Paper

An organic film/SiO_2 /Si heterostructure as a novel biological interface of a light-addressable potentiometric sensor.

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A light-addressable potentiometric sensor (LAPS) with Poly-L-Ornithine with Laminin (PLOL)-coated SiO_2 layer as the only insulating material on the bulk Si in SiO_2/Si structure, is investigated as a possible biological interface. Comparing with the conventional structure (Si_3N_4/SiO_2/Si) of the LAPS, much steeper photocurrent response was observed for the PLOL-coated SiO_2/Si structure, which shows higher surface potential sensitivity and pH resolution. PLOL-coated SiO_2 layer is kept soaked with the electrolyte for ten days and no appreciable change in the surface potential sensitivity was noticed. The pH sensitivity of the sensor was investigated in the pH range of pH4 to pH10, and a nearly Nernstian sensitivity was observed.

Keywords: Light-addressable potentiometric sensor (LAPS), Surface modification, surface potential sensitivity, pH sensitivity, organic film.

1. Introduction:

Since the first report of Hafeman on the light-addressable potentiometric sensors (LAPS) in 1988 [1], LAPS has found its application in many chemical and biochemical fields. Mainly, the applications of LAPS has been limited to the sensing of the chemical elements that modifies directly or indirectly the surface potential due to the charge bound at the electrolyte-insulator and insulator-semiconductor interfaces [2-6]. Despite the unique feature of light addressable ability in responding to any potential in series with the external bias circuit [7] LAPS has been rarely [8] investigated as a biological interface for bi-directional and noninvasive transfer of information in the form of electrical potentials from the biological system. This is may be due to the present *Surface Potential Sensitivity* (SPS), and spatial resolution of the conventional LAPS to sense the very low extracellular potentials, e.g., extracellular transients of the electrical action potential of the neuron cells [9, 10]. The highest slope at the nearly linear region of the photocurrent response, which we define as the *Transduction Factor* (TF), determines the sensitivity of the LAPS to detect the tiny change in surface potential. The SPS is higher for the higher TF. The photocurrent response is obtained when the bias is scanned on the insulating surface to switch the LAPS from accumulation to inversion, with a modulated light shone at the bulk Si. The ionic sensitivity of LAPS is determined from the shift of the photocurrent response and is principally related to the ionic sensitivity of the top most layer, e.g., Si_3N_4, Al_2O_3, Ta_2O_5 [11] etc., for pH-sensitive LAPS. The slope of the photocurrent response [12] also determines the ionic resolution of the LAPS i.e., how small change in ionic concentration LAPS can detect.

The TF of the LAPS characteristic depends on many parameters, and mainly, on the doping level of the semiconductor, thickness of the semiconductor layer, thickness of the insulating layer, density of the interface state [13] etc, and accordingly, thinning the insulator layer should increase the TF of the LAPS. We investigated a thin layer of polymer (Poly-L-Ornithin with Laminin (PLOL))-coated SiO_2 layer on the bulk Si as a LAPS structure for a novel biological interface [14]. The PLOL-coating, which is very thin (1.5 - 2nm) [15], eliminates the necessity of using Si_3N_4 or other top insulators on SiO_2 layer, and thereby increases the TF. As a biological interface, the LAPS is expected to remain in contact with the electrolyte (cell-culture medium) that contains various ions, for long time i.e., the cell-culturing and recovering period. In this article, after theoretically analyzing the TF of the LAPS, the long-time study of TF in contact with the cell-culture medium is presented. The pH response of the novel LAPS in the range of pH 4 - pH 10 is also discussed.

2. Theoretical consideration and sensor preparation:

Theoretically it appears that the optimization of LAPS heterostructure of Si_3N_4/SiO_2/Si might provide the steeper photocurrent response and thereby provide higher resolution of the LAPS. The electrical equivalent circuit of the LAPS [16] is shown in figure 1. Where,

I_p = Light induced minority carriers
I = Measured photocurrent.
C_d = Depletion capacitance
C_i = Insulator capacitance

