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Light Addressable Potentiometric Sensor as Cell-Based Biosensors for Biomedical Application

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1. Introduction

One of most enduring topics in the field of biosensors and bioelectronics is cell-based biosensors, which are able to convert cellular biological effects to electrical signals, via living cells. As the archetypal interface between a biological and an electronic system, the research and development of cell-based biosensors are essentially dependent on the combined knowledge of engineers, physicists, chemists and biologists. In recent years, the miniaturization and expanding applications of cell-based biosensors in biology, environment and medicine fields, have drawn extensive attention.

Light addressable potentiometric sensor (LAPS) is a semiconductor device proposed by Hafeman in 1988, and it is now as commonly used as ISFET (Hafeman et al., 1988). LAPS indicates a heterostructure of silicon/silicon oxide/silicon nitride, excited by a modulated light source to obtain a photocurrent. The amplitude of this light induced photocurrent is sensitive to the surface potential and thus LAPS is able to detect the potential variation caused by an electrochemical even. Therefore, in principle, any event that results in the change of surface potential can be detected by LAPS, including the change of ion concentration (Parce et al., 1989), redox effect (Piras et al., 1996), etc. LAPS shows some advantages comparing to ISFET while constructing cell-based biosensor. The easier fabrication process of LAPS is fully compatible with the standard microelectronics facilities. The encapsulation of LAPS is much less critical since no metal contact is formed on the surface. Besides, the extremely flat surface makes it compatible to incorporate into very small volume chamber, which is important for small dose measurement. Therefore, LAPS seems promising for biomedical application.

Due to the spatial resolving power, LAPS also has an advantage for array sensing application (Shimizu et al., 1994). Usually, no additional sensor structure is needed to realize the LAPS array sensing. In fact, LAPS is an integrated sensor itself, whose integration level is defined by the spatial resolution and the illuminating system. Thus, miniaturization with high integration level can be achieved. Many efforts have been drawn on the integration of LAPS (Men et al., 2005; Wang et al., 2005). Among these attempts, most are focused on the

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multi-sensing of different ions. Our lab proposed an electronic nose with MLAPS for environmental detection, which can detect H^+, Fe^{3+} and Cr^{6+} simultaneously (Men et al., 2005). Schooning et al. proposed a 16-channel handheld pen-shaped LAPS which can detect pH of 16 spots on the surface (Schooning et al., 2005). For biomedical sensing, our lab reported a novel microphysiometer to detect several different ions in cell metabolism (Wu et al., 2001a). Besides integrating LAPS to detect different ions, other possible attempts are also performed to integrate both abilities of ion concentration detection and extracellular potential signal detection, although it is still a long term from realistic application (Yu et al., 2009).

While constructing cell-based biosensors, one of the biggest obstacles is that the target cells are required to be deposited on predetermined electrodes. Due to the light addressing ability, the light addressable potentiometric sensor (LAPS) can overcome this geometrical restrict, which makes LAPS an outstand candidate among various cell-based biosensors. LAPS show great potential for constructing miniaturized and integrated biosensors. One promising solution is the LAPS array for integrated cell-based biosensors. By combining the IC techniques, mechanisms, and signal sampling methods, the LAPS array system has been greatly improved and miniaturized for biomedical applications.

LAPS as cell-based biosensors are able to perform longtime monitoring of several different cell physiology parameters, including extracellular acidification rate, various metabolites in extracellular microenvironment and action potential. These distinguish functions provide LAPS some promising applications in biomedical fields, such as cell biology, pharmacology, toxicology, etc (Parce et al., 1989; Mcconnell et al., 1992; Wada et al., 1992; Hafner, 2000; Wille et al., 2003). Furthermore, the multi functions of LAPS array as integrated cell-based biosensors makes the LAPS array system a good platform for drug analysis.

This chapter covers design and fabrication rules, systems and applications of LAPS. LAPS as cell-based biosensors are described in details, including principle, developments, and applications. Promising aspects and developments in miniaturization of LAPS array systems are introduced for cell-based biosensors. Applications of LAPS as cell-based biosensors are provided in biomedical fields, including the interaction of ligands and receptors, drug analysis, etc. Some future developments of LAPS as cell-based biosensors are proposed in the last part of this chapter.

2. Principle

LAPS is a semiconductor based potential sensitive device that usually consists of the metal-insulator-semiconductor (MIS) or electrolyte-insulator-semiconductor (EIS) structure. As for constructing cell-based biosensor, electrolyte is needed for cells living, thus LAPS with EIS structure is always adopted. LAPS with EIS structure is schematically shown in Figure 1A. The LAPS consists of the heterostructure of $Si/SiO_2/Si_3N_4$. An external DC bias voltage is applied to scan the EIS structure to form accumulation, depletion and inversion layer at the interface of the insulator (SiO_2) and semiconductor (Si), sequentially. When a modulated light pointer illuminates the bulk silicon, light induced charge carriers are separated by the internal electric field and thus photocurrent can be detected by the peripheral circuit. The amplitude of the photocurrent depends on the local surface potential. By detecting the photocurrent of LAPS, localized surface potential can be obtained (Hafeman et al., 1988).

The basic function of LAPS is for pH detection. Usually, a layer of Si_3N_4 is fabricated on the surface of LAPS as the H^+-sensitive layer. According to the site-binding theory, a potential

difference which is related to the concentration of H+ in the electrolyte forms at the interface of insulator (Si_3N_4/SiO_2) and solution (Siu et al., 1979; Bousse et al., 1982). This potential is coupled to the bias voltage applied to the sensor. Larger concentration of H+ provides a larger value of this potential difference, causing the I-V curve to shift along the axis of bias voltage (Figure 1B). When the bias voltage keeps constant in the middle region, change of the photocurrent indicates the pH change of the electrolyte. With the microchamber specified for biological assay, the extracellular acidification rate of cells can be monitored in the microenvironment by the commercialized Cytosensor™ Microphysiomter system.
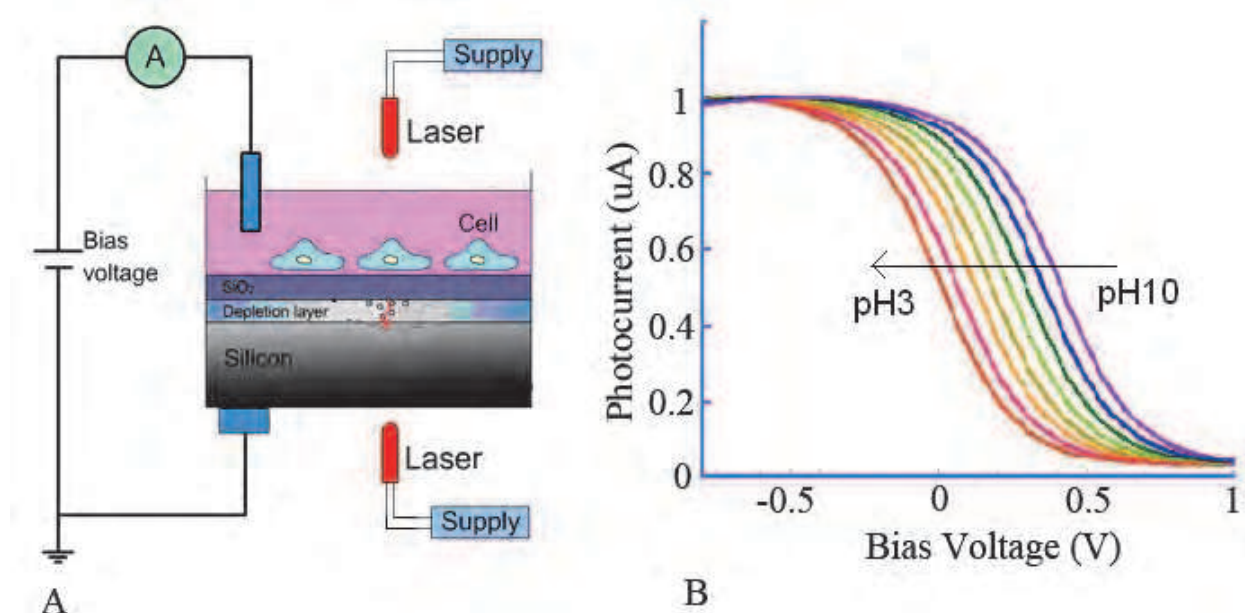

