chloral hydrate (0.4 mg/kg for the first 2 h, and maintained at a surgical level with supplemental doses as needed).

Intramuscular EMG activity of the hindlimb was used to monitor the responses of ESCS at different stimulation parameters. The surgical implantation of intramuscular EMG recording electrodes and the headplug was performed following the protocol (Roy et al., 1991). Briefly, a 12-pin connector (SamTech) was secured to the skull and the Teflon-coated stainless wires (Cat No. 793500, A-M System Inc, http://www.a-systems.com/) were passed subcutaneously either to the back or to the hindlimb. Skin incisions were made to expose the tibialis anterior (TA) and medial gastrocnemius (MG) muscles bilaterally. A small notch (~2 mm) of the Teflon coating was removed from each wire to form EMG electrode. Bipolar intramuscular EMG electrodes were then inserted into the muscles.

The implantation of the stimulator and ESCS electrode began five days after the headplug and EMG implant surgery. Prior to the implantation the stimulator was immersed in 75% ethanol to minimize the risk of infection. A partial laminectomy was performed at the T9 spinal segment, and then ESCS electrodes were placed over the lumbar-sacral segment and fixed to the tendon of latissimus dorsi. The stimulators were subcutaneously implanted in the back of the rats. A rat with an implanted stimulator is shown in Fig. 5.

All incision areas were irrigated with saline and sutured in layers (i.e. investing fascia and superficial fascia then the skin). The analgesics (Buprenex, 0.5–1.0 mg/kg, three times per day) were initiated after surgery and continued for 2 days.

Fig. 4. Block diagram of the external controller.

Fig. 5. A rat with an implanted stimulator and a head connector.

2.7. Testing procedures

ESCS test started immediately after the stimulator was implanted. All testing was performed when the rats were anesthetized. A charge balanced biphasic stimulation waveform was applied to the dorsal face of the spinal cord. The hindlimb EMG responses to the epidural electrical stimulation were recorded. A low-noise amplifier (RM6240BD, Chengdu Instrument Factory, China) was used to collect the four channels of intramuscular EMG signals through the headplug. Raw signals were digitally sampled at a rate of 2000 Hz, and then band-pass filtered at 30–1000 Hz.

The hindlimb responses to lumbar-sacral ECS at different stimulation parameters were investigated in three rats. ECS was administrated initially by stimulation at 1 Hz frequency and 400 μs pulse width to study the motor responses of the different stimulation amplitude. The stimulation amplitude was increased at 0.1 V incremental step, and restricted not to induce discomfort or pain in rats. Thereafter, the changes in the responses at different stimulation frequencies were analyzed. The average peak-to-peak amplitudes of the EMG responses at stimulation frequencies of 3, 5, 10 Hz and at a pulse duration of 400 μs were compared with responses at 1 Hz. Subsequently, ECS was applied at 1 Hz and amplitude above motor threshold to investigate the responses of the different pulse widths. The average peak-to-peak amplitudes of the EMG responses at stimulation pulse widths of 800, 1200, 1600 μs were compared with responses at 400 μs. All statistical results were reported as means ± SE.

3. Results

3.1. Impedance of the electrode-lead system

The impedance of the electrode-lead system in 0.9% saline was 21.079 ± 0.589 kΩ at 100 Hz and 22.699 ± 1.353 kΩ in vivo condition.

3.2. Stimulation output in vitro

In our design, the output voltage could be adjusted from 0.1 to 10 V with approximately 2.4 mV resolution and the stimulation frequency could be changed from 1 Hz to 255 Hz, with a 1 Hz increment. ECSs for motor effects needed longer pulse width than that for chronic pain treatment likely due to the chronaxie effect on interneuronal cells (He and Herman, 2008). Table 2 gives the output waveform range, resolution and technical specifications of the implantable stimulator.

3.3. Hindlimb response in vivo test

Examples of the responses elicited at different stimulation amplitude in the TA muscle of one rat are shown in Fig. 6. Stimulation with 0.2 V did not induce observable changes in EMG response. The EMG responses showed a progressive increase in peak-to-peak amplitude with increased stimulation amplitude. The threshold for generating a small response in hindlimb muscles at 1 Hz was 0.3 ± 0.1 V. Fig. 6b illustrates the characteristics of a single response on the expanded time scale. The mean threshold for a muscle response induced by ECS (1 Hz, 400 μs pulse duration) of the three rats was 0.3 ± 0.1 V.

At stimulus frequencies between 1 and 10 Hz, we observed hindlimb extension movements synchronized with stimulation frequency in rats. When the frequencies were normalized to the values at 1 Hz, there was a progressive decrease in the mean peak-to-peak amplitude with increasing frequency of stimulation (Fig. 7). At 10 Hz, the mean peak-to-peak amplitude for the TA muscle was roughly 50% of the values at 1 Hz.