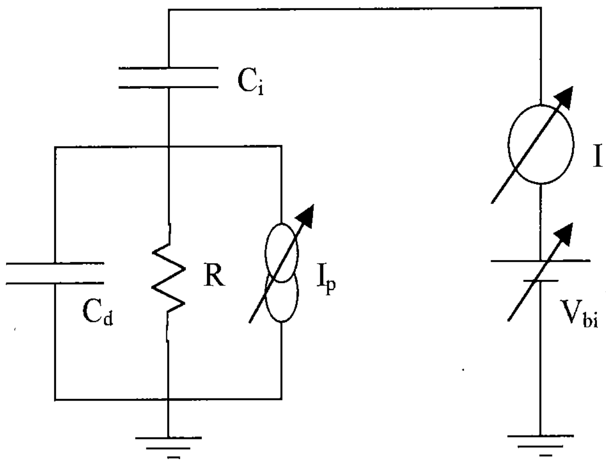

Figure 1. AC equivalent circuit of LAPS device

The measured current is given by

$$I = I_p \frac{j\omega C_i R}{1 + j\omega R(C_i + C_d)} \qquad (1)$$

The term $[2\pi R(Ci + Cd)]^{-1}$ is defined as the cutoff frequency for the LAPS. The measured current, at a frequency below the cutoff, is given by

$$I \approx j\omega C_i R I_p \qquad (2)$$

and at above the cutoff frequency

$$I = \frac{C_i}{C_i + C_d} I_p \qquad (3)$$

The equation (2) and (3) shows that, at a frequency below or above the cutoff frequency, the measured current can be increased by increasing the insulator capacitance C_i ($=\varepsilon_i/d$), and hence decreasing the thickness of the insulating layer. Conventionally the insulating layer of LAPS is a combination SiO_2 and Si_3N_4, and insulator capacitance of

$$C_{i(SiO2)} >> C_{i(SiO2+Si3N4)}$$

Therefore, using only SiO_2 as the insulating layer, the measured current and hence the TF can be increased. Regarding pH response, for various reasons, such as hydration, hysteresis, drift, and insulating properties, SiO_2 or Si_3N_4 alone cannot be used as insulating layer of LAPS [17]. But, SiO_2 alone can be used if it is prevented from getting hydrated in contact with the electrolytic solution. An organic polymer film on SiO_2 can do this job and we chose Poly-L-Ornithine with Laminin (PLOL) as the organic film for the following reason (15).

1) The coated layer can be made very thin e.g., 1.5 - 2nm.
2) Being a polymer, PLOL prevents leaking of water through its membrane and thus prevents hydration of the SiO_2 surface.
3) The attachment of the biological cells to the surface is very strong due to the negative surface charge of PLOL layer.

The LAPS is prepared as a heterostructure of thermally grown SiO_2 layer of 30nm thick as the insulating layer on the 225μm thick phosphorous-doped (n-type) Si <100> having resistivity of 1-10Ω-cm. The performance of this novel structure is compared with conventional type, i.e., Si_3N_4 + SiO_2 insulator layer-based LAPS. Both the SiO_2 and Si_3N_4 + SiO_2-based sensors are processed on the wafers of same physical specifications. After 30nm thick SiO_2 is thermally grown in a batch process, few sensors are separated and 100nm-thick layer of Si_3N_4 is deposited by LPCVD on the SiO_2 layer to form conventional LAPS sensor. PLOL is chosen for the coating of the SiO_2 surface.

The experimental setup is shown in figure 2, in which 830nm unfocused IR-LASER is used for illuminating the back surface of Si. The intensity of the LASER is modulated at frequency of 10kHz. The bias is applied through a DC power supply by an Ag/AgCl rod electrode and the photocurrent is converted to voltage by a pre-amplifier with a factor of 10^6 Volt/Ampere. A PC collects the data through a lock-in amplifier. PLOL-coated SiO_2 surface is sealed with silicone rubber sheet with only a circular opening (ϕ=8mm) just above the back-illuminated point, through which the surface contacts the electrolyte. The wafers were bought and processed from the Silicon Sense Inc., Nashua, USA.