Fig. 1. (A) Working principle of the LAPS. (B) Characteristic I-V curves of n-type LAPS

Beside the pH detection, attempt has been made for the extracellular detection of electrical signals. LAPS is a surface potential detector with spatial resolution. Light pointer used for LAPS detection can be focused by microscope and optical lens, which suggests the LAPS possible for cell analysis on any non-predetermined testing site. After cells are cultured on the LAPS, a focused laser, 10 μm in diameter, is used to illuminate the front side of the chip to address the cells to be monitored. Excitable cells such as cardiac myocytes or neuron cells can generate extracellular action potential. This potential is coupled to the bias voltage applied to the LAPS, which correspondingly changes the amplitude of the photocurrent. Thus, by monitoring the photocurrent at a constant bias voltage, extracellular potential signals can be detected (Xu et al., 2005).
Illuminating different sensing areas with modulated lights of different frequencies generates a photocurrent signal, from which corresponding information of each testing site can be obtained by FFT (Fast Fourier Transform) methods (Cai et al., 2007). Comparing with conventional surface potential detectors such as FET or MEA, integration of LAPS array has many advantages. The most important feature of LAPS array is the great reduction of the required leads. For MEA, the number of required leads is the same as the number of electrodes, while for LAPS array, only one lead is necessary, regardless of the number of testing sites, which is important for high level integration (George et al., 2000). Besides, LAPS can detect extracellular potential as well as ion concentrations (Wu et al., 2001b),

which makes it suitable for multi-functional integration. Sensing surface of LAPS is extremely flat and free of metal contact. Thus it's easy for encapsulation of LAPS array and fabrication of micro testing chamber.

3. Device and system

The LAPS device is a typical EIS structure. Fabrication procedure is easy and fully compatible with standard microelectronics facilities. We have introduced in our publications the most commonly used LAPS device and system (Xu et al., 2005). In this section, we mainly introduce the devices and fabrication process of LAPS array sensors.

3.1 Devices

As mentioned before, the LAPS can be treated as an array sensor with no extra structures due to the spatial resolution. However, since only a little part of the LAPS surface is illuminated with the modulated light pointer, unilluminated parts, where no photocurrent flows, act as stray capacitance and cause noises. Therefore, the smaller the total capacitance of the device is, the better the potential sensitivity will be. Small effective areas as well as a thick insulating layer reduce the total capacitance, and thus improve the potential sensitivity. Nevertheless, by reducing the effective LAPS surface to small areas, the advantage of the LAPS against surface potential detectors with discrete active sites is lost (George et al., 2000). According to our experience in cell experiments, we found 200µm×200µm a compromised size between cell culture and the noise level (Xu et al., 2006). One typical structure of the integrated LAPS array sensor reported for multifunctional detection of extracellular pH detection and extracellular potential signals is schematically shown in Fig.2 (a). (Yu et al., 2009) The chip has a total area of about 1cm×2cm. Testing areas of two different sizes are fabricated on the same silicon chip by heavily doping the silicon between the testing areas. For extracellular potential signal detection, about 400 small square wells were fabricated in size of 200µm×200µm and the plateau between two adjacent testing areas was 100µm in width. Cells were cultured on the areas with small wells for potential detection. The depth of the well shaped structure was about several hundred nanometers, and we found that cells are more likely to grow on the testing areas of the arrays, which had lower altitude. Four larger wells for detection of cell acidification were 3mm×3mm in size and 1mm away from each other.

The fabrication process of such LAPS array structures was shown in Fig.2(b). A p-type silicon wafer (thickness of 450µm) with <100> crystal orientation was used. First, a thick layer of silicon oxide was thermally grown on the surface. Then, after the pattern was transferred to the surface using photolithography, all silicon oxide, except that grown on testing areas (acting as a protector of substrate at testing areas from being doped in the following step), was removed by etching. Thermal diffusion doping was then carried out. As the silicon wafer is p-type, boron was selected as the impurity. There were two steps in doping process. First, a glass layer containing boron was pre-deposited on the sensing surface. Then pre-diffusion doped the surface of silicon to a small depth. After pre-diffusion, the glass layer was removed, followed by the redistribution step. During the redistribution step, a thick layer of silicon oxide about several hundred nanometers formed on the surface of doped areas, which participated in forming a well shaped structure. The doped part of the semiconductor was several micrometers in depth to cut off the depletion layer of adjacent detection sites. After the doping procedure, silicon oxide layer on testing areas was

