Biosensors and Bioelectronics 25 (2010) 1434–1438

Contents lists available at ScienceDirect

Biosensors and Bioelectronics

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

Synergistic contributions of fullerene, ferrocene, chitosan and ionic liquid towards improved performance for a glucose sensor

Wei Zhilei[a], Li Zaijun[a,*], Sun Xiulan[b], Fang Yinjun[c], Liu Junkang[a]

[a] School of Chemical and Material Engineering, Jiangnan University, Wuxi 214122, China
[b] School of Food Science and Technology, Jiangnan University, Wuxi 214122, China
[c] Zhejiang Zanyu Technology Co., LTD., Hangzhou 310009, PR China

ARTICLE INFO

Article history:
Received 22 August 2009
Received in revised form 24 October 2009
Accepted 28 October 2009
Available online 6 November 2009

Keywords:
Fullerene
Ferrocene
Ionic liquid
Glucose
Biosensor

ABSTRACT

The paper describes an ingenious approach for the fabrication of a promising glucose sensor, GOx/C_{60}-Fc-CS-IL, that exploits the synergistic beneficial characteristics of fullerene (C_{60}), ferrocene (Fc), chitosan (CS) and ionic liquid (IL) for glucose oxidase (GOx). Cyclic voltammetry, impedance spectroscopy and chronoamperometry were used to evaluate performance of the biosensor, respectively. Since efficient electron transfer between GOx and the electrode was achieved, the biosensor exhibits a high sensitivity (234.67 nA nM^{-1} cm^{-2}), low detection limit (S/N = 3) (3×10^{-9} M), fast response time (0.752 s), wide calibration range (from 1×10^{-8} M to 1×10^{-5} M) and excellent long-term stability (30 weeks). The apparent Michaelis–Menten constant (K_M) of GOx on the composite medium, 0.03 mM, shows high bioelectrocatalytic activity of immobilized enzyme toward glucose oxidation. Due to low operating potential (100 mV), the biosensor is relatively insensitive to electroactive interfering species in human blood such as ascorbic acid, and uric acid, which are commonly found in blood samples. Excellent electrochemical reversibility, high sensitivity and stability, technically simple and possibility of preparation at short period of time are of great advantages of these glucose biosensors.

1. Introduction

Glucose sensors have been extensively investigated due to their important clinical application. Glucose oxidase (GOx) is the most popular enzyme in the development of glucose sensors, which catalyzes the oxidation of glucose that can be amperometrically detected at electrodes (Murphy et al., 2009). To improve the performance of GOx based biosensors, effective immobilization of GOx in a biocompatible environment and inclusion of components that augment electron transfer between electrode and GOx are strongly required (Rappathy et al., 2009). An ideal immobilization method for enzyme should employ mild chemical conditions and allow effective enzyme immobilization within a short period. In particular, electrode materials with a large surface-to-volume ratio can increase the amount of immobilized enzyme, minimize the barriers for mass transportation between the substrate and the product, and provide a chemically and mechanically robust system. Fullerene (C_{60}), ferrocene (Fc), chitosan (CS) and ionic liquid (IL) have proven to meet the above-mentioned requirements (Franzoi et al., 2009; Fu et al., 2009; Wang et al., 2009; Goyal et al., 2007). Strategies have been independently developed either on the development of matrix for enzyme immobilization or preparation of electrocatalytic components. However, studies devoted on concurrent improvement on both aspects toward GOx based sensors are scarce.

Fullerene possesses remarkable physical and chemical properties that make them interesting building blocks for supramolecular assemblies (Mukherjee et al., 2009). Fullerene-C_{60}, a truncated icosahedron, because of its triply degenerate, low lying LUMO at −4.3 eV is an excellent electron acceptor capable of accepting as many as six electrons reversibly. The delocalization of charges within the giant spherical carbon framework as well as rigid structure of the π sphere offers unique opportunities for stabilizing charges entities. The spherical fullerenes accelerates electron transfer and charge shift, but slows down charge recombination (Moore et al., 2002). Due to above remarkable electrochemical properties, C_{60} has been applied as novel mediator to fabricate biosensors for determination of dopamine (Goyal et al., 2008), nandrolone (Goyal et al., 2007), organic vapors (Lin and Shih, 2003), oxygen (Bouchtalla et al., 2002), glucose (Chuang and Shih, 2001), odorants (Szymanska et al., 2001) and urea (Wei and Shih, 2001).

