Sensors and Actuators B 237 (2016) 150–158

Contents lists available at ScienceDirect

Sensors and Actuators B: Chemical

journal homepage: www.elsevier.com/locate/snb

Optical property and adsorption isotherm models of glucose sensitive membrane based on prism SPR sensor

Xi Yang[a], Yinquan Yuan[a,b,*], Zichun Dai[c], Fang Liu[a], Jun Huang[a]

[a] National Engineering Laboratory for Fiber Optic Sensing Technology, Wuhan University of Technology, Wuhan 430070, China
[b] Key Laboratory of Fiber Optic Sensing Technology and Information Processing, Ministry of Education, Wuhan University of Technology, Wuhan 430070, China
[c] School of Life Sciences, Central China Normal University, Wuhan 430079, China

ARTICLE INFO

Article history:
Received 31 December 2015
Received in revised form 15 June 2016
Accepted 15 June 2016
Available online 16 June 2016

Keywords:
Surface plasmon resonance sensor
Glucose sensitive membrane
Immobilized enzyme
Optical property
Adsorption isotherm model

ABSTRACT

For the prism based SPR glucose sensors, glucose sensitive membrane (GSM) is a key factor that decides the performance of sensor. GSM consists of glucose oxidase (GOD) and matrix material (for example, polyacrylamide gel, PAM). As the prism based SPR glucose sensor coated with GSM is put in the glucose solution, some glucose and dissolved oxygen can diffuse into the GSM and produce by-products, then the by-products will change the refractive index (RI) of GSM and cause the spectrum shift of SPR sensor. In this paper, our proposes are to build the quantitative relationships among the resonance angle of SPR sensor, the RI of GSM, the concentration of glucose in GSM and the concentration of glucose in solution, and then to provide an efficient research method for the GSM of optical sensors. Firstly, we have reported the fabrication and characterization of GSM which is made of immobilized GOD on SiO_2 nanoparticles and polyacrylamide gel. Secondly, we have finished a serial of SPR sensing experiments and obtained the resonance spectra of SPR sensors coated with pure PAM gel film, with free GOD@PAMfilm, and with immobilized GOD@PAM film in PBS solution, respectively, also displayed their variation relationship of resonance angle with the concentration of glucose in PBS solution. Thirdly, we have investigated the effects of solution pH and the content of immobilized GOD on the performance of the sensor, and obtained the optimum solution pH and the optimum content of GOD. Finally, we have related the resonance angle of SPR sensor and the RI of GSM by the combination of SPR experiment data and theoretical simulation, related the RI of GSM and the concentration of glucose in GSM according to Gladstone-Dale mixing rule, related the concentration of glucose in GSM and the concentration of glucose in solution by using the adsorption isotherm model of GSM.

1. Introduction

The measurements for the concentration of glucose in solution are of importance in biochemical research [1] and diagnosis of diabetes [2,3]. In recent years, many methods are used to detect the concentration of glucose in aqueous solution, therein the major methods are electrochemical sensors [4–6] and optical sensors [7–11]. The electrochemical sensors, using electrical signal sensing, have some disadvantages, such as susceptible to electromagnetic interference and vulnerable to pollution. Among the optical glucose sensors, the commonly used methods are by measuring fluorescence signals [7–9], chemiluminescent signals [10], and surface plasmon resonance signals [11–13]. Surface plasmon resonance (SPR) refers to the optical excitation of surface plasmon wave at the interface between a noble metal (gold or silver) and a dielectric. SPR sensors can be divided into two kinds: prism based SPR sensors [14] and optical fiber SPR sensors [15]. The most commonly used SPR sensors are the prism based SPR sensors in Kretschmann configuration, which comprise of a glass prism and a noble metal film, working in wavelength interrogation mode [11,12] or in angular interrogation mode [14,16].

Utilizing of enzymes is an increasing trend to develop new glucose sensors. Especially, immobilized glucose oxidase (GOD) has drawn significant attention for glucose monitoring [17]. Enzyme immobilized on a support has been realized for its stability, reusability and high activity [18]. The technique of GOD immobilization and the support property are key factors to develop an excellent glucose sensor. The common methods for GOD immobilization are adsorption [19], encapsulation [20] and chemical

* Corresponding author at: National Engineering Laboratory for Fiber Optic Sensing Technology, Wuhan University of Technology, Wuhan 430070, China.
E-mail address: ymyyq@whut.edu.cn (Y. Yuan).

http://dx.doi.org/10.1016/j.snb.2016.06.090
0925-4005/

crosslinking [21]. Although physical adsorption and direct encapsulation can achieve high activity, they work only for small size supports. Crosslinking has better stability [21], therefore we use the chemical crosslinking to immobilize the GOD on the surface of SiO_2 nanoparticles. In most cases, gel was used as porous host matrix, in which enzymes are entrapped and smaller molecules can diffuse. Both inorganic and organic materials, such as chitosan [10], silica gels [17,20] and hydrogels [22,23], are often used as host matrix for enzyme immobilization. The polyacrylamide (PAM) gel can be fabricated to meet the desired requirements by using a simple procedure and controlling the ratios of ingredients [24], moreover, the thickness of gel film and the pore size in gel could be adjusted by changing the process parameters and the ratios of monomers and cross-linking agent. The PAM gel is stable, nontoxic and biocompatible, these advantages allow it to be a kind of excellent host matrix.