removed. Instead, a thin layer of silicon oxide, 30nm in thickness, was thermally grown on testing sites and then a 60nm silicon nitride layer as the sensitive layer to H+ was deposited on the sensor chip by PECVD. At last, a thin layer of aluminum (thickness of 200nm) was evaporated on the backside of the silicon chip to form an ohm contact.
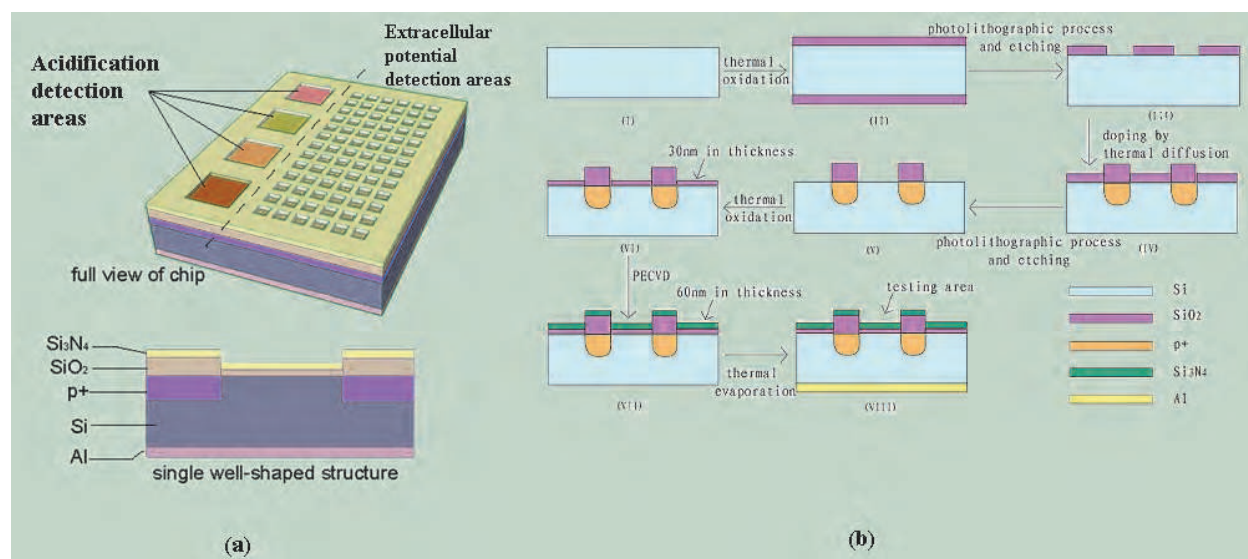

Fig. 2. (a) Schematic diagram of LAPS array sensor structure: the upper one is the full view of chip; the lower one is the well-shaped structure of single testing area. (b) Fabrication process of the LAPS array.

3.2 System

The LAPS system usually requires LED driver, chemical working station, lock-in amplifier, sampling card, flow control system, etc. In our work, a potentiostat (EG&G Princeton Applied Research, M273A) is employed to control the bias voltage across the silicon bulk to form the depletion layer inside. In running process, the bias voltage of LAPS is applied to the platinum counter electrode versus the silicon working electrode and the photocurrent flows through the working electrode to be detected by peripheral equipment. Preamplification is also performed in the potentiostat, which transforms the current signal into potential signal.

In LAPS system, the surface potential signal is amplitude modulated by the high frequency light signal, resulting in the high frequency photocurrent signal. Thus, to obtain the original surface potential signal, demodulation is required after preamplification. Lock-in amplifier is always used for small signal detection, as it can greatly increase the signal to noise ratio (SNR), usually an improvement of the SNR for over 10^6 times. In Our system, the lock-in amplifier (Stanford Research System, SR830) is employed. The lock-in amplifier only detects the signals in narrow band near the object frequency, determined by the reference frequency. Thus, in order to get corresponding surface potential signal from the photocurrent signal, it is important to keep the internal reference frequency exactly the same as the carrier frequency of the photocurrent signal, which is the light frequency. The laser generator supply is controlled by external reference signal generated by lock-in amplifier, which has the same frequency as the internal reference signal used for demodulation. Therefore, the result of the lock-in amplifier includes the amplitude and phase information of the photocurrent signal, which reflects the change of the surface potential signal of the

LAPS chip. After signal demodulation by lock-in amplifier, data is then sampled by a 16-bit acquisition card to the computer for data screening and further processing by the software. Programming can be performed with different programming languages, among which, labVIEW is recommended.

For LAPS array detection, different LAPS array system were established. The simple way is to scan the light pointer along the LAPS devices. Each sample contained information at corresponding detecting area. However, this solution suffered from low resolution and long scanning time, which prevented it from wide application (Nakao et al., 1996). An alternative way to perform the LAPS array detection is using multi light sources as the illumination. Several light sources were modulated at different frequencies and illuminating different area of the LAPS devices. In this situation, each sample contained information of several detecting areas. To extract each signal from the overall mixed signal, Fast Fourier Transform (FFT) technique is a preferred way. Our lab has reported a novel design that could significantly increase the measurement rate of LAPS (Zhang et al., 2001). By illuminating the LAPS simultaneously at several different positions, each of which is illuminated with a light pointer modulated with different frequencies, the surface potential at all illuminated regions can be measured simultaneously by analyzing the resulting photocurrent. Using this method, the rate to obtain a complete image of the surface potential distribution across a LAPS wafer can be drastically increased compared to the conventional system. However, the multi-light LAPS needs to equip a signal generator for each light source. To obtain an 8×8 image, the system needs to provide 64 signal generators. With LEDs as the light sources, this system has a lower resolution and precision. So this method is unsuitable for accurate imaging. Moreover, the problem lies in the big volume of the illuminating system, which was a main obstacle for highly integrated system, and the longer time for digital demodulation, which is not suitable for fast detection such as the detection of extracellular action potential.

Researchers have paid attentions in solving the problems in constructing LAPS array system. Our lab also has presented a novel imaging system, shown in Figure 3. With microlens array, a single laser is separated into a focused laser line array. Every focused laser is modulated separately to a different settled frequency. With a line-scanning control, an 8×8 image can be obtained that only needs 8 scanning. Moreover, with different sensing materials, this device can be used to detect several components of sample in parallel. (Cai et al., 2007)