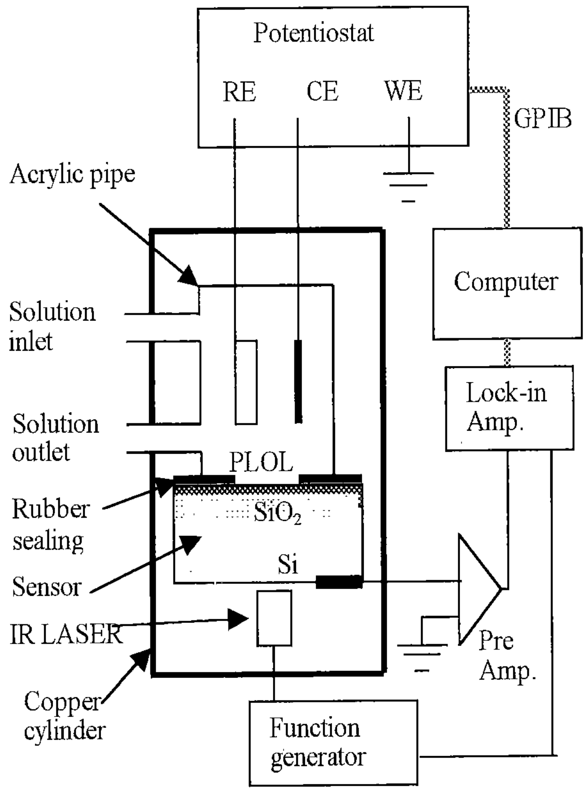

Figure 2. Experimental setup for the LAPS measurement

3. Experimental results and discussion:

The long-term TF is first studied for the novel LAPS response to cell-culture medium that contains 40.0mM NaCl, 1.7mM KCl, 4.1mM $CaCl_2$, 1.5mM $MgCl_2$, 5.0mM glucose, 50unit/ml penicillin, 50unit/ml streptomycin. The pH value of the medium is adjusted to 7.9-8.1 by HEPES buffer solution. The photocurrent response of the sensors is observed for two situation; just after applying the culture medium on the sensor and after keeping few days soaked with the culture medium, imitating the total culture and recovery period of biological cells. The response of novel (PLOL-coated SO_2) and conventional (Si_3N_4 + SiO_2) LAPS are given in figure 3, for the culture medium (pH 7.9). As the theory predicts the amplitude of the photocurrent and the TF of the response characteristics for the SiO_2-based LAPS are much higher than the SiO_2+Si_3N_4-based LAPS.

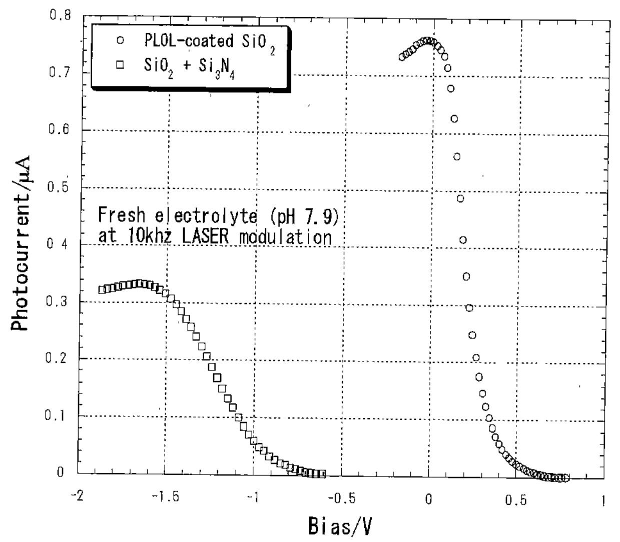

Figure 3. Comparative photocurrent response of the LAPS.

Figure 4 shows the LAPS response of the PLOL-coated SiO_2 sensor measured several times during ten days of observation with culture medium. Here we can see that the amplitude of the photocurrent did not change appreciably during the ten days in contact with the culture medium. This is may be due to the PLOL layer, which prevents leaking of the water and ions into the SiO_2 and thereby keeping the dielectric property intact.

The TF for the LAPS can be defined as the change of photocurrent for the unit change in surface potential, at the linear region of the photocurrent response characteristics. Since in our experimental setup, we convert the current into voltage by a factor of 10^6V/A, we define the TF as the change of output voltage for the unit change of bias potential at the linear region of the LAPS characteristics.

