CrossMark

Fluoroaromatic substituents attached to carbon nanotubes help to increase oxygen concentration on biocathode in biosensors and biofuel cells

Maciej Karaskiewicz [a], Jan F. Biernat [b], Jerzy Rogalski [c],
Kenneth P. Roberts [d], Renata Bilewicz [a,*]

[a] *Faculty of Chemistry, University of Warsaw, Pasteura 1, 02-093 Warsaw, Poland*
[b] *Department of Chemical Technology, Gdansk University of Technology, Narutowicza 11/12, 80-233 Gdańsk, Poland*
[c] *Department of Biochemistry, Maria Curie Sklodowska University, Akademicka 19, 20-031 Lublin, Poland*
[d] *Department of Chemistry and Biochemistry, The University of Tulsa, 800 S. Tucker Drive, Tulsa, OK 74104, USA*

ARTICLE INFO

ABSTRACT

Based on the known ability of perfluorodecalin/perfluorohydrocarbons to enhance oxygen solubility we modified oxygen sensitive biocathode by adding perfluorinated components to the catholite. This procedure improved the efficiency of the oxygen sensitive cathodes. Glassy carbon electrodes covered with single-wall carbon nanotubes (SWCNTs) with covalently bonded perfluoroaromatic groups were shown to be more sensitive to oxygen, and higher catalytic reduction currents were observed using laccase modified biocathodes allowing to improve the performance of bioelectrodes for fuel cells and oxygen monitoring devices. Maximum current and power density was found for the glucose/oxygen fuel cell composed of anode modified with side-aminoethylated SWCNTs conjugated with glucose dehydrogenase and cathode covered with side-perfluorophenylated SWCNTs and laccase. The open circuit potential of this cell is 0.57 ± 0.05 V and the maximum power density is 0.865 ± 0.090 mW/cm^2. In case of applications of biofuel cells in the biological media, in order to avoid addition of NAD$^+$ to the solution, we propose binding this cofactor to the aminoethylated SWCNTs. For applications of this biofuel cell in biological media, we propose binding NAD$^+$ cofactor to the aminoethylated SWCNTs instead of its addition to the electrolyte. The power output 0.226 ± 0.031 mW/cm^2 of such cell is somewhat smaller but still satisfactory, and the open circuit potential is 0.65 ± 0.08 V.

1. Introduction

In recent years there has been substantial progress in designing enzyme covered electrodes for enzymatic biofuel cells and biobatteries. These devices that generate energy from readily available fuels, e.g. sugars or alcohols, attract much attention since they are cheap, work at room temperature and at pH close to physiological, and use enzymes catalyzing oxygen reduction and glucose oxidation. Catalyzed oxygen reduction could proceed directly to water without any harmful intermediates and side products [1–11]. The enzymes are also specific, so no additional membranes separating the cathodic and anodic compartments are needed when the biocathode is used in the enzymatic biofuel cell [4,10–12]. Recently, several promising glucose–oxygen biofuel cells and biobatteries

have been reported; increased stability and significantly improved power output have been demonstrated using carbon fiber, carbon nanotubes, carbon black and other nanostructured carbonaceous electrode matrices [13–22]. The effect of oxygen concentration on the biocathode performance was studied both numerically and experimentally [23,24]. As oxygen solubility in aqueous solutions is rather low, improvement of oxygen transport into the enzyme matrix and maintaining its stable concentration at the electrode surface should remove one of the constraints in the application of oxygen based electrodes [25]. During work of the biocathode in the fuel cell, rapid decrease of oxygen concentration in the vicinity of the electrode surface is observed. Thus low concentration of oxygen and its fast depletion at the biocathode are problems to be tackled in order to achieve the stage of practical applications. To solve these issues flow systems, gas-diffusion cathodes and pressed-air-breathing biocathodes have been proposed [26–28].

In the present study, we made use of the ability of perfluorocarbons, to accumulate large quantity of oxygen. This behavior we studied using as an example perfluorodecalin film placed on

---

the electrode surface. To improve oxygen accumulation on the electrode surface we also designed and synthesized fluoroarylated carbon nanotubes. In our recent papers [29–36] we demonstrated that arylated carbon nanotubes provide efficient pathways for the transfer of electrons between the active center of laccase and the conducting support. The 3-dimensional nanotubes network of modified carbon nanotubes on the electrode is able to accommodate and accumulate huge amount of catalyst [37–40]. The phenyl, naphthyl or terphenyl groups bonded to the nanotubes allow stable and durable adsorption of the enzyme and increase the amount of enzyme molecules in suitable orientation to easy exchange electrons with the electrode.

Formerly we presented the synthesis, purification and characterization of arylated SWCNTs [35]. In the present paper we describe the synthesis of SWCNTs equipped with perfluoroaryl residues. These materials retain the laccase binding feature to the arylated carbon nanotubes, and at the same time increase the amount of oxygen concentration close to the electrode surface as it was observed for perfluorodecalin.

