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A flexible and wearable glucose sensor based on functional polymers with Soft-MEMS techniques

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Abstract

A novel biosensor for glucose measurement using functional polymers was fabricated and tested. The biosensor utilizes the physical and chemical functions of hydrophobic polydimethyl siloxane (PDMS) and hydrophilic 2-methacryloyloxyethyl phosphorylcholine (MPC) copolymerized with dodecyl methacrylate (DMA). The glucose sensor was constructed by immobilizing glucose oxidase (GOD) onto a flexible hydrogen peroxide electrode (Pt working electrode and Ag/AgCl counter/reference electrode). The electrodes were fabricated using microelectromechanical systems (MEMS) techniques onto those functional polymers. The sensor showed novel functions of flexibility and it was stretchable so that the sensor could normally work when it was released after expanding to 120% longer than that of normal length. Also, basic characteristics of the sensor were evaluated. The output current of the hydrogen peroxide electrode was linearly related to the hydrogen peroxide concentration in a range of 0.20–2.50 mmol/l, with a correlation coefficient of 0.998. GOD was then immobilized onto the surface of the sensor using MPC polymer. In this case, the current output of the glucose sensor related to the glucose level over a range of 0.06–2.00 mmol/l, with a correlation coefficient of 0.997. The calibration range includes the reported concentration of tear glucose in normal human subject (0.14 mmol/l).

Keywords: Biosensor; Phospholipid polymer; MPC; PDMS; Flexible; Glucose sensor

1. Introduction

As diabetes mellitus is rapidly increasing, the development of a safe, convenient and continuous blood sugar level monitoring technology is a pressing need throughout the world. “Finger pricking” is well known method of blood sampling. The sample is applied to a reagent strip for analysis in a portable meter (Pickup, 2003; Pickup et al., 2005). While “finger pricking” is a relatively painless and commonly used method by diabetic patients for measuring their own blood sugar level, there are several issues to be considered. The one is inconvenience and the required compliance by patients and the second is that this is not a continuous and non-invasive monitoring method (Badugu et al., 2003). Although many kinds of blood sugar monitoring method (March et al., 1982; Rabinovitch et al., 1982; Schier et al., 1988; Clarke et al., 1988; Meadows and Schultz, 1988; Trettnak and Wolfbeis, 1989; Heise et al., 1994; Tolosa et al., 1997, 1999; D’Auria et al., 2000) have been reported, critical method has not been carried out. Thus, one of the major trends in research on glucose measurement is continuous in vivo glucose monitoring. Particularly, continuous blood sugar monitoring method which does not become interfere with daily life is attractive.

Usually, continuous glucose monitoring do not measure blood glucose directly, but rely instead on measurement of the glucose levels in other biological fluids (Wilson and Gifford, 2005). With reference to continuous monitoring, relationships between general physical conditions and constituents of biological fluids such as tears, mucus, sweat and saliva were reported (Daum and Hill, 1982; Man et al., 1979; Mitsubayashi et al., 1994; Romano and Rolant, 1988; Stolwijk et al., 1991). For

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example, correlation between glucose concentration in tears and blood glucose was reported (Sen and Sarin, 1980). Tear glucose level changes with a delay of approximately 5 min in comparison with blood sugar level (March et al., 2001). On the other hand, we reported flexible electrochemical sensors for bioinstrumentation on eye site (Mitsubayashi et al., 2003; Iguchi et al., 2005) in previous works.

On the basis of such a technical backgrounds, the sensor for tear glucose measurement was fabricated and tested. The most significant feature of the sensor is that flexible and biocompatible polymers were used for all structural members. In particular, phospholipid polymer, which is so-called MPC polymer, was used for the sensing region. MPC polymer has molecular configuration, which is similar to a cell membrane. Such a configuration was carried out by the techniques of polymer chemistry (Ishihara et al., 1990, 1998). Utilizing this sort of a polymer as a contacting part to measuring site, biocompatible sensor can be achieved. In this work, poly (MPC-co-DMA) (PMD) was used for the sensor. To utilize PMD as a sensor material, a flexible PDMS membrane was used as the substrate on which Pt working electrode and Ag/AgCl reference/counter electrode were formed. Applying film electrodes formed by "Soft-MEMS" techniques, the sensor showed excellent flexibility that enables itself to fit the shape of measuring site. In this paper, details of the structure, fabrication, basic characteristics of the glucose sensor using phospholipid polymer is presented. Also, the result of hydrogen peroxide measurement through PMD membrane is reported.