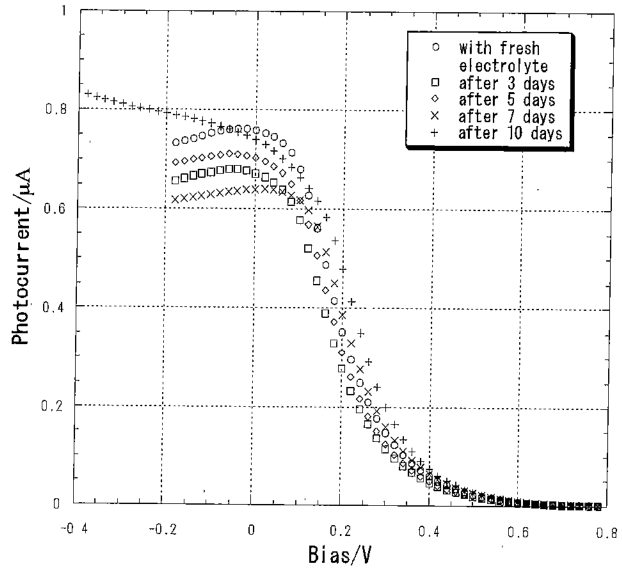

Figure 4. Response of PLOL-modified SiO_2-based LAPS in contact with the electrolyte

The average TF of the novel LAPS response for several times measurement during its ten days period in contact with cell-culture medium is shown in figure 5. The TF decreases until 3rd day, then increases with time, and, finally shows a saturation tendency at the tenth day in contact with the cell-culture medium. This variation with time was not clearly understood, but it may be due to the complex diffusion mechanism of various ions in the cell-culture medium through the PLOL layer. Though the TF varies from 3 to 3.5 in ten days period, still, it is much higher than that of conventional LAPS, the average value of which is 0.65.

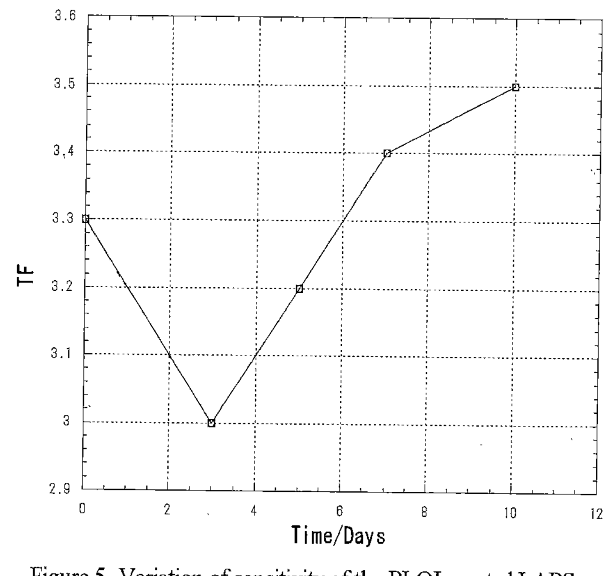

Figure 5. Variation of sensitivity of the PLOL-coated LAPS.

The temporal shift of the novel LAPS output signal is analyzed to study the long-term behavior in contact with the electrolytic solution that shows the electrochemical stability of the sensor.

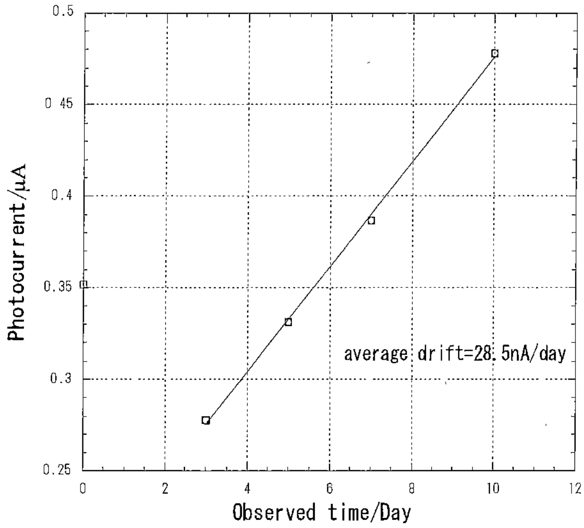

Figure 6. Drift nature of the PLOL-coated SiO_2 -based LAPS

Figure 6 shows the shift of the sensor signal at pH 7.9 over the measurement period of ten days. The high drift of the sensor represents the diffusion of ions from the electrolyte to the oxide layer as well as to the oxide-semiconductor interface. This is also confirmed by the LAPS characteristics at the tenth day (Figure 4) in contact with the electrolyte where it shows some DC leakage.