At the anode of the biofuel cell, glucose dehydrogenase (GDH) enzyme is used since it does not react directly with oxygen as opposed to glucose oxidase. In the latter case, the performance of anode is decreased because oxygen from the cathode accesses the anodic part and competes with the enzyme; moreover, the H_2O_2 produced in this case destabilizes the enzyme [41,42]. Nicotinamide adenine dinucleotide dependent glucose dehydrogenase (GDH) was chosen; however binding of its NAD^+ cofactor is relatively weak, and therefore it should be either added to the solution, the matrix on the electrode, or immobilized directly together with the enzyme onto the electrode surface [43,44]. Recently, we have shown the immobilization of NAD^+ jointly with the enzyme in the liquid-crystalline lipid matrix and the application of diaphorase to decrease the overpotential of NADH oxidation [37]. Here we propose binding of NAD^+ onto the carbon nanotubes together with the enzyme as the best way to nanostructurize the anode surface in the biological fuel cell.

2. Experimental

2.1. Materials and chemicals

All reagents were of analytical grade; solutions were prepared using MilliQ water. SWCNTs were from CheapTubes, Brattleboro, USA (purity >90%); or from Nanocyl, Sambreville, Belgium. Perfluorodecalin, perfluoroaniline and perfluorobenzoic acid were from Sigma–Aldrich, while aniline, benzoyl chloride and analytical grade solvents were from POCh Gliwice, Poland.

Thermogravimetric analyses (TGA/DTA) were conducted using Universal V4.3A TA instrument in argon atmosphere at a heating rate of 10°/min. The derivatization degrees were calculated according to recommendations [43–45], assuming temperature of 600 °C as sufficiently high for complete detachment of substituents. The calculated results are collected in Table 1.

FT-IR spectra were registered using Genesis II (Mattson) instrument. Materials were prepared in the form of KBr pellets. The characteristic bands for the fluorinated species are given in the synthesis section and include C–F frequencies. The spectra for benzoyl derivatives and anilides do not show expected specific bands of noticeable intensity.

Raman spectra were collected using a Witec confocal Raman microscope system (Ulm, Germany) equipped with a fiber-coupled Melles Griot (Carlsbad, CA) argon ion laser operating at 514.5 nm focused through a 60× objective. Collected light was dispersed through a triple monochromator (600 g/mm, 500 nm blaze) and detected with a thermoelectrically cooled (−60 °C) charge-coupled

device. In the sample preparation procedure a small amount of carbon nanotubes in powder form was placed between a microscope slide and a cover slip.

2.2. SWCNTs purification and modification procedures

2.2.1. Purification of SWNTs
To remove metallic and carbonaceous impurities, the pristine single walled carbon nanotubes purchased from CheapTubes and Nanocyl, were treated with 4 N HCl. The suspension was maintained at 60 °C for 6 h under permanent sonication [46,47] the solid was collected, exhaustively washed with water and vacuum dried.

2.2.2. Syntheses of amides
The synthetic pathways are shown in Scheme 1.

2.2.3. Side 2-aminoethylated-SWCNTs, material **1**
The nanotubes modified with aminoethyl groups on the sidewalls were synthesized according to procedure described by Mugadza et al. [48]. Purified SWCNT (600 mg), $NaNO_2$ (800 mg; 12 mmol) and ethylenediamine (1.2 g, 1.35 ml; 12 mmol) were combined and thoroughly triturated. To this, almost solid material in closed test-tube conc. sulfuric acid (0.52 ml; 10 mmol) was added, mixed with a glass rod and maintained at 60 °C for 1 h. The mixture was diluted with water, the solid filtered off and washed with water to neutrality. Finally, the product was washed with methanol and dried in vacuum.

2.2.4. Benzoylated side aminoethyl-SWCNTs, material **2**
A mixture of 100 mg commercial benzoyl chloride added to a suspension of 50 mg of aminoethyl-SWCNTs **1** and 1 ml pyridine was sonicated for 2 h at 50 °C and then left overnight. The solid was filtered off, washed exhaustively with methanol, next with methylene chloride and vacuum dried.

2.2.5. Pentafluorobenzoylated side aminoethyl-SWCNTs, material **3**
A mixture of 50 mg of pentafluorobenzoic acid, 1 ml thionyl chloride and 0.01 ml dry pyridine was maintained for 2 h at 50 °C and left for 2 days at room temperature. Then 2 ml toluene was added and the volatiles were exhaustively removed under reduced pressure. The residue was dissolved in 1 ml dry pyridine, 50 mg of aminoethylated SWCNTs was added and the mixture was sonicated for 4 h at 50 °C and then left for 2 days at room temperature. The solid was filtered off, washed several times with methanol, finally with methylene chloride and dried under reduced pressure. IR: 3422, 2921, 2851, 2371, 2345, 2323, 1654, 1636, 1559, 1542, 1522, 1457 cm^{-1}.

2.2.6. SWCNTs with terminal carboxylic groups, material **4**
SWCNTs oxidized by means of a mixture of conc. nitric and sulfuric acid produce carboxylic groups at the ends/defect sites of the nanotubes. Thus 260 mg of purified SWCNTs were treated with 50 ml of 3:1 (v/v) sulfuric acid and nitric acid mixture and maintained at 50 °C for 4 h under permanent sonication. The mixture was diluted with water on cooling and left overnight. Then the suspension was centrifuged, the sediment mixed with water, sonicated and centrifuged again. The last procedure was repeated five times. Finally the sediment was mixed with methanol, sonicated, centrifuged and dried under reduced pressure; yield 200 mg, c.f. [49].

2.2.7. Terminal chlorocarbonyl carbon nanotubes, SWCNTs-COCl, material **5**
The dried oxidized SWCNTs **4** (200 mg) were treated with an excess of thionyl chloride (4 ml) and the mixture was sonicated