Fc and its derivatives are attracting keen interest in the area of electroanalysis for their unique redox behavior. Because Fc is one of the most popular electrochemical active groups and the redox reaction of Fc^+/Fc is completely reversible, many scientists have used it in chemical modified electrodes (Elanchezhian and Kandaswarny,

* Corresponding author. Tel.: +86 5105811863; fax: +86 5105811863.
E-mail address: zaijunli@263.net (L. Zaijun).

0956-5663/$ – see front matter © 2009 Elsevier B.V. All rights reserved.
doi:10.1016/j.bios.2009.10.045

2009; Qiu et al., 2009; Raoof et al., 2009). However, Fc and its derivatives can be poorly adsorbed onto the electrode surface. Thus, many materials had been used to improve the attachment of Fc and its derivatives to the electrode surface such as CS and IL (Zhao et al., 2008; Zhou et al., 2008).

ILs are compounds consisting entirely of ions that exist in liquid state around room temperature. The investigation of ILs has gained increasing attention because of their unique chemical and physical properties such as negligible vapor pressure, low toxicity, wide potential window, high ionic conductivity and good solubility. ILs had been widely used in extraction (Li et al., 2007) and non-aqueous biocatalysts (Shan et al., 2008). Recently, ILs were also investigated as biocompatible materials for the fabrication of biosensors (Shangguan et al., 2008; Sun et al., 2009; Tu et al., 2009). These researches have confirmed enzyme can maintain high activity and stability in a suitable IL. However, IL lacks enough film forming ability to immobilization enzyme on the electrodes and strongly requires an additional material to aid forming film such as CS (Ragupathy et al., 2009).

Considering the benefits of C_{60}, Fc, CS and IL, we have integrated them in a biosensor fabrication to exploit their synergistic contributions on the improvement of sensor characteristics in this study. To the best of our knowledge, a glucose biosensor has not been reported so far with these integrated components. Further, we have utilized ingenious methodologies to integrate C_{60}, Fc, CS and IL for the fabrication toward glucose biosensor.

2. Experimental

2.1. Materials and reagents

C_{60} (99.5%), Fc, D-(+)-glucose (99.5%), GOx (EC 232-601-0, 136300 U/g, from *Aspergillus niger*) and CS (85%) were obtained from Sigma–Aldrich Chemical Company (Mainland, China) and used without further purification. IL, 1,3-dibutylimidazolium bromide was synthesized as described elsewhere (Li et al., 2007). [DBIM][TF_2N] was obtained by anion exchange of 1,3-dibutylimidazolium bromide with bis(trifluoromethylsulfonyl)amine lithium salt. A phosphate-buffered saline (PBS, pH 7, Na_2HPO_4-KH_2PO_4-NaCl-KCl, 0.01 M) was prepared. A 0.01 M glucose stock solution was prepared by dissolving it in pH 7.0 PBS and stored in a refrigerator at 4 °C. A 150 μM C_{60} solution was prepared by dissolving it in dichloromethane. Fc saturated solution was prepared by dissolving it in ethanol. CS solution was prepared by dissolving 5 g of CS in 100 mL of 1% (v/v) acetic acid. IL solution was prepared by dissolving 5 mL of the IL in 50 mL ethanol. All other reagents employed were of analytical reagent grade or with highest quality and were purchased from Shanghai Chemical Company (Shanghai, China), and ultra pure water (18.2 ΩM cm) purified from a Milli-Q purification system was used throughout the experiment.

2.2. Apparatus

Electrochemical experiments were performed with an IM6e electrochemical system (ZAHNER Elektrik, German) and CHI660B electrochemical workstation (Shanghai, China). A conventional three electrode system was used with Ag/AgCl (saturated KCl) electrode as the reference electrode, platinum wire as the counter electrode, and a modified glassy carbon electrode (GCE, 2 mm in diameter) as working electrode. If not mentioned, all potentials given below were relative to Ag/AgCl (saturated KCl) electrode.

