A multichannel neural signal detecting module: Its design and test in animal experiments*

Wang Yufeng[1,2], Wang Zhigong[1**], Lü Xiaoying[2], Gu Xiaosong[3], Li Wenyuan[1], Wang Huiling[2], Jiang Zhenlin[3], Lü Guangming[3] and K. P. Koch[4]

(1. Institute of Radio Frequency & Optical Electronics Integrated Circuits, Southeast University, Nanjing 210096, China; 2. State Key Laboratory of Bioelectronics, Southeast University, Nanjing 210096, China; 3. Key Laboratory of Neural Regeneration of Jiangsu Province, Nantong University, Nantong 226001, China; 4. Fraunhofer Institute for Biomedical Engineering, 66386 St. Ingbert, Germany)

Accepted on November 9, 2006

Abstract A four-channel neural signal detecting module with an implantable 12-contact cuff electrode was designed for real-time neural signal recording on peripheral and central nerves. The mathematic coupling model between nerve and electronic system was analyzed. Electrode connection configurations were considered. The detecting circuit included an input coupling network, a pre-amplifier, and some filtering and notching stages. Shield guarding and the right-leg-driven circuit were developed for further elimination of common mode interference. By electrode switches, the module could cooperate with a nerve functional electrical stimulation circuit, building a neural channel bridge-connection system. It was tested by recording experiments on rat's sciatic and spine nerves. The signals in spontaneous and evoked conditions have been captured successfully. In addition, an implantable neural signal detecting CMOS IC has been introduced.

Keywords: Cuff electrode, quasi-tripolar configuration, neural signal recording, neural channel bridge-connection.

The research on neural signals is essential to understand the information and behavior of life bodies. Since Galvani discovered bioelectricity in 1791, great progress has been made in studies of neural signal generation and transmission. Especially due to the contributions of Hodgkin and Huxley, who were awarded the Nobel Prize in Physiology or Medicine in 1963, applying cathode ray oscillograph and voltage clamp amplifier to identify the change in membrane ion channel, and Neher and Sahkman, who were awarded the Nobel Prize in Physiology or Medicine in 1991, inventing patch clamp amplifier, neuroscience is more and more connected with engineering technologies, e. g. electronics, which creates abundant multi-disciplinary researches.

Patch clamp technique enables precise measurements of transmembrane electrical signals. At present, the attention on neuron group network properties is increasing, as neural information encoding should be obviously regarded as neuron group cooperation functions. It is required to record neuron networks simultaneously in parallel, under a same trigger condition, to determine both sequence and connectivity of neuron activations. Conventional neural signal recording technique is for single neuron only and does not meet this requirement. Multichannel extracellular *in vivo* neural signal recording is the supposed means to observe the nervous system functional connectivity.

Moreover, multichannel extracellular recording can also contribute to clinic neural prosthesis[1—3]. As the nerve is damaged or interrupted, the remaining neuron stumps can keep alive while the neural channel is physically cut out. There exists the possibility of implanting an electronic device, connected to two stumps of the interrupted nerve, to bridge them and achieve an active neural functional regeneration. Neural signal detecting is the indispensable frontend for such a neural prosthesis system.

1 Coupling model in extracellular recording

A myelinated nerve fiber with Ranvier nodes is shown in Fig. 1. A Ranvier node can be regarded as a point source of transmembrane ionic current. If the transmembrane current at node $R(n)$ is $I_m(t)$, according to "all or none" rule of action potentials, the ionic current at other nodes can be presented in the

* Supported by National Natural Science Foundation of China (Grant Nos. 69825101, 90377013 and 60121101)

** To whom correspondence should be addressed. E-mail: zgwang@seu.edu.cn

same way except the delay, which results in

$$I_{m,n}(t) = I_m(t - kT) \quad (1)$$

where k is the serial number of Ranvier nodes between an arbitrary node and the origin, and T is the constant conduction delay from a node to the adjacent one. According to Gauss's law in Maxwell Equations, i.e.,

$$I_s = -\lim_{V_\varepsilon \to 0} \iiint_{V_\varepsilon} \nabla \cdot J_s \mathrm{d}V = 4\pi \cdot r^2 \sigma \frac{\mathrm{d}\varphi}{\mathrm{d}r} \quad (2)$$

and supposing a homogeneous medium in the extracellular space, each transmembrane ionic current source leads to a potential at an arbitrary extracellular point, which can be solved by Eq. (2). Summing the ionic current induced potentials together, the compound potential contributed by all the point sources along the nerve fiber can be given by[4]

$$\varphi(t) = \frac{1}{4\pi\sigma} \sum_{n=-\infty}^{\infty} \frac{I_m(t - x_n/v)}{\sqrt{h^2 + (x - x_n)^2}} \quad (3)$$