To illustrate the constructing of LAPS array system for cell-based biosensors, our system for multi detection of cellular parameters were shown in Fig.4 (Yu et al., 2009). A laser light with the wavelength at 690nm (red) was used for illumination of extracellular potential detection. The laser was modulated at 10kHz by the lock-in amplifier (SR830, Stanford Research System), and the power is about 0.2mW. The laser was focused to about 10µm in diameter through an optimized microscope so that it can be used to address a cluster of cells on the sensor chip. Four LEDs with the wavelength at 625nm (red) were respectively driven at four different frequencies of crime numbers to avoid harmonic interference with a power of 50mW. These LEDs illuminated the relative four testing areas for acidification detection. These five lights illuminated the sensor chip at the same time. A photocurrent signal including signals at all these five different frequencies was generated, respectively representing information of the five different testing sites. The detecting system was designed to sample the overall signal and extract signals at the five different frequencies.
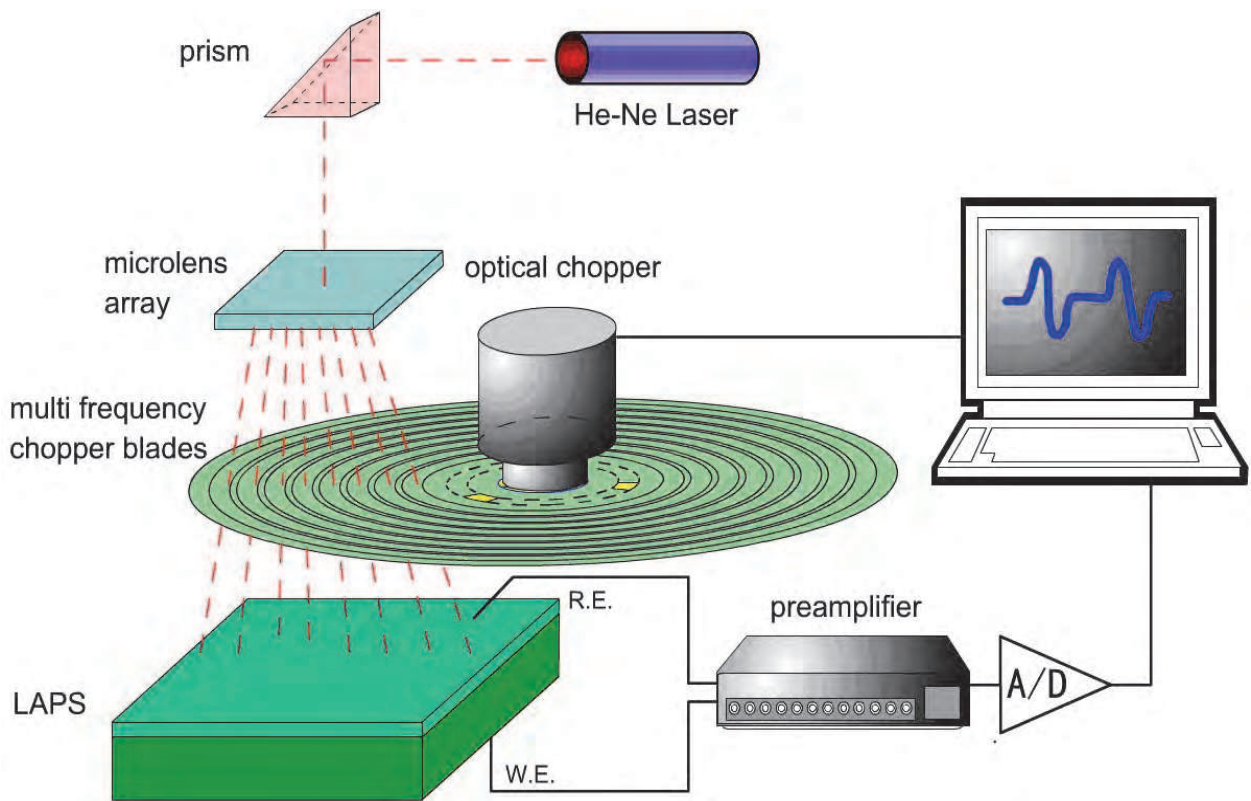
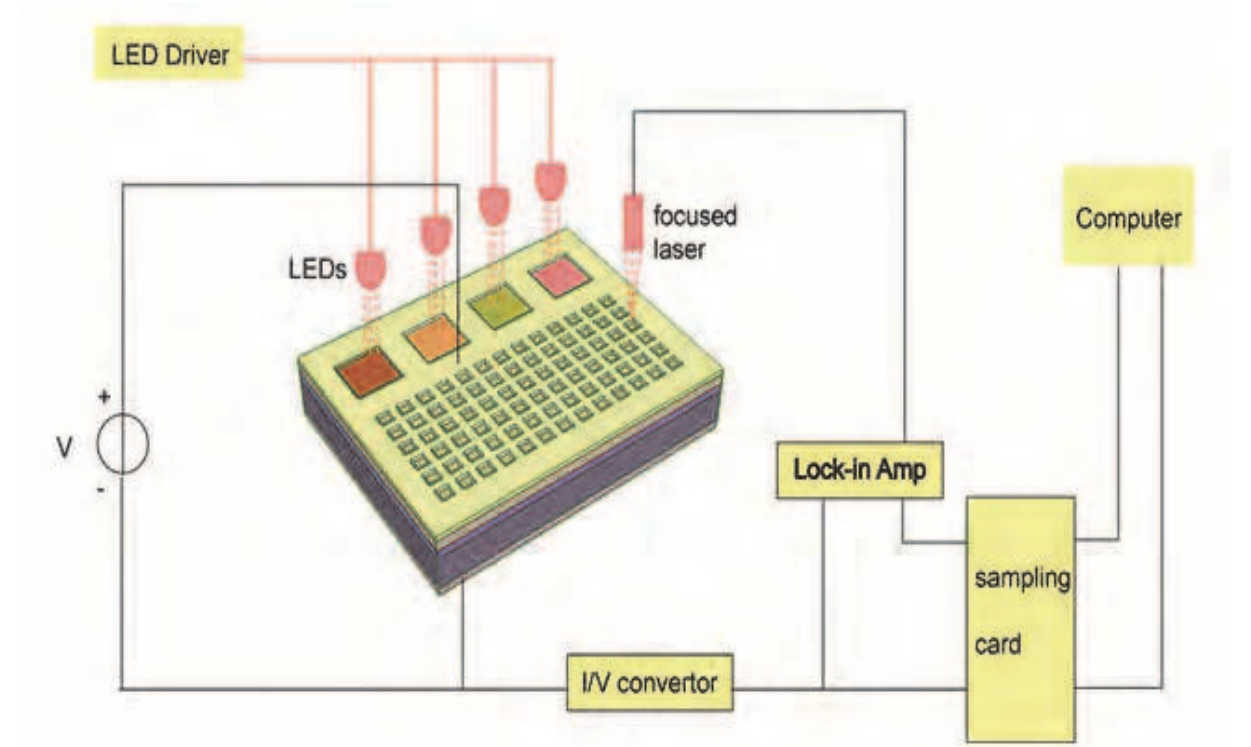Fig. 3. Schematic diagram of the line-scanning light sources based on microlens array.

Fig. 4. Multi-functional LAPS system for simultaneously detecting extracellular acidification and extracellular potential signals.

Basically, single spike of action potential recorded lasts only several hundred milliseconds or even less (Sprössler et al., 1999; Fromherz.P et al., 2002; Sprössler et al.,1998). Thus, the sampling frequency should be high enough not to miss any action potential signals. Besides, for real-time monitoring of extracellular potential signals, time delay between sensing and display was preferred to be as small as possible. It's a different situation for acidification detection. Usually, the extracellular pH change is a long time effect. Accumulation of H+ in extracellular environment will not cause significant signals until several minutes (Hafner, 2000). Thus, for acidification detection, time delay between sampling and display was less critical.

Usually, there were several seconds interval between two times of sampling. A potentiostat (Model 273A, EG&G) was used to apply the bias voltage to the sensor chip and perform the I/V-converting. Due to the different requirements in the time delay, two different methods were combined for signal recording. For extracellular potential detection, after the overall photocurrent signal was I-V converted, the lock-in amplifier was used. As the laser was modulated at 10kHz by the internal reference signals of the lock-in amplifier, the output of the lock-in amplifier was also the component at 10kHz. Thus, only the signal generated at the testing site illuminated by the focused laser was preserved and demodulated, which indicated the extracellular potential signal. High sampling frequency up to 100kHz was set to monitor the action potential. The lock-in amplifier can perform a fast demodulation of signals, and thus little delay was introduced for real-time monitoring.