2.3. Electrodes preparation

Procedure of the electrode preparation includes five assemble processes, i.e. pretreatment of GCE, immobilization of C_{60}, Fc, CS-IL and GOx on the electrode surface (shown in Fig. 1). (1) GCE was polished successively with 1.0, 0.3, and 0.05 μm alumina powder, and sonicated in a 6.0 M nitric acid/doubly distilled water and ethanol/doubly distilled water for 20 min, respectively. Then, GCE as working electrode was subjected to cyclic scanning in 0.5 M H_2SO_4 solution in a potential range from −0.1 V to 1.0 V. When the cyclic voltammogram was almost unchanged, the electrode was taken out, cleaned with water and dried under a stream of nitrogen. (2) A 40 μL of the C_{60} solution was coated onto the surface of the pretreated GCE using a microsyringe and dried in a stream of hot air (50 °C). (3) 0.1 mL of the Fc solution was well mixed with 0.1 mL of the IL solution, and 2 μL of the mixture solution was then coated on the surface of C_{60}-GCE using a microsyringe and dried in a stream of hot air (50 °C). (4) 50 μL the IL was well dispersed in 0.2 mL of the CS solutions. After the mixture was sonicated for 30 min, its 5 μL was coated on the surface of the C_{60}-Fc-GCE using a microsyringe and dried in a stream of hot air (50 °C). (5) GOx stock solution was prepared by dissolving 10 mg of it in 1.0 mL of pH 7.0 PBS. 8 μL of the enzyme solution was dropped onto the surface of C_{60}-Fc-CS-IL-GCE to fabricate the glucose biosensor.

2.4. Electrochemical studies

Cyclic voltammetry and electrochemical impedance spectroscopy were performed using an IM6e electrochemical system. The impedance spectra were measured in the frequency range from 10^5 Hz to 1 Hz in a potential of 0.20 V versus Ag/AgCl (saturated KCl), with a voltage amplitude of 5 mV. The impedance Z was expressed in terms of a real (Z') and an imaginary (Z'') component. Impedance signals were recorded after reaction between the electrodes and the unstirred substrate solution for 5 min. Impedance measurements were performed in 20 mL of a pH 7.0 PBS containing 1.0 mM $K_3[Fe(CN)_6]$/$K_4[Fe(CN)_6]$ (1:1 mixture as redox probe). The current–time curves were recorded on the CHI660B electrochemical workstation in a stirred cell for successive additions of glucose solution at an operating potential of +100 mV, in which a 5 mL of home-made cell and agitation speed of 2000 rpm were employed for the measurement.

3. Results and discussion

3.1. Electrochemical properties of GOx/C_{60}-Fc-CS-IL modified electrode

The interfacial properties of electrodes were investigated by electrochemical impedance spectroscopy. Fig. 2A presents typical Nyquist plots. From Fig. 2A, we observed the clear semicircle portions in Nyquist plots of electrochemical impedance spectra for the bare GCE, C_{60}-GCE, C_{60}-Fc-GCE, C_{60}-Fc-IL-CS-GCE and GOx/C_{60}-Fc-CS-IL-GCE, indicating they are the electron transfer limited process, the electron transfer resistances of the redox for all modified electrodes are obviously higher than that of bare electrode, which is because materials-modified on the surface of the electrode will partly block the electron transfer of $[Fe(CN)_6]^{3-/4-}$ solution to the electrode, and the electron transfer resistances of GOx/C_{60}-Fc-CS-IL-GCE is remarkably lower than that of C_{60}-Fc-IL-CS-GCE, that reveals efficient electron transfer between the electrode and GOx was achieved for the glucose sensor.

Cyclic voltammograms of Fc-GCE, C_{60}-Fc-GCE and GOx/C_{60}-Fc-CS-IL-GCE were recorded in a pH 7.0 PBS at a scan rate of 100 mV/s (Fig. 2B). It can be clearly seen from Fig. 2B cyclic voltam-

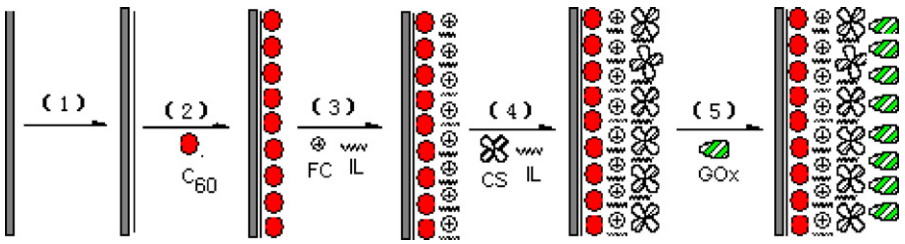

Fig. 1. General procedure for fabrication of GOx/C_{60}-Fc-CS-IL-GCE.

mograms for modified electrodes showed a pair of redox peaks. In particular, synergistic electrocatalytic properties of C_{60}, Fc, CS and IL lead to lowering of peak potentials with enhancement in the peak current for C_{60}-Fc-GCE and GOx/C_{60}-Fc-CS-IL-GCE, this would improve the selectivity and sensitivity of the sensor.