The responses of the lumbar-sacral ECSs at different stimulation pulse widths were also investigated. When compared to the values at 400 μs, there was a general trend for an increase in the mean peak-to-peak amplitude in the TA muscle (Fig. 8). At 1600 μs, the mean peak-to-peak amplitude for the TA muscle was roughly 200% of the values at 400 μs.
Table 2
Specifications of the implantable stimulator.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Regulated (constant) current or voltage</td>
<td></td>
</tr>
<tr>
<td>Frequency</td>
<td>1–255 Hz (1 Hz step)</td>
</tr>
<tr>
<td>Amplitude</td>
<td>0.1–10 V output (2.4 mV resolution)</td>
</tr>
<tr>
<td>Pulse width</td>
<td>0–4000 μs (step 400 μs)</td>
</tr>
<tr>
<td>Electrode configuration</td>
<td>Bipolar electrode</td>
</tr>
<tr>
<td>Current consumption in receiving mode</td>
<td>17 mA</td>
</tr>
<tr>
<td>Current consumption in transmitting mode</td>
<td>20 mA</td>
</tr>
<tr>
<td>Current consumption in sleeping mode</td>
<td>40 μA</td>
</tr>
<tr>
<td>Current consumption in stimulation mode (1 V, 400 μs, 40 Hz) with 10 kΩ resistor</td>
<td>1 mA</td>
</tr>
</tbody>
</table>

4. Discussion

4.1. Development of the implantable ESCS device

ESCS has been suggested as a promising therapy for functional recovery in walking after SCI. The implantable ESCS device available for modulating locomotor function in animal model has been in great demand due to the need to assess the effectiveness of this therapy in chronic animal experiments. To meet the high demand, we developed an implantable stimulator with bi-directional wireless communication for ESCS in rats. This stimulator is small in size, low in cost, easy to build and use with a radio frequency controlled stimulus waveform output.

In practical applications, the spinal cord stimulation could be easily disabled by encapsulation leakage, extension leads fracture, electrode displacement, and/or battery consumption after implantation. To avoid such failures from happening, a series of procedures are implemented in the design and manufacture process. First, five elastomer coatings are applied to encapsulate the IPG in the proposed system. The encapsulation method has been proved to be feasible in previous literatures (Dennis, 1998; Millard and Shepherd, 2007). Second, the medical silicone tube is used to protect the extension leads because it is common that the Teflon coating peels off after several weeks of implantation. The tube can also prevent stainless steel wires from being fractured while rats move freely. Third, in order to avoid dislocation of the electrode, eight holes on the substrate are used to attach ESCS electrode to the nearby tendon with the surgical suture. Furthermore, a polyethylene mesh is attached to the encapsulated stimulator for anchoring the stimulator after implantation. In addition, a good compromise between the battery size and its capacity is sought to develop a fully implantable stimulator. The IPG supplied by a primary 3 V button battery with 270 mAh capacity can enable the chronic stimulation of spinal cord for about two weeks in this pilot study. For a longer lifetime application, a primary battery with larger capacity or a rechargeable battery with inductive power transmission system can be considered. When powered by a 3 V, 2.3 Ah primary battery, the dog implant with CC1000 transceiver chip can work for about 3.2 years (Jallian et al., 2007).

The stimulation parameter is adjustable by RF communication, which also provides the state information feedback of the IPG after the implantation. Although inductive communication link between internal and external coils has been used in the implantable devices for many years, the RF transceiver chip CC2500 is very convenient in the development of the implantable stimulator for animal experiments. As far as the power consumption is concerned, a starting current of about 20 mA in the CC2500 could result in a decrease of the stimulator’s lifetime. It is desirable that some newly developed commercial transceiver chip, e.g. TMS 37157 (TI, USA), with micropower consumption (maximum 150 μA in active mode and 60 μA in shutdown mode), can serve the purpose in the future implantable stimulator for animal experiments.

4.2. ESCS for inducing motor responses in hindlimb muscles

We have demonstrated that the developed ESCS system is capable of inducing motor responses in hindlimb muscles. Moreover, the chosen stimulation parameters, e.g. the site, the intensity, the frequency and the pulse width, are all crucial for producing motor responses. It has been previously reported that in chronic spinal rats, epidural stimulation at the lumbar-sacral level elicited rhythmic hindlimb movements, with coordinated bilateral stepping being evoked more often when ESCS was applied at L2 (Ichiyama et al., 2005) or S1 (Gerasimenko et al., 2008; Lavrov et al., 2006). In our experiments, ESCS at lumbar-sacral level induced hindlimb extension movements in normal anesthetized rats. Meanwhile, two stimulation electrode contacts were separated at 8 mm distance, with the cathode placed at the sacral segment and anode at the...
lumbar segment. More contacts on the stimulation electrode can provide different contact configurations in future experiments.

It appears that the stimulation parameters, e.g. the amplitude/current, the frequency and the pulse width, are critical for the production of the locomotor behavior. We did not observe significant stimulus response between muscle activity and stimulation intensity below the motor threshold. In contrast, with ESCS above the motor threshold, the EMG responses showed a progressive increase in the peak-to-peak amplitude with the increased stimulation intensity. Moreover, it was reported that there was a general trend for a decrease in the amplitude of each responses in all muscles with an increase in stimulation frequency (Lavrov et al., 2006). Our experiment showed similar results with stimulation frequencies between 1 and 10 Hz. Furthermore, the motor effects on the TA muscle could be notably enhanced by increasing the pulse duration. In the future, the optimal combinations of site, amplitude, frequency, and pulse width will be jointly considered to investigate the motor functions of ESCS in small animals with our developed stimulation device.

5. Conclusion

We have developed and validated an implantable ESCS system for small laboratory animals. The IPG and ESCS electrode can successfully deliver biphasic balanced electrical stimuli to the lumbar-sacral segment area of the spinal cord in rats for about two weeks. The stimulation parameters of the IPG can be adjusted wirelessly by the operator using a user interface from a computer. In the preliminary animal experiments, we showed that ESCS at lumbar-sacral segment could produce acute changes in hindlimb EMG activity with our developed system. We further demonstrated the different motor effects caused by varying stimulation parameters. In conclusion, the proposed implantable ESCS system can be used in the investigation of ESCS for motor effects in small laboratory animals.

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