Eq. (3) is the mathematic expression of the coupled potential on the electrode contact in extracellular space, where I_s is the transmembrane ionic current and can be precisely measured and mathematically modeled by patch clamp technique, J_s is the current density, σ the conductivity of the extracellular media, φ the coupled potential, r the distance between the point P and the Ranvier nodes which equals to $[h^2 + (x - x_n)^2]^{0.5}$, v is the velocity of neural spike conduction. The obtained potential will be further sent to the electronic module and be amplified or processed.

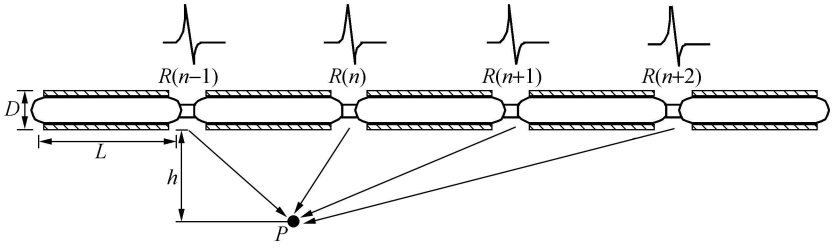


Fig. 1. The compound potential in the extracellular space of a myelinated nerve fiber with nodes of Ranvier.

It is shown that all the parameters or vectors in Eq. (3) can be obtained. So Eq. (3) can precisely model the coupling between a single nerve fiber and a neural microelectrode in the homogeneous media. However it is still an ideal and simple model. Considering that there are thousands of nerve fibers bindled into a nerve fascicle rather than only one, the structure and the internal medium can be very complex. And the number of the activated neuron can hardly be estimated. So there leaves more difficulty in practical cases for the mathematic solution of the electrode potentials. Referring to the extracellular cuff electrode in this paper, the obtained electrode signals are compound or summing potentials, which reflect the activation in the whole nerve fascicle.

2 Cuff microelectrode and quasi-tripolar configuration

Cuff microelectrode is one of the most effective electrodes for extracellular neural signal recording. Cuff electrodes are commonly constructed on silicon or polymer substrate with platinum or iridium electrode contacts by microelectronics technology, e.g. lithography and etching[5,6]. After implantation, the cuff electrode can curl and enwrap the nerve fascicle automatically, forming a restricted extracellular space, which can greatly improve the obtained signal quality. Electrode leads are connected to the inputs of the neural signal detecting module. According to the electrode contacts connection, there are two configurations for a cuff electrode, i.e. quasi-tripolar and true-tripolar, as shown in Fig. 2.

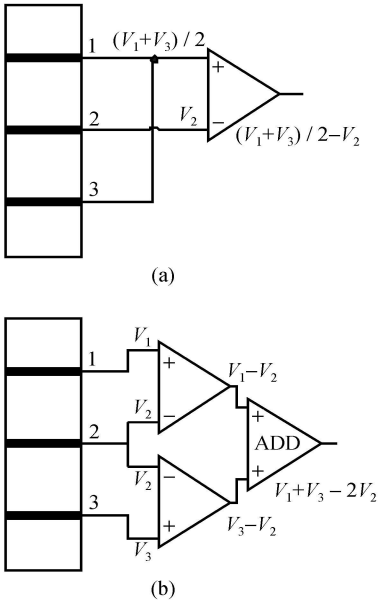


Fig. 2. Quasi-tripolar (a) and true-tripolar (b) configurations for cuff electrode.

The outputs of both quasi-tripolar and true-tripolar configurations contain the same signal components, while the amplitude in the latter one is twice the former, which implies that true-tripolar is better for improving signal-to-noise ratio (SNR). In quasi-tripolar, the two outboard contacts are shortend, which can reduce the potential difference between the outboard contacts and thus prevent the current flow along the nerve fascicle and through the cuff. The common mode potential of the short connected contacts 1 and 3 is equal to the mid-contact 2, which contributes to the common mode interference rejection in the implant environment. Quasi-tripolar is also advantageous in large dynamic range by avoiding electrode interface DC offset driving the neural signal detecting amplifier into saturation. Considering the requirements of common mode rejection and circuit simplification, quasi-tripolar was applied in this study.