For acidification detection, after the overall photocurrent signal of five different frequencies was converted to a potential signal, it was directly sampled to the computer for analysis. Signals generated at the four different sensing areas were gained separately by digitally demodulating the signal by software with FFT methods (Cai et al., 2007) at respective illuminating frequencies of the four LEDs, which were four different crime numbers. The overall signal was also sampled at 100kHz. Data of one second was sampled every five seconds. Thus, there was four seconds for the software to perform digital demodulation of the signals at these four different frequencies and then display each part.

4. Application

LAPS has many advantages for constructing cell-based biosensors. Since the first publishing of the Cytosensor™ Microphysiometer, it has been widely used by researchers. Besides, the newly proposed cell-semiconductor LAPS device for extracellular potential detection is considered as a useful tool for cell electric biology study. Applications of LAPS for cell-based biosensor are introduced in cell biology, pharmacology, toxicology, environment measurement, etc. Several reviews have been published to introduce the applications of the microphysiometer (Parce et al., 1989; Mcconnell et al., 1992; Wada et al., 1992; Hafner, 2000; Wille et al., 2003). In this section, the application of LAPS sensors, especially the LAPS array biosensors for drug analysis, was introduced.

4.1 Multi-parameter monitoring of cell physiology by LAPS array for drug analysis

The primary functions of LAPS as cell-based biosensors are monitoring the extracellular acidification. Researchers have been working with the Cytosensor Microphysiometer on various aspects including the ligand/receptor binding, pharmacology, toxicology, etc. However, the microphysiometer suffered a major problem that only H+ can be monitored. In recent work, the microphysiometer was usually used together with other instruments for

biological detection. To solve this problem, getting more information about the multi-functional cellular processing of input- and output-signals in different cellular plants is essential for basic research as well as for various fields of biomedical applications. Therefore, research work with LAPS for extracellular potential detection and multi-parameter detection of cell physiology was preferred.

Liu et al. have constructed a cell-semiconductor hybrid device for some applications in drug analysis (Liu et al., 2007a). As an agent of β-adrenoceptor agonist that contributes to cardio-activity, isoproterenol (ISO) enhances the L-type calcium channel activity, which caused an increase in Ca^{2+} signal. As shown in Figure 5, it is obvious that after administration of ISO, the beating frequency, amplitude and duration of cardiomyocytes were all increased in a dose-depended manner (0.1, 1, 10 µM). The cellular contractibility all recovered after washing drugs out at above concentrations. Whereas, as a negative one, carbamylcholine (CARB) had opposite effect to ISO, increasing K^+ conductance in cardiacmyocytes, and signals indicated a decreasing trend. Figure 5A showed the changes of curves to ISO and CARB at concentration of 1 µM. We could see that the parameters display the two drugs distinct. Furthermore, if we differentiated parameters to each stroke shown in Figure 5BCD, more approving results could be got. According to those changes, we know that ISO and CARB have direct effects on the duration and amplitude of the strokes 2 and 3, which accord with the pharmacological increase of the Ca^{2+} or K^+ ion current, respectively. Thus, cooperated with Na^+, K^+ and Ca^{2+}, targets of a concrete drug can be evaluated synchronously by the biosensor system.

The concentrations of the extracellular ions, such as Na^+, K^+, Ca^{2+}, may change along with the alteration of cell physiology. In order to analyze simultaneously the relations of the extracellular environmental H^+, Na^+, K^+, Ca^{2+} under the effects of drugs, our lab has developed a novel microphysiometer based on multi-LAPS (Wu et al., 2001a; Wu et al., 2001b). The surface of the LAPS is deposited with sensitive membranes by silicon microfabrication technique and the poly- (PVC) membrane technique. Three different sensitive membranes are illuminated in parallel with light sources at different frequencies, and measured on-line by parallel processing algorithm, Figure 6A. Different sensitive (H^+, K^+, Ca^{2+}) membrane is illuminated on the sensor, simultaneously with three light sources at different frequencies (3kHz for K^+, 3.5kHz for Ca^{2+}, 4kHz for H^+). The photocurrent comprises these three frequency components, and the amplitude of each frequency component might be measured on-line by software FFT analysis, as shown in Figure 6B. Dilantin, i.e. phenytoin sodium, a sort of anti-epilepsy drugs, has significant effects of transqulizing and hypnotic and anti-seizure. Moreover, dilantin is also one of the anti-arrhythmia drugs. It is proved that dilantin has membrane stabilizing action on neural cells because it can reduce pericellular membrane ions (Na^+, Ca^{2+}) permeability, inhibit Na^+ and Ca^{2+} influx, stave K^+ efflux, thus, prolong refractory period, stabilize pericellular membrane, decrease excitability (Figure 6C).

Besides combining detection of different metabolites, integration of different functional biosensors is also attractive. In our work, we have proposed a LAPS array system for simultaneously detection of both the acidification rate and the extracellular signals []. Although this system is some distance from realistic application for drug screening, this integrated cell-based biosensor can be used for simultaneously detection of both the acidification rate and the extracellular signals under certain drug effect. Comparing to