The electrochemical stability of GOx/C_{60}-Fc-CS-IL-GCE was also checked by a repetitive potential sweep at a scan rate of 100 mV/s. It was found that peak currents maintained the same original intensity after 100 scans (no shown), which demonstrated the electrochemistry of the modified electrode is fairly stable and the leakage of C_{60} and Fc form the surface of the GOx/C_{60}-Fc-CS-IL-GCE can be neglected. Moreover, the effect of varying the scan rate on the performance of the electrode was also studied and the results were shown in Fig. 2C. With an increasing scan rate, the CV peak currents of the GOx/C_{60}-Fc-CS-IL-GCE increased in the scan rate range from 10 mV/s to 600 mV/s, but peak potentials were nearly independent of the scan rate, indicating that the C_{60}-Fc-CS-IL mediator was efficiently connected on GCE for facile charge transfer.

3.2. Electrochemical response of GOx/C_{60}-Fc-CS-IL-GCE biosensor to glucose

The modified electrode exhibits stable and reversible electrochemistry, it can be used as electron transfer mediator to shuttle electrons between GOx and the modified electrode. GOx was selected as a model enzyme to evaluate the electrocatalytic ability of the C_{60}-FC-CS-IL-GCE. Fig. 3 shows the CVs of the GOx/C_{60}-FC-CS-IL-GCE in the absence and presence of glucose in 0.1 M pH 7.0 PBS. In the absence of glucose, the reversible electrochemical behavior of Fc was observed on the GOx/C_{60}-FC-CS-IL-GCE (a). After addition of 1.0 μM glucose (b) and 5.0 μM glucose (c) in pH 7.0 PBS at 100 mV/s, the cathodic and anodic peak current increased dramatically, which clearly showed the catalytic properties of modified electrode to glu-