Referencing to our group's previous study in the chemic orientation and the morphological measurement of rat's spine nerve fascicle section, a medium type 12-contacts cuff electrode obtained from Fraunhofer Institute for Biomedical Engineering IBMT, Germany (shown in Fig. 3) was used for rat's sciatic and spine nerve signal detecting.

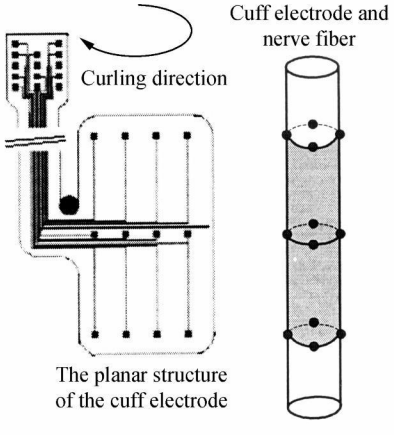


Fig. 3. The 12-contacts cuff electrode.

3 Design of multichannel neural signal detecting module

A four-channel neural signal detecting module was designed. Based on the analysis of a cuff electrode, the 12 contacts were divided into four groups. The three contacts located along the nerve fascicle in each group received a group of neural signals which were sent to the recording module outside the rat's body via leads. The leads were extra shielded by a copper silkscreen for better interference rejection. Fig. 4 shows the schematic of the detecting circuit for one channel, together with its connection with rat spine. The detecting circuit included an RC network, a pre-amplifier, an active band-filtering stage, a notch network, a shield guarding circuit and a right-leg-driven circuit.

The RC network was the first stage and was made up by two C_a and four R_a. The design did not simply introduce the commonly used dc coupling or ac coupling configuration. The reason was that: (1) in dc coupling, due to the electrode polarization voltage, the high-gain pre-amp (pre-amplifier) stage might be induced into saturation. A trade-off was to limit the pre-amp gain to a moderate level, e.g. less than 100, to avoid saturation. As well known, the common mode rejection ratio (CMRR) and noise characteristics greatly depended on the high-gain first stage. So dc coupling could worsen the characteristics of the neural signal detecting module; (2) in ac coupling, because of the implant electrode contacts imbalance and the passive components tolerance, the high-pass filter in front of the pre-amp could possibly convert common mode voltage into differential and degrade the effective CMRR. The finally proposed RC network in Fig. 4 provided ac coupling for differential signals and a dc path for amplifier bias. Because it was not grounded, no current flowed through the network when a common mode input voltage was applied, which led to an infinite CMRR theoretically. The tolerance of passive components could degrade CMRR but had been proved to be negligible[7].

The pre-amp, band-filtering stage and the notch circuit were conventional structures in analog signal amplification and processing. The pre-amp was composed of parallel amplifiers A_1 and A_2, with gain of R_b/R_g, determined by the feedback resistors. In the design, the gain could be adjusted by switches in the range between 500 and 10000. The active filter was composed of A_3 and A_4. The feedback of A_3 determined the mid-band gain R_{c2}/R_{c1}. The feedback of A_4 determined the lower cutoff frequency $R_{c3}C_c$. The higher cutoff frequency was limited by the unity gain bandwidth of the op-amp (operational amplifier). In the notch circuit, an auxiliary op-amp in positive feedback extra increased the notch attenuation. According to 50 Hz interference, R_{d1} and C_{d1} were selected as 47 kΩ and 68 nF, respectively.