conventional microphysiometer, this system combined both the electrical signal and the metabolism signals of cells, which could be of great help in analyzing the cellular response to drugs.
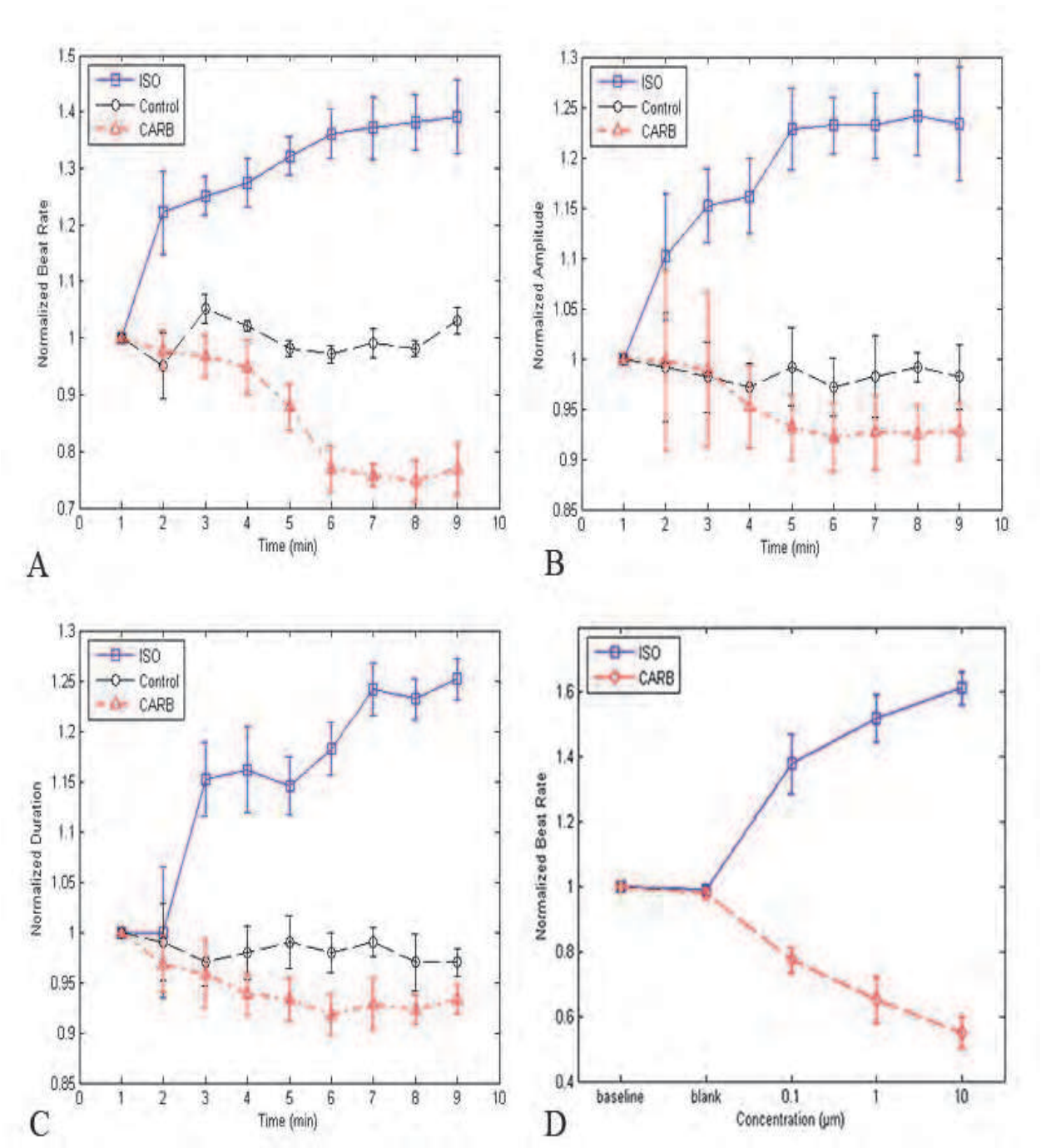

Fig. 5. Plots comparing the response of cardiomyocytes to the carbamylcholine (CARB), isoproterenol (ISO) and physiological solution as control. The concentration of drugs are all 1 μM. Drugs effect on the beat rate (A), amplitude (B) and duration (C) of each extracellular potential. Effect of different drug concentration to beat rate (D). Each data point represents an average over 50 s. The experimental data is the average value of six times of repetition.
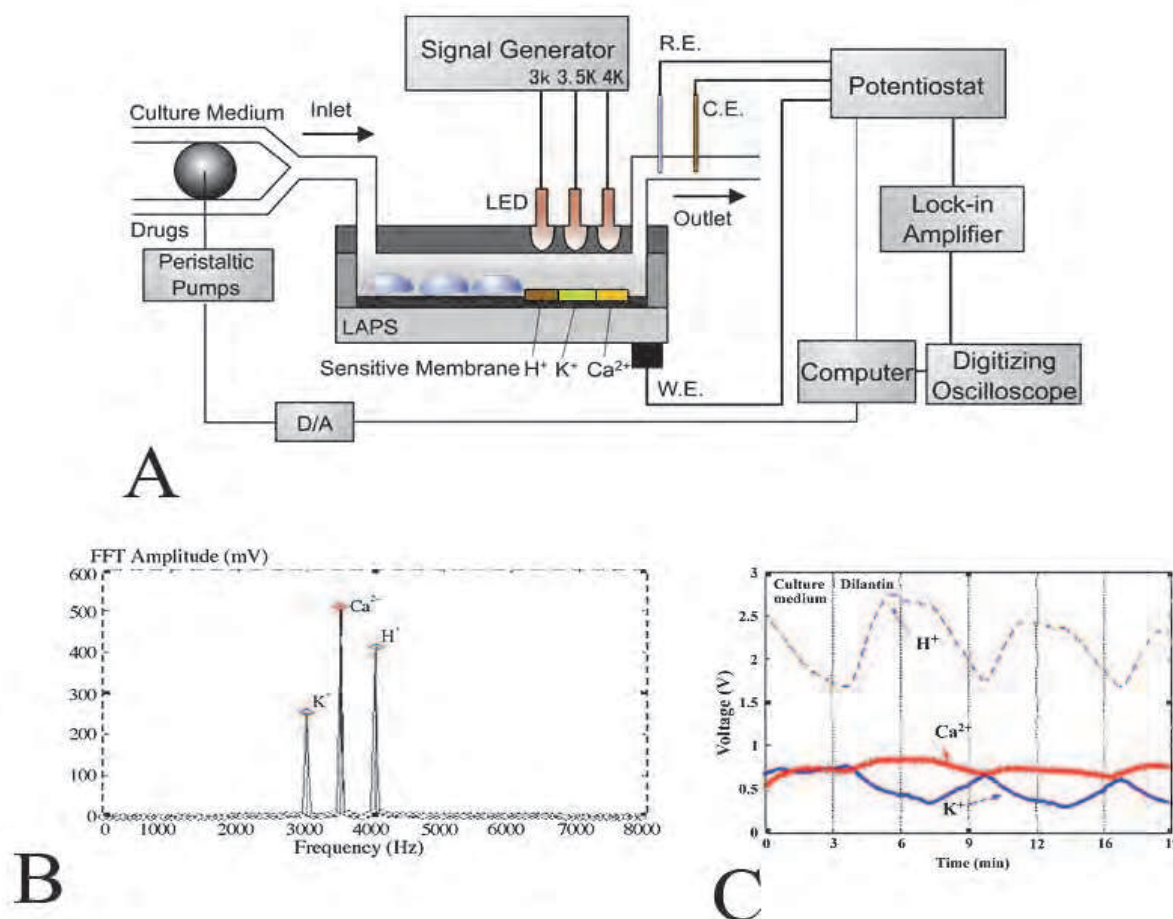
Fig. 6. Microphysiometer studies based on multi-LAPS. (A) The schematic drawing of the system of the multi-LAPS to different extracellular ions (H+, K+, and Ca2+). (B) Illuminate simultaneously at the three sensitive membranes with three light sources at different modulated frequencies. (C) H+, K+, Ca2+ analyzed simultaneously by multi-LAPS.