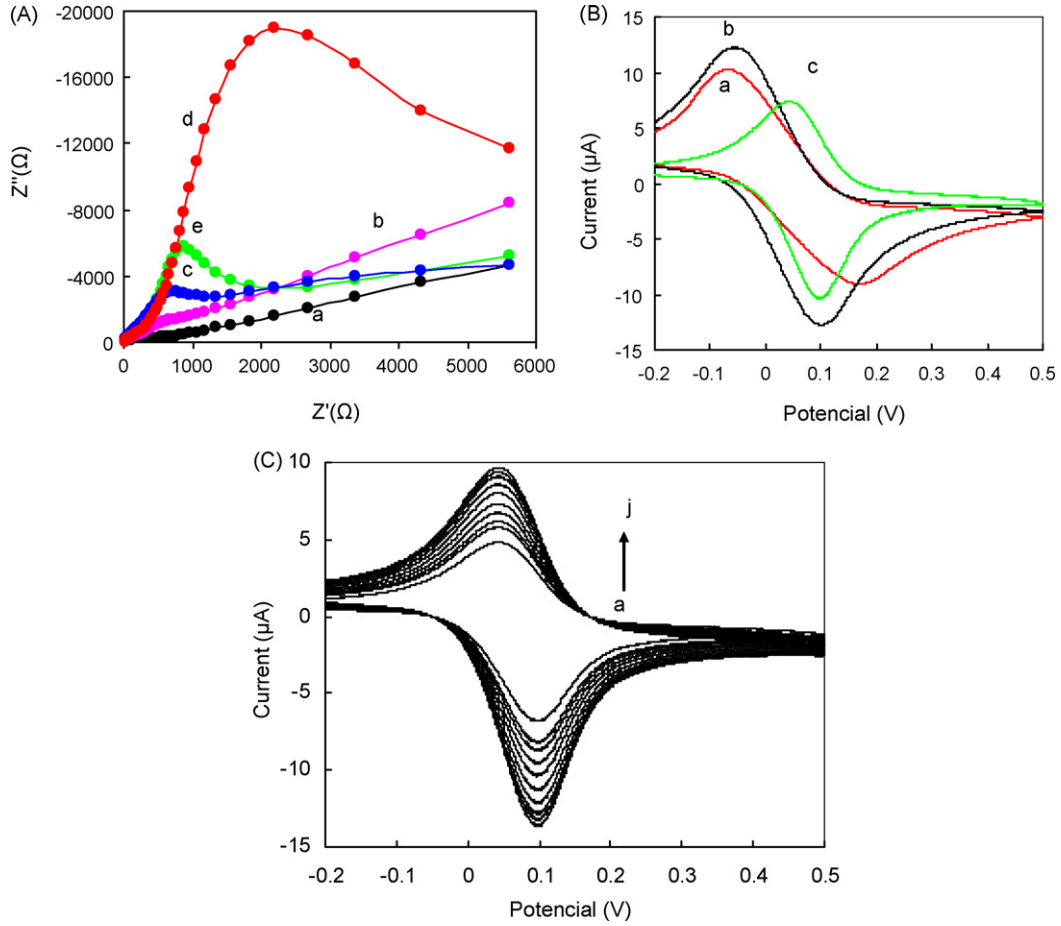

Fig. 2. (A) Faradaic impendence spectra that corresponded to bare GCE (a), C_{60}-GCE (b), C_{60}-Fc-GCE (c), C_{60}-Fc-CS-IL-GCE (d) and GOx/C_{60}-Fc-CS-IL-GCE (e) in the pH 7.0 PBS containing 1.0 mM of $[Fe(CN)_6]^{3-/4-}$, respectively. (B) Cyclic voltammograms of Fc-GCE (a), C_{60}-Fc-GCE (b) and GOx/C_{60}-Fc-CS-IL-GCE (c) in pH 7.0 PBS at 100 mV/s. (C) Cyclic voltammograms of GOx/C_{60}-Fc-CS-IL-GCE in pH 7.0 PBS at 10, 25, 50, 75,100, 200, 300, 400, 500 and 600 mV/s (from a to j), respectively.

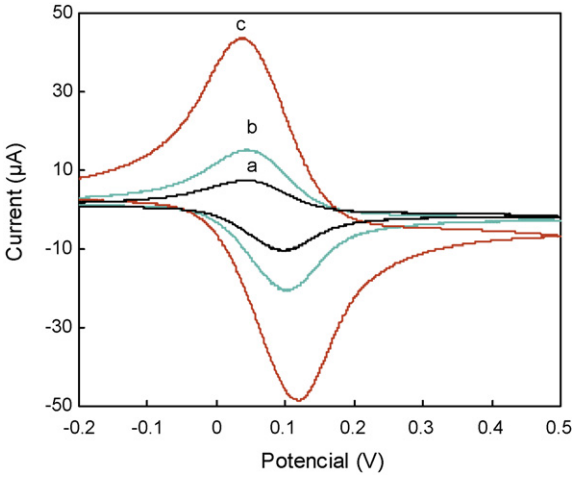

Fig. 3. Cyclic voltammograms of GOx/C_{60}-Fc-CS-IL-GCE in the absence glucose (a) and presence of 1.0 μM glucose (b) and 5.0 μM glucose (c) in pH 7.0 PBS at 100 mV/s.

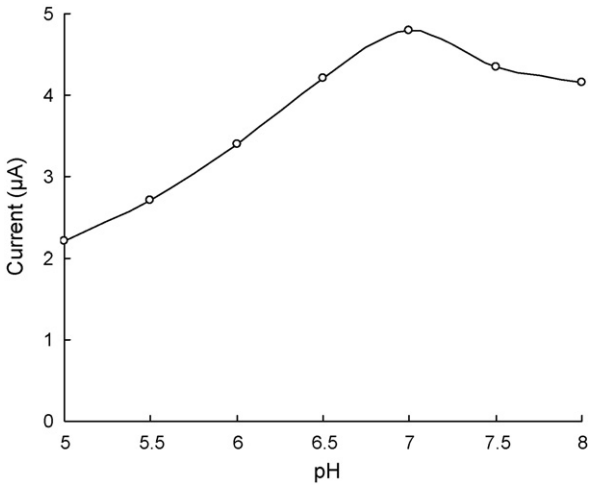

Fig. 4. Amperometric response of GOx/C_{60}-Fc-CS-IL-GCE in different pH PBS containing 0.8 μM of glucose. Potential: +100 mV.

cose. Since the reduction current responses are obviously higher than the oxidation current responses, the amounts of glucose is determined by monitoring the increase in the reduction peak current of the GOx/C_{60}-FC-CS-IL-GCE in oxygen saturated solution. Because background currents influence readings, the operation of the sensor at low potential is analytically desirable to reduce the electrochemical interferences, this allowing high selectivity. Given these considerations, a low operating potential of 100 mV was chosen to demonstrate the applicability of this biosensor to the sensitive detection of glucose.