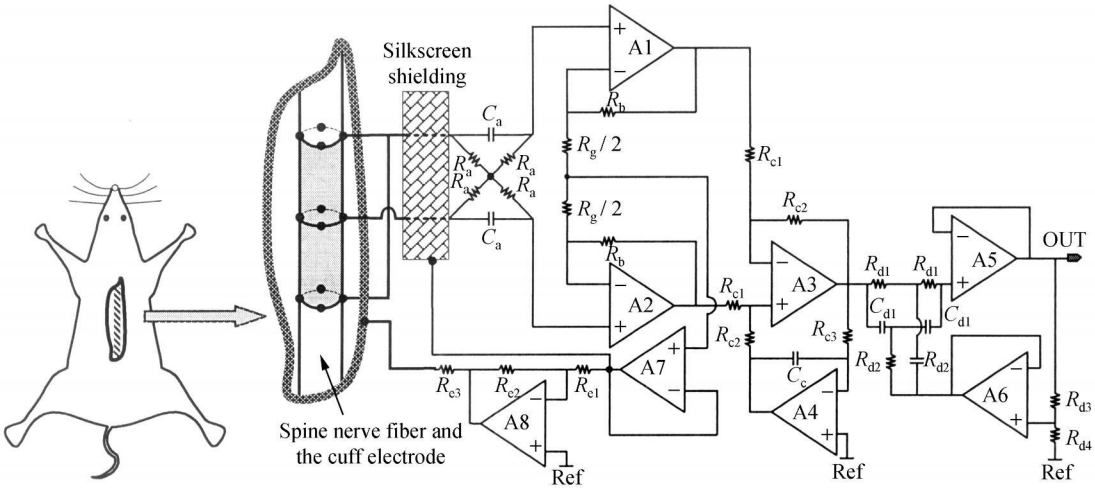


Fig. 4. The schematic of the recording circuit and its connection with rat spine.

Shield guarding and right-leg-driven circuit were particularly useful for additional enhancement in biomedical applications[8]. Shield guarding was made up by a unity gain op-amp A_7, which buffered the output common mode voltage of the differential preamp and clamped the shield to the same voltage. It was a practical way of eliminating of the capacitance between the shield and the signal lead, which is approximately 100 pF/m. The guarding also eliminated the leakage current of the coaxial cables and improved the input impedance, CMRR and amplifier frequency range. In fact, the shield was ac grounded from the low resistance output of A_7. Op-amp A_8 and a negative feedback loop constituted the right-leg-driven circuit, which was an established name for biomedical grounding using feedback op-amps. In biopotential measurements, animals in experiments were grounded from skin to reduce the interference, while the right-leg-driven circuit in our design was a more efficient or active means. As shown in Fig. 4, the common mode voltage was inversely amplified and applied back to the body, which drove the common mode interference to a low level of $i_d \cdot R_{e3}/(1+R_{e2}/R_{e1})$ with a strong negative feedback. Also the safety was improved because the current path to the ground went through the right-leg-driven circuit where some high resistances limited the current to a safe level.

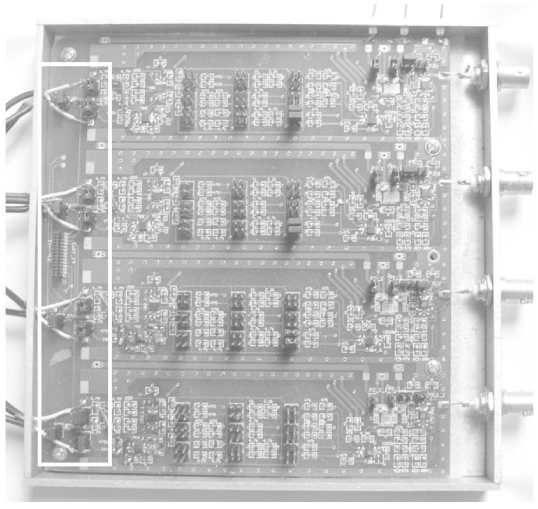
The module was fabricated on a printed circuit board (PCB). A commercial op-amp MAX4168 (Maxim Corp.) was selected in the circuit. The characteristics were simulated by Hspice and listed in Table 1, using op-amp macro-model. An aluminumshell and panel were also designed and the whole size was 15 cm×15 cm×2 cm. The photos of the circuit board and the module are shown in Fig. 5. The board contained four channels of circuits and connected the cuff electrode via a plug. An extra electrode switches unit was also included (shown in the pane of Fig. 5) to easily alter the connection of electrodes as recording or stimulating. Therefore, the module could be readily combined with neural electrical stimulation module to further build up a neural channel bridge-connection system.

Table 1. Characteristics of the neural signal recording circuit

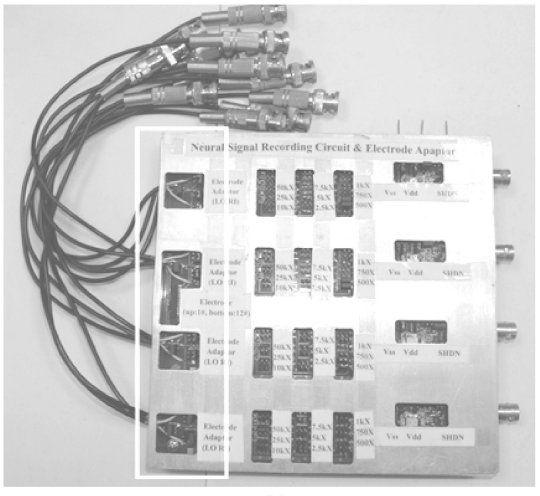
Supply voltage (V)	±1.5	CMRR (dB) @1 kHz	164
Current (mA)	9.6	PSRR (dB)	120
Power (mW)	28.8	Equivalent input noise voltage (nV/ $\sqrt{\text{Hz}}$) @1 kHz	9.2
Range of the gain	500—10000	Input offset voltage (mV)	4.7
3dB-bandwidth (kHz)	0.13—10.3	Notch attenuation (dB)	45