4.2 LAPS for environment monitoring

Environment monitoring is a very important aspect in LAPS application. In our work, we mainly treated situation with heavy metal ion. We have reported the electronic tongue system with LAPS for heavy metal ions monitoring in sea water [][]. However, this system requires the ion sensitive membranes of corresponding heavy metal ions which increases the cost and was indirect to study the effect of the target sea water to the biological object.

The LAPS biosensor system has been reported to detect heavy metal ions according to changes in parameters describing spontaneous beating of cardiomyocytes under the different toxic effects (Liu et al., 2007b). The effects of heavy metal ions on cell function were evaluated by comparing the changes of the sensor signals before and after the cells were exposed to the toxins. Figure 7 shows the change of frequency, duration and amplitude of the signals after the addition of 10 M heavy metals for each type (Fe^{3+}, Hg^{2+}, Pb^{2+}, Cd^{2+}, Cu^{2+} and Zn^{2+}). Exposure of beating cardiomyocytes to 10 μM Fe^{3+} decreases the frequency, amplitude and duration to about 50% of the basal signal. Similar curves were found for Pb^{2+} and Cd^{2+} solutions with a smaller decrease of amplitude and duration, however a slight

progressive increase of frequency was observed. On the contrary, the three parameters all increased in Hg^{2+} solution. There were no apparent trends with regard to Cu^{2+} and Zn^{2+} toxic effects on measured parameters (only duration on Zn^{2+} showed a slight increase). Comparing with biosensors using pure enzymes, cell-based biosensors, which use whole cells as the bio-recognition elements, LAPS for biosensors can detect agents functionally (Bousse, 1996; Stenger et al., 2001). Metal ions are found to have effects on the cellular organelles and components, such as cell membrane, mitochondrial, lysosome, endoplasmic reticulum, nuclei, and some enzymes involved in metabolism, detoxification, and damage repair (Squibb and Fowler, 1981). All these systems are considered to influence metal induced cellular responses simutaneously. Therefore, incorporated with whole cells, cell based biosensors would offer potential physiological monitoring advantages over devices based on isolated enzymes or proteins. And with the help of living cells, especially mammalian cells, we could not only detect but also evaluate toxicities of heavy metals with cellular physiological changes.
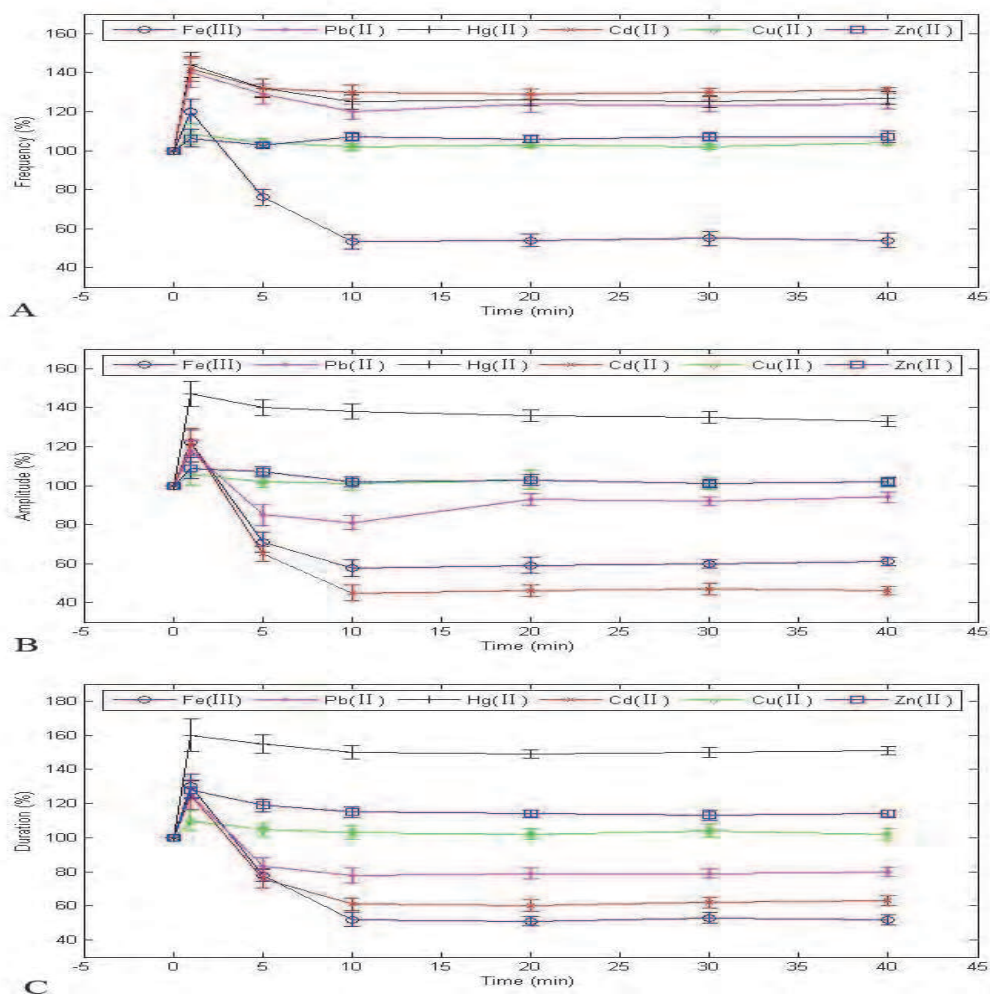

Fig. 7. Plots comparing the response of cardiomyocytes to the carbamylcholine (CARB), isoproterenol (ISO) and physiological solution as control. The concentration of drugs are all 1 µM. Drugs effect on the beat rate (A), amplitude (B) and duration (C) of each extracellular potential. Effect of different drug concentration to beat rate (D). Each data point represents an average over 50 s. The experimental data is the average value of six times of repetition.

4.3 LAPS as bioelectronic nose and bioelectronic tongue

Göpel and his colleague first proposed utilizing olfactory neurons as sensitive materials to develop a bioelectronic nose (Gopel et al., 1998; Ziegler et al., 1998; Gopel et al., 2000). They suggested the biomolecular function units can be used to develop highly sensitive sensors (like the dog's nose to sense drugs or explosives) as one of independent trends for electronic nose or tongue chip.

When olfactory receptors were expressed on the membrane of a heterologous cell system, the binding of olfactory receptors with specific odorant molecules can be detected by QCM or SPR. However, these cells were not excitable, so the action potentials produced by the interaction of receptors and glands can not be detected. Here will present the olfactory and taste cell-based biosensors in our laboratory. The implementation of olfactory and taste cell sensors system based on LAPS with artificial olfactory and artificial taste sensor system for odor and ion sensor array are described (Liu et al., 2006; Wang et al., 2007; Zhang et al., 2008).