The electrochemical reaction in the system could be described by a well known following mechanism (Zhang et al., 2004): In the presence of glucose and enzyme GOx, glucose is oxidized to gluconolactone and the coenzyme GOx (ox) is converted to GOx (red). The resulting reduced form of the enzyme GOx (red) is then reoxidized by the ferrocene ion, yielding ferrocene, which in turn is reoxidized at the underlying electrode with generation of an amperometric catalytic anodic current. During above process, synergistic contributions of C_{60} and Fc as composite mediator improve the electron relays for activation of oxidation of the analyte (Goyal et al., 2007), which would accelerate electrochemical reaction, and network of CS and IL offers an excellent microenvironment for GOx, this results a high enzyme activity and rapid response to glucose.

The influence of the buffer pH is very essential to the sensitivity of the biosensors, because the pH affects not only the electrochemical behavior of C_{60} and Fc but also the bioactivity of GOx. The optimal pH reported for GOx is usually in the range of 6.5–7.5 (Liu et al., 2003), which varies with immobilization method and microenvironment around the enzyme. Thus, the effect of pH was examined in the range of pH 5.0–8.0. This biosensor showed a maximum response at pH 7.0 (Fig. 4). As a consequence, the performance of the glucose biosensor was evaluated at pH 7.0 in this work.

3.3. Amperometric detection of glucose at GOx/C_{60}-Fc-CS-IL biosensor

Fig. 5 shows the typical current–time responses at GOx/C_{60}-Fc-CS-IL biosensor for successive addition of glucose. The time required to reach 95% of the maximum steady-state current was less 0.752 s, indicating a very fast response, which was mainly due to the existence of C_{60} and Fc as mediator of electron transfer and the well-conductive properties of C_{60} and IL. With the increasing glucose concentration the amperometric response increased linearly in the range from 1×10^{-8} M to 1×10^{-5} M. The linear equation was $I_p = 7.3679C + 1.2068$, with a statistically significant correlation coefficient of 0.9998 and a slope of 234.67 nA nM^{-1} cm^{-2} (sensitivity), which I_p is in μA and concentration in μM. The detection limit was 3×10^{-9} M that was obtained from the signal-to-noise characteristics of these data (S/N = 3). The glucose biosensor was measured ten times in 2.0 μM glucose standard solution under the same conditions repeatedly. A relative standard deviation of 1.3% for the measurements was obtained, this indicated the measurement has high precision for the sensor. The proposed sensor was stored in air at ambient conditions and its sensitivity was checked every week. The response of the sensor was 95% of its initial value after 30 weeks which shows long-term stability and very good sensitivity for the analysis of real samples. These analytical parameters are better than results previously reported for electroanalytical determination of glucose with difference sensors and biosensors (Qiu et al., 2009; Wang et al., 2009).

3.4. Michaelis–Menten constant

When glucose concentration higher than 0.1 mM, a response plateau was observed, showing the characteristics of the Michaelis–Menten kinetic mechanism. The apparent Michaelis–Menten constants (K_M) can be calculated according to

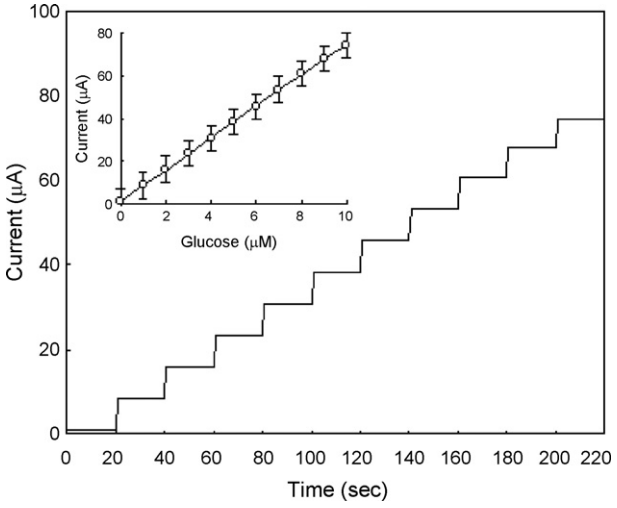

Fig. 5. Chronoamperometric current response of GOx/C_{60}-Fc-CS-IL-GCE to successive addition of 1 μM of glucose at +100 mV in pH 7.0 PBS solution. Inset: Calibration plot of concentration of glucose (0–10 μM) vs. current.

Table 1
Determination of glucose in blood serum samples.*.