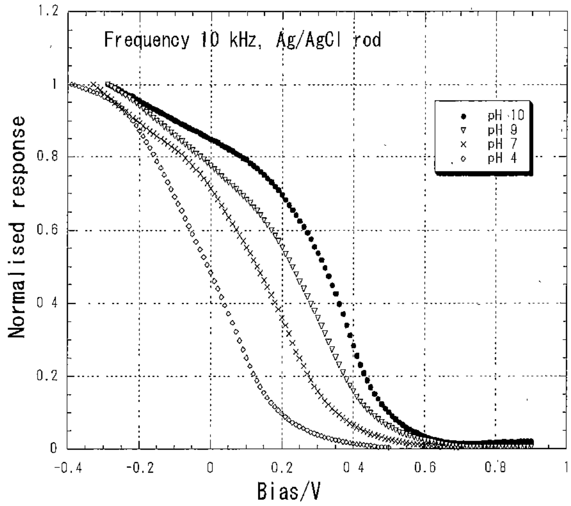

Fig 7. PLOL-coated LAPS response to various pH buffers

The pH response of the sensor is investigated for the pH buffer in the range of pH 4 to pH 10 and their photocurrent response is shown in figure 7. The inflexion points, which are the zero crossing potentials of second derivative of the LAPS response for various buffers, are plotted against their pH value to see the pH sensitivity and are shown in figure 8. The average pH sensitivity is found to be 58.33mV/pH, which is quite near to the Nernstian value of about 59mV/pH, in the experimented pH range.

The total system noise, after converting the photocurrent to voltage by a factor of 10^6 using a pre-amplifier, is found to be $\approx 2mV_{pp}$. Considering an average TF of 3.3, the sensor is sensitive to a series potential change of 600μV with the present noise level. With the average pH sensitivity of 58.33mV/pH for this sensor, 600μV change in surface potential corresponds to a pH change of 10mpH. Therefore, a pH resolution of 10mpH is possible. Decreasing the average noise level and thinning the SiO_2 as well as the bulk silicon can further increase this sensitivity.

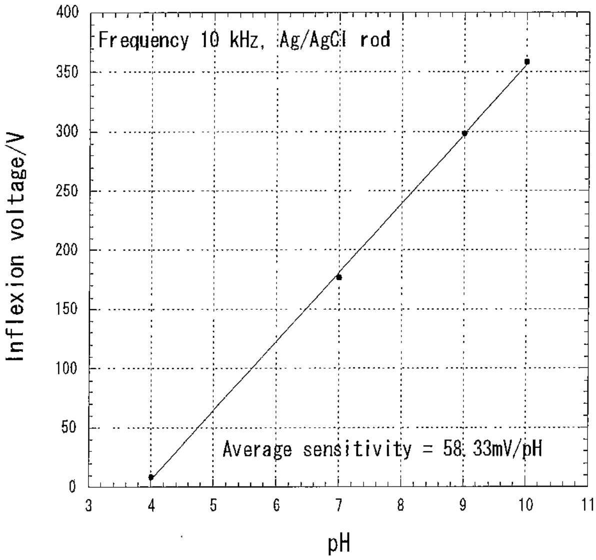

Fig. 8 Calibration curve for the PLOL-coated SiO_2-based LAPS

4. Conclusion:

In this article a novel light-addressable potentiometric sensor structure containing a thin layer of Poly-L-Ornithine with Laminin-coated SiO_2 as the only insulating layer on Si is investigated. Using PLOL-coated SiO_2 we tried to enhance the Transduction Factor of the LAPS response so that higher Surface Potential Sensitivity in the measurement of tiny change of surface potential as well as resolution in the ionic concentration measurement is achieved. Complying with the theoretical consideration, experimentally we also obtained much higher TF over the conventional LAPS. The pH response of the novel LAPS is also high. Despite the high drift, the experimental result shows a very good promise for the PLOL-coated SiO_2/Si structure as a LAPS with the ability to be a novel biological interface.

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Biography:

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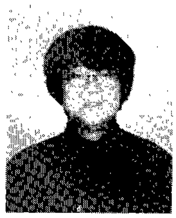

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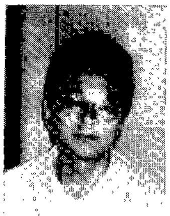

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