4 Experimental results of neural signal detecting

Two *in vivo* recording experiments were carried out in Key Laboratory of Neural Regeneration of Jiangsu Province, Nantong University in September and October 2005, respectively. The experimental objects included 3 rats and 2 rabbits. Here introduces the implant surgery procedure in rat spine nerve as an illustrative instance: a 250 g SD rat was anaesthetized by hypodermic injection of 10% chloral hydrate. The rat body was fixed and the skin was sterilized on the back. An insection was made to expose the spinal



(a)



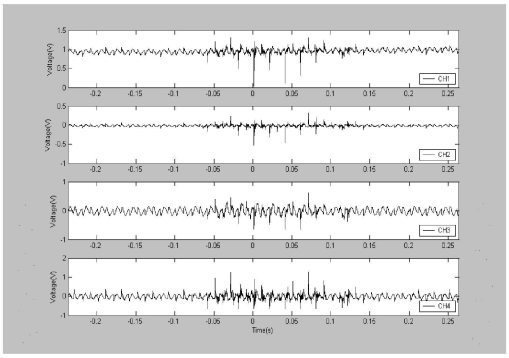
(b)

Fig. 5. Four-channel neural signal recording circuit board (a) and the completed module (b), with the electrode switches in the pane.

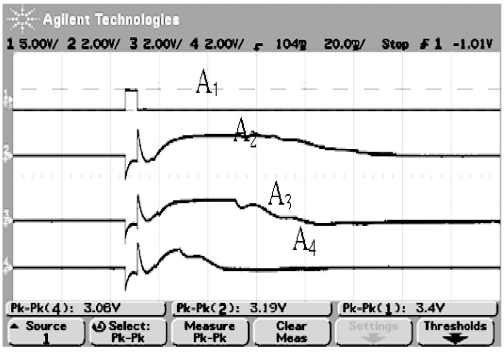
centrum. And the cuff electrode was enwrapped and fixed at the site. Then the electrode leads were connected.

Spontaneous and evoked neural signals were successfully detected from both spine and sciatic nerve of the rats and rabbits. Spontaneous signal recording condition was no stimulus in the environment, with the animal in anaesthesia. Evoked signal could be recorded from three groups of electrodes when applying 2—5 V amplitude, 1 ms width pulses to the rest one group of electrodes as stimulus. The spontaneous and evoked neural signals recorded in the experiments are shown in Fig. 6.

In Fig. 6(a), the spontaneous spinal neural spikes persisted for about 0.2 s. The maximum amplitude in the four channels reaches 2 mV. The width of a single spike is around 0.5—3 ms. The sequence relationships between the four channels, such as beginning and ending, showed high coherence. From the aspects of amplitude and phase, the spikes in channels 1 and 2 were almost the same, resulting in the highest pertinence. The signals in channel 3 showed the same spike sequence and phase relationship as channels 1 and 2 but a different amplitude shape. Channel 4 showed more distinctness, with both its amplitude and phase having a different trend from the other three channels.



(a)



(b)

Fig. 6. The four-channel spontaneous spinal neural spikes (a) and the evoked potentials (b), where A1 is the stimulus and A2—A4 are the evoked potentials.

In Fig. 6(b), A_1 is the signal from the stimulation electrode group, while A_2—A_4 are the recorded signals from the rest three groups. In A_2—A_4, evoked compound neural signals appear after the stimulus artifact. From the amplitude, the evoked signals are a little larger than the spontaneous ones and reach 6 mV. However, the width of spikes remarkably increases to 10—60 ms, which is probably due to that a large number of neurons in the fascicle were simultaneously activated by stimulation and contributed to the compound evoked potential.

Further signal processing and analysis can locate the activation zone in the nerve fascicle and trace the

signal evolvement in the network.