Samples	Glucose found by proposed method (mM)	Glucose found by hexokinase method (mM)
Serum 1	8.41 ± 0.12 $F = 1.19$, $t = 0.72$	8.37 ± 0.11
Serum 2	3.23 ± 0.09 $F = 2.09$, $t = 0.28$	3.25 ± 0.13
Serum 3	3.66 ± 0.06 $F = 2.25$, $t = 1.32$	3.70 ± 0.09
Serum 4	4.53 ± 0.10 $F = 1.00$, $t = 0.40$	4.51 ± 0.15
Serum 5	5.11 ± 0.11 $F = 1.19$, $t = 1.08$	5.05 ± 0.12

* Results expressed as: $X \pm st/\sqrt{n}$ ($n = 5$), where X is the mean of n observations of x, s is the standard deviation, t is distribution value chosen for the desired confidence level, the t- and F-values refer to comparison of the proposed method with the hexokinase method. Theoretical values at 95% confidence limits: $F = 6.39$, $t = 2.78$.

the Lineweaver–Burk equation:

$$\frac{1}{I_{ss}} = \frac{1}{I_{max}} + \frac{K_M}{I_{max}}\frac{1}{C} \quad (1)$$

Here I_{ss} is the steady-state current after glucose addition, C is the buck concentration of substrate and I_{max} is the maximum current measured under saturated substrate (glucose) solution. The apparent K_M of the proposed sensor was calculated for glucose according to data in the insert in Fig. 5. The results indicated the K_M value is between 0.034 mM and 0.029 mM for different glucose concentration and their mean value is 0.03 mM, implying that the biosensor exhibits an excellent affinity for glucose. This value of K_M for GOx in this work is much smaller than that obtained the native GOx in solution (27 mM) (Rogers and Brandt, 1971) and at glucose biosensor based on sol–gel organic–inorganic hybrid material (20 mM) (Wang et al., 1998), Pt nanoparticles/mesoporous carbon matrix (10.8 mM) (Yu et al., 2008), GOx immobilized at chitosan and Au nanoparticles (10.5 mM) (Wu et al., 2007), boron doped carbon nanotube modified electrode (15.19 mM) (Chin et al., 2008), single-walled carbon nanotube modified electrode (8.5 mM) (Liu et al., 2008), ferrocene-modified multiwalled carbon nanotube nanocomposites (3.12 mM)(Qiu et al., 2009), and immobilization of osmium complex and glucose oxidase onto carbon nanotubes modified electrode (0.91 mM) (Salimi et al., 2009). The smaller value of K_M validates synergistic contributions of C_{60}, Fc, CS and IL results in a higher enzymatic activity and affinity for glucose.

3.5. Interferences study

The influence of electrochemical interference to glucose on the current response of Gox/C_{60}-Fc-CS-IL-GCE was evaluated. The results indicated the injection of 100-fold ascorbic acid and 100-fold uric acid did not influence the current response of glucose (1.0 μM). Electrochemical detection of glucose was possible at a lower potential (100 mV) due to the synergistic electrocatalytic influences of C_{60}, Fc, CS and IL. Hence, there is negligible interference from other electro-active components.

3.6. Application of the biosensor for determination of glucose in human serum samples

To evaluate the ability of the biosensor for routine analysis, the biosensor was applied to the determination of glucose in blood serum samples. Here, 10 μL of blood serum sample was added to 10 mL of pH PBS and the amperometric response of glucose was recorded at +100 mV. The results are presented in Table 1. This table also shows that there is a very good agreement between the results obtained by the proposed biosensor and those obtained by application of a routine enzymatic method (using hexokinase method) in a local hospital.

4. Conclusion

A highly sensitive and stable glucose biosensor based on the synergetic contributions of C_{60}, Fc, CS and IL has been successfully fabricated. The electrocatalytic activity of C_{60} and Fc remarkably improves the electron relays for activation of oxidation of the glucose and accelerate electrochemical reaction. The network of CS-IL provides a favorable microenvironment to keep the bioactivity of GOx, and the electron conduction pathways for GOx through Fc and C_{60}. Excellent sensitivity, selectivity, stability, fast response time and ease of preparation, low cost and acceptable accuracy for sample determination made this sensor ideal for detection of glucose in real samples such as blood serum samples.

Acknowledgements

The authors acknowledge the financial support from the National Natural Science Foundation of China (No. 20771045 and 20676052), the National High Technique Development Plan of 863 (2007AA10Z428), sponsored by Qing Lan Project and the Natural Science Foundation of Zhejiang Province (No. Y4080404).

