—Note—

Development of a Micro Glucose Sensor Using an Electrodeposited Platinum Black with Mechanical Stability

Shoichiro IKEDA*, Hirokazu WATANABE and Iseimi IGARASHI†

Received June 16, 1995; Accepted August 22, 1995

1 INTRODUCTION

Diabetes meritus is one of the serious diseases of not only adults but also young generation. The intermittent injections of insulin (1 to 2 times a day) are widely accepted as the therapy of controlling the blood glucose levels (BGLs) to normal values but can not stop the advancement of complications such as diabetic nephropathy and retinopathy. Therefore, artificial pancreas, so called, constructed with an automatic insulin injector and continuous monitoring system (or sensor) of BGL has been researched and developed^{1,2)}. The key point of the development of implantable artificial pancreas is that of a small implantable glucose sensor which can measure BGL directly up to 500 mg/dl.

The authors have previously reported several kinds of O₂ electrode type glucose sensors³⁻⁶⁾. Yamauchi et al.⁷⁾ reported EEE-type glucose sensor based on an electrodeposited platinum (Pt) black electrode embedded glucose oxidase (GOD).

We have proposed an on-line ultrasonic cleaning method to remove the adhered materials on the sensor surface from blood or body fluid such as proteins. However, Pt black deposited by the conventional method easily

separates from the base metal. We have developed a micro glucose sensor fabricated in a needle using a newly developed electro-deposition method of Pt black with physical stability.

2 EXPERIMENTAL

A Pt wire (0.127 mm ϕ) was sealed in the capillary of a 23-gauge needle (OD = 0.65 mm ϕ , ID = 0.45 mm ϕ) with epoxy resin as shown in Fig. 1. The wall of the needle had two small windows (2.00 $\times$ 0.45 mm) to expose the test solution. Pt black was electrodeposited under irradiation of ultrasonic wave from a solution of H₂PtCl₆·6H₂O (33 mg/ml) and Pb(CH₃COO)₂ (0.6 mg/ml) for 80 s at -0.080 V vs. Ag-AgCl⁷⁾ using a potentiostat (Hokuto Denko; HA-151), a coulometer (Hokuto Denko; HF-201), and an ultrasonic cleaner (Shindengen Kogyo; SUC4003A, 40 kHz, 30 W). GOD (Sigma Chemicals; Type VII-S, 165,800 U/g) was adsorbed to the Pt black surface from 0.75

Fig. 1 Schematic drawing of a micro glucose sensor.

GOD: glucose oxidase, GDA: glutaraldehyde, PTFE: Teflon sheath.

Department of Applied Chemistry, Nagoya Institute of Technology (Gokiso-cho, Showa-ku, Nagoya, 466, Japan)

† Toyota Physical and Chemical Research Institute (Aza-Yokomichi, Ooaza-Nagakute, Nagakute-cho, Aichi-gun, 459, Japan)

Key Words: Glucose Sensor, Platinum Black, Ultrasonic, Immobilized Enzyme

ml of 5300 U/ml solution (phosphate buffered (PB), pH 7.45) at 6 °C for 30 min and air-dried. Then the sensor was dipped into 1% glutaraldehyde solution (PB) at 6 °C for 10 min and dried, and then dipped into 0.5% Nafion solution (Aldrich Chemicals Co., Inc.; 5% solution: diluted with a 1:1 wt ratio solution of water and 2-propanol) at 6 °C for 10 min, and then dried in order to immobilize GOD. The latter process was repeated 3 times. The activity of immobilized GOD on the sensor was measured by 3-Methyl-2-benzothiazolinone-hydrazone (MBTH) – Diethylaniline (DEA) method⁸⁾ at 37 °C.