5 Design of implantable neural signal detecting IC

The module in Fig. 5 is not suitable for implantation, limited by its size. Therefore, it is necessary to integrate the circuit for implanted recording. Thus, a single-chip and low-power CMOS neural signal detecting amplifier had been designed and tested[9]. The chip was designed in 0.6 μm CMOS 2P2M technology of CSMC and simulated in SILVACO's Smartspice. The chip was laid out and verified with a PDK (process design kits) developed by Institute of RF- & OE-ICs, Southeast University, in Huada's Zeni layout environment. The chip was taped out via MPW (Multi Project Wafer) service of ICC, Shanghai. The chip size was 0.86 mm×1.2 mm, as shown in Fig. 7.

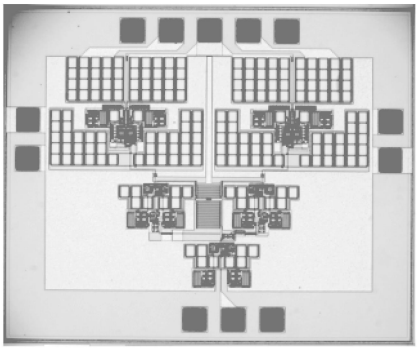


Fig. 7. An implantable single chip and low power neural signal detecting CMOS amplifier.

The chip had been tested on wafer in a time domain. The measurement instruments include a Cascade Microtech probe station, an Agilent 33220A arbitrary waveform generator and a Tektronix TDS5104 oscillograph. Test results showed that the neural detecting amplifier could operate under a single supply of 2.5±1.25 V with a power consumption of 180 μW. The gain reached 10000. The 3 dB bandwidth was 9.5 kHz. The slew rate was 16.7 V/μs and 114 V/μs for positive and negative slope, respectively. The output swing was 2.5 V and the input offset voltage was 3.2 mV (Fig. 8).

Further, the chip will be packaged for frequency response measurements and implanted neural signal detecting experiments. Another neural signal detecting CMOS amplifier, whose circuit design completely follows the module as a prototype, had already been taped out.

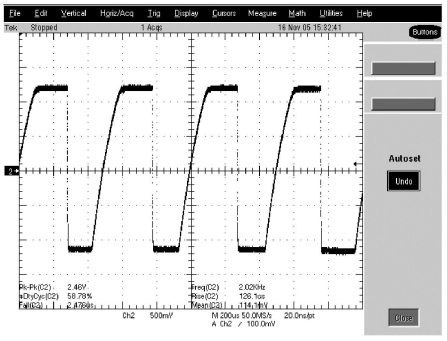


Fig. 8. The output waveform when applying 2 kHz 20mVpp square wave as input.

6 Conclusion

A four-channel neural signal detecting module for *in vivo* recording was designed and fabricated and successfully applied in animal experiments. An implantable neural signal detecting amplifier in 0.6 μm CMOS technology was designed and tested. The design and fabrication of the module and the chip promote the exploration of the neural channel bridge-connection system and neural disease prosthesis.

References

1 Wise KD. Wireless implantable microsystems: coming breakthroughs in health care. IEEE Symp on VLSI Circuits Digest of Technical Papers, 2002, 106—109
2 Wang ZG, Lü XY and Gu XS. Research of central nerve signal recording, processing and regeneration with microelectronics devices. The 14th Conference on Neural Networks of China, Anhui, China, 2004, 10—15
3 Wang ZG, Lü XY, Li WY, et al. Study of microelectronics for detecting and stimulating of central neural signals. IEEE Proc of Neural Interface and Control, 2005: 192—200
4 Horch KW and Dhillon G. Neuroprosthetics: Theory to Practice. (Bioengineering and Biomedical Engineering S.). Singapore: World Scientific Publisher, 2002, 348—370
5 Stieglitz T, Beutel H, Keller R, et al. Flexible, polyimide-based neural interfaces, microelectronics for neural, fuzzy and bio-inspired systems. IEEE Conf on MicroNeuro, 1999, 112—119
6 Wang YF, Lü XY and Wang ZG. Microelectrodes for neural signal recording and functional electrical stimulation. Biomedical Engineering Foreign Medical Sciences, 2005, 28(3): 129—133
7 Spinelli EM, Pallas-Areny R and Mayosky MA. AC-coupled front-end for biopotential measurements. IEEE Trans. on Biomedical Engineering, 2003, 50(3): 391—395
8 Cai JX and Zhang WZ. Biomedical Electronics. Beijing: Peking University Publishing House, 1997, 82—85
9 Wang YF, Wang ZG, Lü XY, et al. Fully integrated and low power CMOS amplifier for neural signal recording. IEEE Proc of Engineering in Medicine and Biology, Shanghai, China, 2005, 5250—5253