2. Experimental section

2.1. Evaluation of PMD membrane

To confirm the availability as a sensor material, hydrogen peroxide permeability of a PMD membrane was investigated. A Pt disc electrode (diameter: 0.5 mm) was dipped into a PMD solution (PMD: 10 wt.%, ethanol: 90 wt.%) for 10 s and cured for 60 min at room temperature. The PMD-coated Pt electrode and an Ag/AgCl wire were then placed into a 50 ml measuring cell filled with phosphate buffer solution (PBS: pH 7.0, 50 mmol/l). A two-electrode electrochemical method was employed for investigation. A constant voltage of 550 mV versus Ag/AgCl was applied to the PMD-coated Pt electrode by a computer controlled potentiostat (HAB-151, HOKUTO DENKO Co., Japan). The change of output current induced by adding hydrogen peroxide (final concentration, 0.2 mmol/l) was recorded using PC, which was connected to the potentiostat via 16 bit A/D converter (ADC-16, pico Technology Ltd., UK).

2.2. Construction of a glucose sensor with functional polymers

The structure of the flexible glucose sensor is illustrated in Fig. 1. The sensor was constructed by immobilizing glucose oxidase (GOD: EC 1.1.3.4, Wako Pure Chemical Industries Ltd., Japan) onto a flexible hydrogen peroxide electrode. The hydrogen peroxide electrode has a Pt working electrode and an Ag/AgCl reference/counter electrode formed on a 3 mm × 50 mm × 350 μm hydrophobic polydimethyl siloxane (PDMS) membrane. The gas-permeable PMD membrane was placed on the other surface of the electrodes.

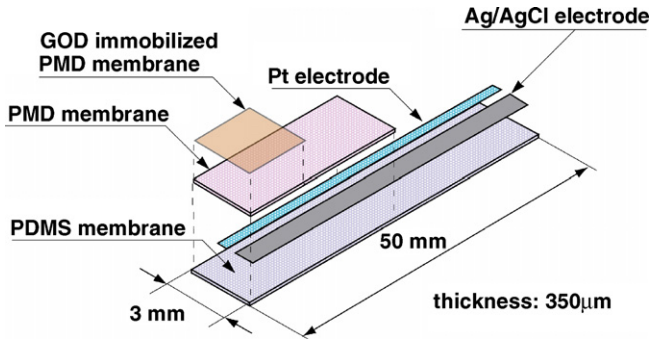

Fig. 1. Schematic structure of the flexible glucose sensor using MPC polymer.

Fig. 2 shows the fabrication process of the glucose sensor. The glucose sensor was fabricated by utilizing MEMS techniques to functional polymer membranes. At first, a 350 μm thick PDMS (Silpot 184, Dow Corning Toray Co., Ltd., Japan) was spin-coated on a dummy silicon wafer and cured at room temperature for 24 h. Then, a positive photoresist (Shipley S1818, Rohm and Haas Electronic Materials Co., USA) was spin-coated. The photoresist was pre-baked under low temperature (60 °C, 30 min). Then the photoresist was patterned into the electrode using a mask aligner (PEM-1000, Union Optical Co., Ltd., Japan). After that, the substrate was placed into a sputtering chamber of an ion beam sputtering (IBS) system (EIS-220, Elionix Co., Ltd., Japan) and 200 nm thick Pt film was sputtered with the acceleration energy of 1900 V. As shown in Fig. 2, the pattern of the Pt electrode was covered using the positive photoresist and 300 nm thick Ag film was sputtered using IBS with the acceleration energy of 1900 V. The Pt and Ag electrode was then formed using lift-off process. Acetone was used to remove the photoresist. The Ag electrode was electrochemically chloridized and finally, PMD was dip-coated. Thus, a flexible hydrogen peroxide electrode was obtained. In order to utilize the functions of PMD membrane with the glucose sensor, GOD was also immobilized using PMD solution casting. A mixture of 100 μl PMD solution (PMD: 10 wt.%, ethanol: 90 wt.%) and 0.5 mg (10 units) GOD was used for casting. Thus, the flexible glucose sensor using phospholipid polymer was obtained.

2.3. Evaluation of the sensor characteristics

At first, response and current dependence of the flexible hydrogen peroxide electrode coated by hydrogen peroxide permeable PMD membrane was investigated. The flexible electrode formed using microfabrication process onto functional polymer membrane was connected to the batch measurement system as mentioned, and constant voltage of 650 mV versus Ag/AgCl reference/counter electrode was applied to the Pt working electrode using the PC-controlled potentiostat, which was used for evaluation of PMD-coated disc electrodes. The change of output currents induced by electrochemical reaction at the electrodes