Besides the cell-based biosensors, tissue-based biosensors with LAPS were reported as the bioelectronic nose, shown in Figure 8 (Liu et al.,2010a; Liu et al., 2010b). The study engaged in designing an olfactory epithelium tissue and semiconductor hybrid neuron chip to investigate the firing modes of the olfactory receptor neurons under different odor stimulations, shown in Figure 9. The theory model of olfactory epithelium coupled to LAPS surface after odor stimulation was established and simulated. Extracellular potentials obtained before and during odor stimulation could be analyzed on basis of local field potentials and differentiated by PCA. All the results reported suggest that the olfactory receptor cells respond to odors in a tissue and semiconductor hybrid neuronchip will be a novel bioelectronics nose with great potential development.
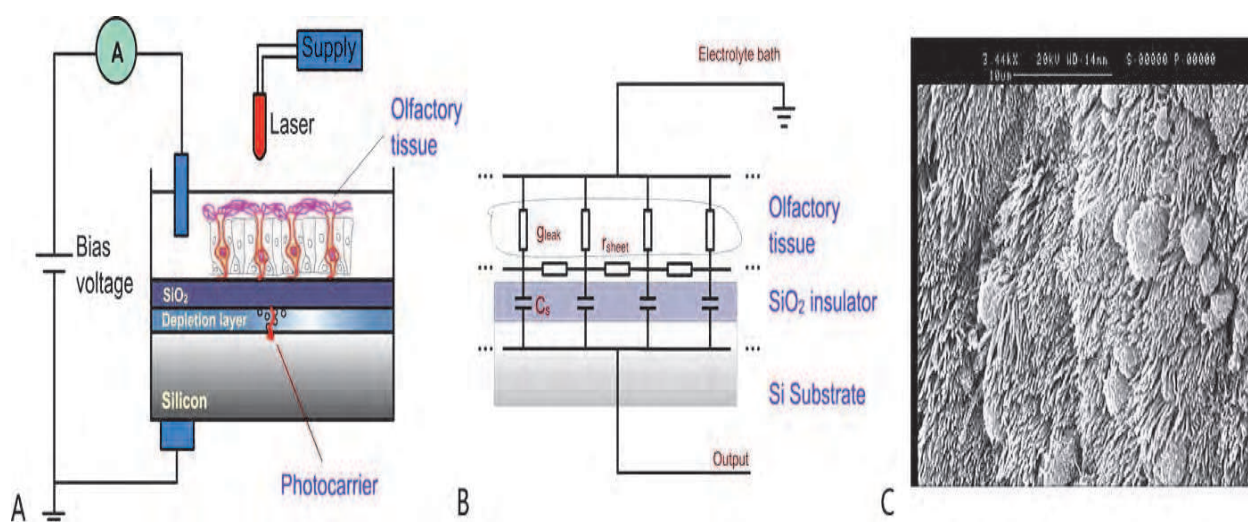

Fig. 8. Olfactory epithelium tissue hybrid with LAPS. (A) LAPS system with the olfactory epithelium on the sensor surface. (B) Sheet conductor model on extracellular potentials recording of the tissue layer between electron conductor and electrolyte bath on LAPS. (C) Olfactory epithelium tissue fixed on the surface of LAPS observed by the scanning electron microscope.
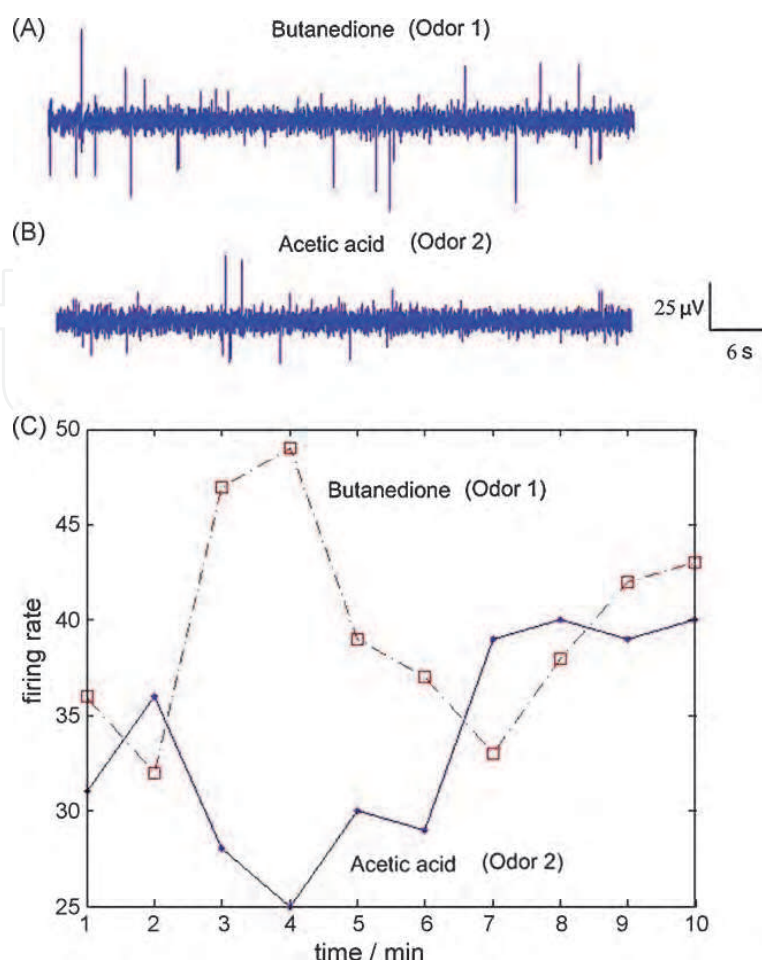
Fig. 9. Responses of the extracellular potential changes of olfactory mucosa tissue to odors of butanedione (A) and acetic acid (B), with different discharge models (C).

5. Conclusion

In this chapter, we engaged the light addressable potentiometric sensor (LAPS) as the cell-based biosensors for biomedical application. The main purpose was to introduce the principle, devices, fabrications, detection systems and applications of the LAPS sensor, especially the recent concept of constructing LAPS array for biomedical application. LAPS has its own advantages while constructing cell-based biosensors and showed promising prospect for application in various areas such as biomedical, environmental, food safety, etc. LAPS array can be used for highly integrated sensors system, which is essential in high throughput drug screening. By improving the sensitivity, spatial resolution, sampling rate, LAPS array could be a super candidate for constructing sensor systems, especially cell-besed biosensors.

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Biosensors for Health, Environment and Biosecurity

Edited by Prof. Pier Andrea Serra

A biosensor is a detecting device that combines a transducer with a biologically sensitive and selective component. Biosensors can measure compounds present in the environment, chemical processes, food and human body at low cost if compared with traditional analytical techniques. This book covers a wide range of aspects and issues related to biosensor technology, bringing together researchers from 16 different countries. The book consists of 24 chapters written by 76 authors and divided in three sections: Biosensors Technology and Materials, Biosensors for Health and Biosensors for Environment and Biosecurity.

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