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An electrochemical glucose biosensor exploiting a polyaniline grafted multiwalled carbon nanotube/perfluorosulfonate ionomer–silica nanocomposite

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ABSTRACT

A glucose biosensor was fabricated with loading of glucose oxidase (GOx) into a new organic–inorganic hybrid nanocomposite. The preparation involves formation of silica network into a Nafion (perfluorosulfonate ionomer) and subsequent loading of polyaniline grafted multiwalled carbon nanotubes (MWNT-g-PANI) onto Nafion–silica nanocomposite. Field emission scanning electron microscopy (FE-SEM) of Nafion–silica/MWNT-g-PANI composite reveals the presence of spherical silica particles (sizes in the range 250 nm–1 μm) and tubular MWNT-g-PANI particles. Chronoamperometry and cyclic voltammetry were used to evaluate the performance of biosensor towards glucose. The Nafion–silica/MWCNT-g-PANI/GOx biosensor exhibited a linear response to glucose in the concentration range of 1–10 mM with a correlation coefficient of 0.9972, good sensitivity (5.01 μA/mM), a low response time (~6 s), repeatability (R.S.D value of 2.2%) and along-term stability. The presence of silica network within Nafion and MWNT-g-PANI synergistically contributes to the performance of the biosensor towards the electrochemical detection of glucose.

1. Introduction

Enzymatic biosensors are attractive as they have many potential applications in various fields that include medical diagnosis, pharmaceutical and environmental controls. Electrochemical sensors hold much promise among the enzymatic biosensors. Sensitivity and stability are the key features for using the biosensors in practical analysis and commercial developments. Electrode modifying materials [1] and techniques for enzyme immobilization [2] have been developed to achieve these key features. A wide variety of electrodes modifying materials such as carbon nanotubes (CNTs) [3,4], polymers [5] and silica [6–8] have been used for the fabrication of biosensors.

For the electrochemical biosensor applications, the electrode modifying material is expected to possess several characteristics such as good electron transduction capability, physical or chemical environment for the stable immobilization of enzyme, bioactivity, easy accessibility towards the analyte and large surface area. Literature reveals that all these important characteristics cannot be inbuilt in a single material. Hence, there is always a demand for the development of composite materials, comprising two or more components, to achieve adequate sensitivity and stability for the biosensors [1,9,10].

Silica-based organic–inorganic composites are attractive materials because they combine both the properties of a rigid three-dimensional porous silica and the organic component [11]. The high specific surface area, mechanical stability, chemical inertness and excellent biocompatibility are the chief characteristics of silica towards fabrication of biosensors [12]. Silica-based materials provide suitable environment for biomolecule entrapment with enhanced electrochemical stability [13]. Besides, the porous and open frame works of silica in sol–gel material validate that the analyte into the active functionalities, which resulted in high sensitivity for the detection of analyte [14]. A simple approach for an efficient entrapment of enzyme into silica has been demonstrated [15]. Thionine doped silica composite was used as a mediator to construct biosensor with the immobilization of horse radish peroxiodase (HRP) [16]. An amperometric glucose sensor was fabricated by in-situ incorporation of glucose oxidase (GOx) within sol–gel silica film on a Prussian blue modified electrode [17]. Cholesterol oxidase was immobilized into chitosan–silica-multiwalled carbon nanotube (MWNT) composite [18]. HRP was

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immobilized onto a dendric Ag particles/silica nanocomposite to fabricate the hydrogen peroxide biosensor [19]. Silica gel/polyvinyl alcohol composite-based biosensor has been fabricated by immobilizing GOx [20]. Thus, literature reveals that sol–gel-derived silica can be combined with another component to prepare functional composites in the process of achieving combinational properties.

Nafion, a perfluorosulfonate ionomer, has been entrapped into sol–gel silica by sol–gel reactions to develop a new class of solid acid catalyst [21,22] and used for the fabrication of electrochemical biosensors [6–8,23,24]. The incorporation of Nafion into the sol–gel-derived silica film minimizes the brittleness of the pure sol–gel-derived silica and enhances the long-term stability of phenol biosensor [25]. GOx was immobilized into Nafion-ordered mesoporous silica composite for the fabrication of glucose biosensor [26]. However, reports on the use of Nafion–silica composites with the incorporation of carbon nanotubes (CNT) or a conducting polymer (CP) towards fabrication of electrochemical biosensor are scarce.

CNT have been extensively used in electrochemical and biosensing studies due to their advantageous properties [27–31]. Recently, CNT-based modified electrodes have been used for the sensitive detection of various analytes [3,4,9,32–34]. Electrochemical studies have clearly demonstrated that CNT-based modified electrodes could exhibit high sensitivity and selectivity for electrochemical detection of analytes [35,36]. The incorporation of CNT into CPs produce new type of composite materials with individual properties of each component as well their synergistic effect on the performance of biosensors [37]. Among CPs, polyaniline (PANI) has been extensively studied due to its high electrical conductivity and redox suitability. To date, several studies have been reported on the use of CNT/PANI or derivatives of PANI composites for the fabrication of biosensors [36,38,39]. The electrocatalytic role of PANI grafted CNT for the electron transfer reaction of biomolecules [32–34] has been well documented.