The dynamic response of the glucose sensor at 37 °C was measured by the stepwise increase in the glucose concentrations in phosphate buffered saline solution (PBSS, pH 7.45) using an injector (Kyoto Electronics; APB-118), an I-V convertor using operational amplifiers⁴⁾, and a data acquisition system composed of an interface (KSK Co., Ltd.; LH-161A) and a personal computer (NEC; PC-9801 ns/T) under different oxygen partial pressures, which were prepared from air or pure O₂ gas diluted with N₂ using mass flow controllers (Brooks Instrument; 5850E).

3 RESULTS AND DISCUSSION

Figure 2 shows the microscopic cross sectional view of Pt black deposited on a Pt wire by the present method after sonication for 1 hr at 38 kHz and 30 W in water. Pt black tightly adhered on the surface of a Pt wire, while that deposited by the conventional method, i.e. without the irradiation of ultra sonic wave, was easily removed from the surface by sonication. Thus, this method is promising to make the firm Pt black layer which resists the ultra sonic cleaning for adhered coagulation or deposited proteins on the surface of Pt black electrode to restore the performance of the sensor.

The activity of GOD immobilized on the Pt black was found to be 5.5 mU/sensor on the first day. It gradually decreased with the passage of time but kept more than 74% of the initial value after 26 days storing in PBSS at 6 °C, except for each period of checking the activity at 37 °C for 15 min, as shown in Fig.

Fig. 2 Scanning electron microscopic sectional view of the Pt black layer on the Pt wire after sonication for 1 hour by a ultrasonic cleaner.

Fig. 3 Time dependence of the enzyme activity of immobilized GOD on the Pt black of a micro glucose sensor stored at 6 °C.

3. Although the effect of the sonication on the activity of the immobilized enzyme has not been examined, it will be required before the practical use of the sensor.

Figure 4 shows the typical dynamic response curves to the stepwise changes of glucose concentration in PBSS under different oxygen partial pressures. The sensor responds to glucose concentration up to about 50 mg/dl within a few minutes and its output current varies with

Fig. 4 Dynamic response to stepwise changes of the glucose concentration in a phosphate buffered saline solution of a micro glucose sensor under different oxygen partial pressures.

oxygen partial pressures. As the stoichiometric concentration of the glucose to that of the dissolved oxygen in the human body is about 40 mg/dl and the sensor has not any membrane to prevent the penetration of glucose⁴⁾, its output current saturates over 50 mg/dl. To use the sensor for the implantable artificial pancreas,

it requires to be attached the semipermeable membrane⁴⁾, because the maximum BGL of the diabetic patients occasionally becomes more than 500 mg/dl.

This work was partially supported by Grant-in-Aid for Developmental Scientific Research No. 05555116 from the Ministry of Education, Science and Culture, Japan.

REFERENCES

- 1) S.P. Bessman and R.D. Shultz, *Holm. Metab. Res.*, **4**, 413 (1972).
- 2) G.F. Cahill, Jr., J.S. Soeldner, G.W. Harris, and R.O. Foster, *Diabetes*, **21**, 703 (1972).
- 3) S. Ikeda, K. Ito, K. Ito, and T. Kondo, *Denki Kagaku*, **46**, 667 (1978).
- 4) S. Ikeda, N. Aoyama, K. Ito, K. Ookura, H. Ichihashi, and T. Kondo, *J. Chem. Soc. Jpn.*, 1554 (1980).
- 5) S. Ikeda, H. Naruse, K. Asai, M. Shimada, K. Ito, K. Ohkura, A. Nakao, H. Ichihashi, and T. Kondo, *Jpn. J. Artif. Organs*, **14**, 297 (1985).
- 6) S. Ikeda, M. Kimura, H. Noda, K. Ito, K. Ohkura, A. Nakao, H. Takagi, and T. Kondo, *Denki Kagaku*, **56**, 1055 (1988).
- 7) N. Yamauchi, Y. Ikariyama and M. Yaoita, "Chemical Sensor Technology Vol. 2", Kodansha Ltd., 205 (1989).
- 8) Toyobo Enzymes Catalogue, Glucose Oxidase, Toyobo Co., Ltd. (1981).