For the prism based SPR glucose sensors, glucose sensitive membrane (GSM) is a key factor that decides the performance of sensor. In Section 2, we have reported the fabrication and characterization of GSM. To improve the performance of GSM, GOD is immobilized on SiO_2 nanoparticles and then entrapped in gel. The merit of immobilized GOD is to prevent enzyme leaking from the gels while allowing free movements of glucoses and its by-products. The glucose and dissolved oxygen can diffuse into the gel layer and produce by-products, then the by-products will change the RI of gel layer and cause the spectrum shift of SPR sensor.

For the GSM, two important issues need to be answered: (i) the relationship between the RI of GSM and the concentration of glucose molecule in GSM; (ii) the relationship between the concentrations of glucose molecules in the GSM and in the solution at chemical equilibrium. In Section 3, we have outlined the monitoring principle of prism based SPR sensor, the RI formula of composite GSM and the isotherm adsorption model of GSM.

In Section 4, based on a serial of the SPR sensing experiments, we have given the resonance spectra of SPR sensors coated with pure gel film, with free GOD@PAM film, with immobilized GOD@PAM film in PBS solution, respectively, also displayed the variation relationship of resonance angle with the concentration of glucose in two cases of free GOD@PAM film and immobilized GOD@PAM film in PBS solution. Moreover, we also have investigated the effect of PBS solution pH and the quantity of immobilized GOD on the performance of the sensor. Finally, to build the relationships among the resonance angles of SPR sensors, the RI of GSM, the concentration of glucose in GSM and the concentration of glucose in solution, we have used SPR experiment data and theoretical simulation result to relate the resonance angle of SPR sensor and the RI of GSM, used Gladstone-Dale mixing rule to relate the RI of GSM and the concentration of glucose in GSM, used the adsorption isotherm model of GSM to relate the concentration of glucose in GSM and the concentration of glucose in solution.

2. Materials

2.1. Reagents

N-tetramethylethylenediamine (TEMED), ammonium persulphate (APS), acrylamide and bisacrylamide were obtained from Sinopharm Chemical Reagent. Glucose oxidase from *Aspergillus niger* (GOD, E C 1.1.3.4, 100 U/mg) was obtained from Biosharp. 3-aminopropiltrietoxysilane (APTES) and glutaraldehyde (GA) were purchased from Alfa Aesar. SiO_2 nanoparticles and glucose reagent were obtained from Sinopharm Chemical Reagent. Phosphate-buffered solution (0.1 M) was prepared by using a commercial pH-analyser, potassium phosphate monobasic and sodium phosphate dibasic from Sinopharm Chemical Reagent in deionized water. All other reagents were of analytical grade and used without further purification. Tri-distilled water was used throughout the experiments.

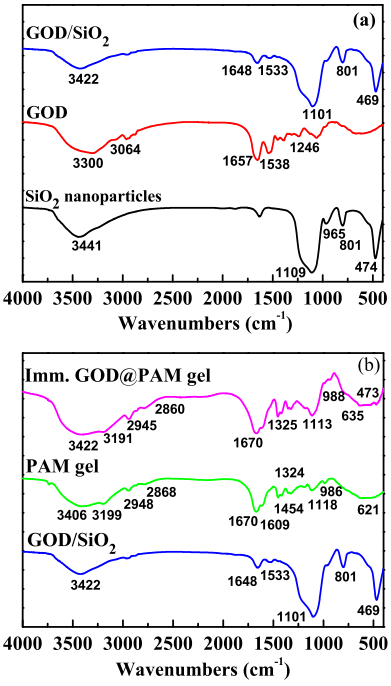

Fig. 1. FT-IR spectra of the GOD/SiO_2/gel film.

2.2. Immobilization of GOD on SiO_2 nanoparticles

The immobilization of GOD in a suitable matrix is beneficial to retention and reuse of isolated enzymes and is a trend to achieve the sensor with high performance. A typical immobilization of GOD on the surface of SiO_2 nanoparticles was performed as following [21]. At first, 4 mL of freshly prepared APTES solution (2% (v/v)) was stirred for 30 min to complete its hydrolyzation. 40 mg of SiO_2 nanoparticles was added into the solution and continued to stir for 18–24 h at room temperature. The modified nanoparticles were collected by centrifugation and washed for several times with deionized water and redispersed in a certain volume of phosphate buffer solution (PBS, 0.1 M, pH = 7.4). 160 μL (25% v/v) of GA was added into the mixture and stirred slowly for 1.5 h at 25 °C. The mixture was washed several times with PBS buffer (0.1 M, pH = 7.4), and once with PBS buffer (0.1 M, pH = 6.5). The activated SiO_2 nanoparticles were obtained. Then, GOD was immobilized on the surface of SiO_2 nanoparticles and the specific steps were as follows. The activated SiO_2 nanoparticles were dispersed in PBS buffer, the GOD was added to the mixture to control the concentration of GOD about 4 mg/dL, and put the mixture into a centrifuge tube at 4 °C for 12 h with occasional shaking. The SiO_2 nanoparticles cross-linked with GOD were washed in PBS buffer for several times to remove the residual reactants and free GOD, and then stored in PBS buffer (0.1 M, pH = 7.0) under 4 °C for use.

In order to verify whether the GOD was immobilized on the surface of SiO_2 nanoparticles, the FT-IR was used to test it, as shown in Fig. 1(a). The FT-IR spectra of GOD shows four infrared bands with the center positions at 3300 cm^{-1} (amide A), 1657 cm^{-1} (amide I), 1538 cm^{-1} (amide II) and 1246 cm^{-1} (amide III). The peak at 3300 cm^{-1} can be related to amide A with a shoulder at 3064 cm^{-1}. A peak at 1657 cm^{-1} is attributed to C=O stretching vibrations of the peptide linkages in the backbone and the band at 1538 cm^{-1} is associated with the combination of C—N stretching and N—H