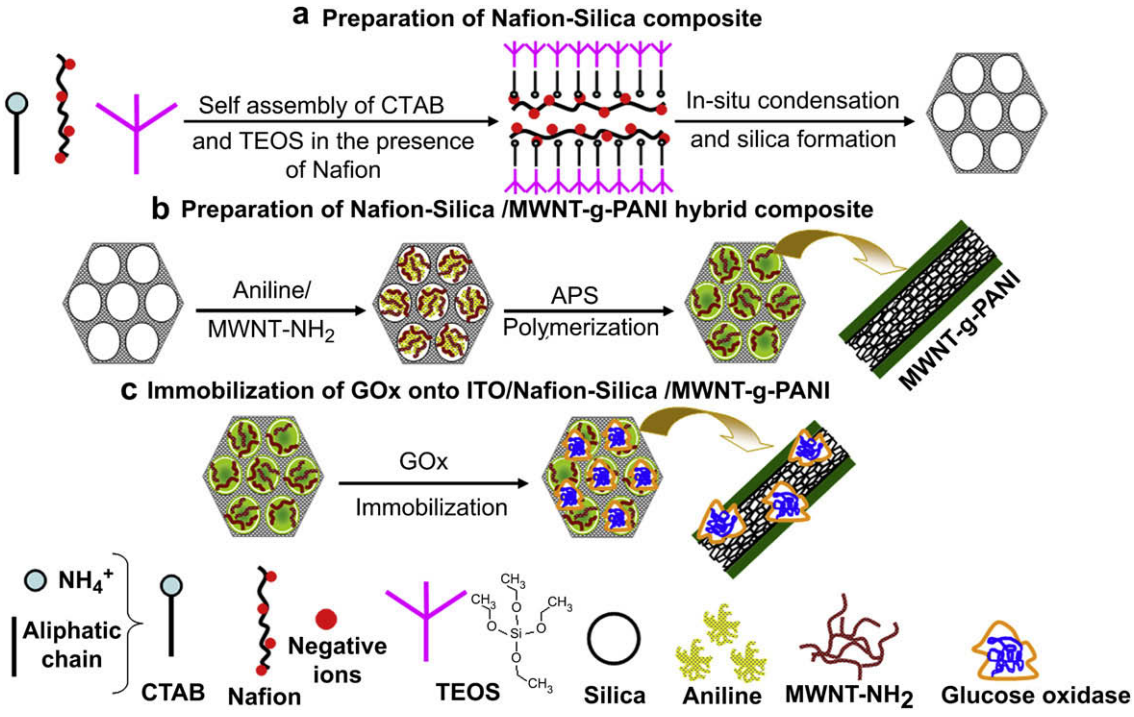
In the past two decades, glucose biosensors received great attention due to the importance in the detection of glucose for the growing population of diabetic patients [40,41]. The ultimate goal of the glucose electrochemical biosensors lies on designing high performance sensor with appropriate characteristics such as sensitivity, selectivity, response time, good linear concentration range, stability and reproducibility. Several approaches have been developed in the process of performance improvements which include development of efficient matrix for enzyme immobilization and preparation of electrocatalytic components [42]. Nafion, silica, MWNT, and conducting polymer have individually been known to improve the electrocatalytic activity [43], electron conduction path [44], sensitivity [45], stability of biosensors [46], respectively. Sol–gel silica, CNT, PANI and Nafion were used either alone or in binary combinations for the fabrication of glucose biosensor, silica gel/GOx [45], Nafion/GOx/CNTs [46], Silica sol–gel/GOx/CNTs [47], Sol–gel/GOx/copolymer [48]. To the best of our knowledge, a multi-component composite comprising of CNT, silica, Nafion and PANI has not been utilized for the fabrication of glucose biosensor.

Considering the advantageous properties of silica–Nafion composites and CNTs–PANI composites, we have developed an electrochemical biosensor-based on a multi-component composite comprising of MWNT, silica, Nafion and PANI. We have judiciously integrated the four components in the composite by employing the following strategy (Scheme 1). First, Nafion–silica composite was prepared by the condensation of silica precursors in a Nafion solution. In the subsequent step, MWNT-g-PANI was incorporated into silica–Nafion composite. The glucose biosensor was fabricated by the immobilization of GOx into Nafion–silica/MWNT-g-PANI composite. The glucose biosensor, Nafion–silica/MWNT-g-PANI/GOx exhibited electrochemical response to glucose over a wide concentration range with a good selectivity and sensitivity.

2. Experimental

2.1. Materials

MWNTs were obtained from CNT Co. Ltd. Incheon, Korea. GOx (256 U mg^{-1}; U: enzyme units) and D(+)-glucose were purchased from Sigma (South Korea). Nafion (5%) solution, aniline, ascorbic acid (AA), uric acid (UA), acetaminophen (AP),



Scheme 1. Fabrication of Nafion–silica/MWNT-g-PANI/GOx biosensor.