References

Bouchtalla, S., Auret, L., Janot, J.M., Deronzier, A., Moutet, J.C., Seta, P., 2002. Mater. Sci. Eng. C: Biol. S 21, 125–129.
Chin, X., Chen, J., Deng, C., Xiao, C., Yang, Y., Nie, Z., Yao, S., 2008. Talanta 76, 763–767.
Chuang, C.W., Shih, J.S., 2001. Sens. Actuators B: Chem. 81, 1–8.
Elanchezhian, V.S., Kandaswarny, M., 2009. Inorg. Chem. Commun. 12, 161–165.
Franzoi, A.C., Dupont, J., Spinelli, A., Vieira, I.C., 2009. Talanta 77, 1322–1327.
Fu, Z.Q., Wan, M.Y., Tu, Y.F., 2009. J. Anal. Sci. 25, 79–82.
Goyal, R.N., Gupta, V.K., Bachheti, N., 2007. Anal. Chim. Acta 597, 82–89.
Goyal, R.N., Gupta, V.K., Bachheti, N., Sharma, R.A., 2008. Electroanal 20, 757–764.
Qiu, J.D., Zhou, W.M., Guo, J., Wang, R., Liang, R.P., 2009. Anal. Biochem. 385, 264–269.
Li, Z.J., Wei, Q., Yuan, R., Zhou, X., Liu, H.Z., Shan, H.X., Song, Q.J., 2007. Talanta 71, 68–72.
Lin, H.B., Shih, J.S., 2003. Sens. Actuators B: Chem. 92, 243–254.
Liu, X., Shi, L., Niu, W., Li, H., Xu, G., 2008. Biosens. Bioelectron. 23, 1887–1890.
Moore, T.A., Moore, A.L., Gust, D., 2002. Philos. Trans. Biol. Sci. 357, 1481–1498.
Mukherjee, P., Bhattachary, S., Nayak, S.K., Chattopadhyay, S., Bhattachary, S., 2009. Chem. Phys. 360, 116–122.
Murphy, R., Tura, A., Clark, P.M., Holst, J.J., Mari, A., Hattersley, A.T., 2009. Diabetologia 52, 154–159.
Raoof, J.B., Ojani, R., Kolbadinezhad, M., 2009. J. Solid State Electrochem. 13, 1411–1416.
Ragupathy, D., Gopalan, A.I., Lee, K.P., 2009. Electrochem. Commun. 11, 397–401.
Rogers, M.J., Brandt, K.G., 1971. Biochemistry 10, 4624–4630.
Salimi, A., Kavosi, B., Hallaj, R., Babaei, A., 2009. Electroanal 21, 909–917.
Shan, H.X., Li, Z.J., Li, M., Ren, G.X., Fang, Y.J., 2008. J. Chem. Technol. Biotechnol. 83, 886–891.
Shangguan, X.D., Zhang, H.F., Zheng, J.B., 2008. Electrochem. Commun. 10, 1140–1143.
Sun, W., Zhai, Z.Q., Wang, D.D., Liu, S.F., Jiao, K., 2009. Bioelectrochemistry 74, 295–300.
Szymanska, I., Radecka, H., Radecki, J., Kikut-Ligaj, D., 2001. Biosens. Bioelectron. 16, 911–915.
Tu, W.W., Lei, J.P., Ju, H.X., 2009. Chemistry 15, 779–784.
Wang, B., Li, B., Deng, Q., Dong, S., 1998. Anal. Chem. 70, 3170–3174.
Wei, L.F., Shih, J.S., 2001. Anal. Chim. Acta 437, 77–85.
Wu, B.Y., Hou, S.H., Yin, F., Li, J., Zhao, Z.X., Huang, J.D., Chen, Q., 2007. Biosens. Bioelectron. 22, 838–844.
Yu, J., Yu, D., Zhao, T., Zeng, B., 2008. Talanta 74, 1586–1591.
Zhao, G.C., Xu, M.Q., Zhang, Q., 2008. Electrochem. Commun. 10, 1924–1926.
Zhang, F.F., Wan, Q., Wang, X.L., Sun, Z.D., Zhu, Z.Q., Xian, Y.Z., Jin, L.T., Yamamoto, K., 2004. J. Electroanal. Chem. 571, 133–138.
Zhou, H., Yang, W.W., Sun, C.Q., 2008. Talanta 77, 366–371.