Electrochemical characteristics of enzyme/graphene electrodes

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Graphene is attractive as electrode materials for biofuel cells because of its high electrical conductivity and chemical stability. The enzyme electrode with high current density and excellent reproducibility can be fabricated by utilizing two-dimensional structure of graphene with a large specific surface area and high uniformity. Stable immobilization of the enzyme is important in applications of enzyme-based biofuel cells. To our knowledge, there are few reports on evaluation of the electrochemical characteristics of enzyme-immobilized graphene planer sheets prepared by chemical vapor deposition (CVD). We focus on CVD-grown graphene sheets as electrode materials for biofuel cells because they are highly uniform, and the shape and size can be accurately controlled by conventional photolithography processes. In this study, we immobilized enzyme on CVD-grown graphene sheets by physical adsorption and evaluated electrochemical characteristics of the enzyme/graphene electrodes.

Graphene sheets were grown by CVD on Cu foil. To evaluate the intrinsic electrochemical characteristics, the graphene sheets were transferred onto electrochemically inert SiO2/Si substrates by a using polymethyl methacrylate (PMMA) method. The transferred graphene sheets were confirmed to be monolayer by Raman spectroscopy. We immobilized an enzyme, glucose oxidase (GOx), in two ways. One was the easiest procedure, deposition adsorption, in which the graphene sheets were soaked for 12 h in GOx solution [1]. The GOx/graphene electrodes were covered by cellulose membranes to prevent desorption of GOx. The other procedure used noncovalent functionalization of the graphene surface. 1-pyrenebutanoicacid succinimidyl ester (PBSE) was used as a chemical linker on graphene [2]. The pyrenyl group of PBSE interacts strongly with the graphene surface by π-stacking and the succinimidyl ester parts combine with the amino base of GOx. The graphene sheets were soaked in 1mM PBSE in pure methanol for 3h, followed by soaking in GOx solution for 12 h. Electrochemical characteristics of these electrodes were measured by cyclic voltammetry (CV) at a scan rate of 10 mV/s in phosphate buffered saline (PBS:pH 7.0) containing glucose of 25 mM.

Figure 1 shows a frequency modulation atomic force microscopy (FM-AFM) image of GOx/PBSE/graphene electrode in ultrapure water. It is evident that GOx is immobilized on the graphene surface because three dimensional structures were observed on initially flat graphene surface. Fig. 1 shows the cyclic voltammograms recorded using graphene (dotted line), GOx/graphene (dashed line), and GOx/PBSE/graphene (straight line) electrodes. GOx catalyzes the oxidation reaction of glucose and current due to the oxidation reaction is generated by the addition of glucose. As shown in Fig. 1, the current responses were clearly observed for two enzyme-immobilized electrodes, in which the current density of the GOx/PBSE/graphene electrode had twice as high as that of the GOx/graphene electrode. The results suggested that the immobilization of the enzyme using PBSE formed strong connectivity between graphene and the enzyme, leading to a higher current density.

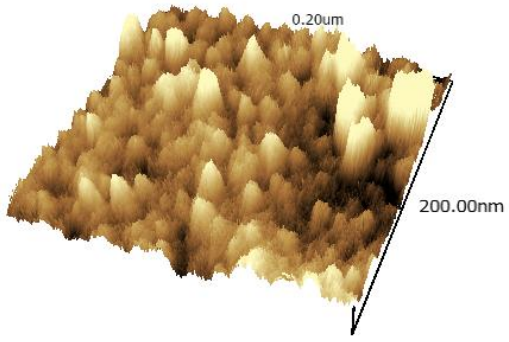

Fig.1 : FM-AFM image of GOx/PBSE/graphene

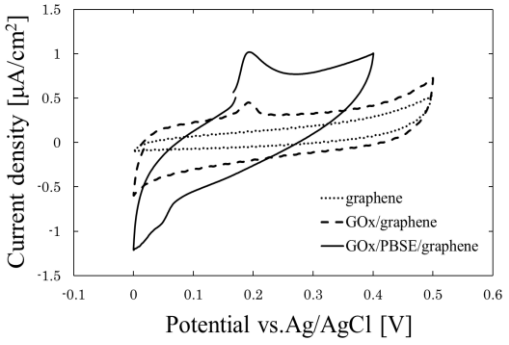

Fig.2: cyclic voltammograms of each electrode

[1] A. Zebra et al., Nature Communications. **2,** 370 (2006).
[2] R. J. Chen et al., J. Am. Chem. Soc. **123,** 3838 (2001).

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