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C_{60}-CdS Composite Films: Electrochemical Preparation And Electrocatalysis Of Hemoglobin In An Aqueous Solution

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ABSTRACT

The electrochemical behavior of a C_{60}/CdS film cast on GC electrode in the acetonitrile solution was investigated. Subsequent cyclic voltammetric scans transformed the precursor to a C_{60}-CdS composite film which exhibited quasi-reversible electron-transfer reactions. The formation and structure of the C_{60}-CdS composite films were characterized by UV-Vis and TEM. To explore the biological activities of the composite films, the electrocatalysis of Hb(hemoglobin) at the C_{60}-CdS composite film was also studied. Moreover, the preferable catalytic activity of C_{60}-CdS/Hb composite film towards hydrogen peroxide(H_2O_2) was demonstrated.

INTRODUCTION

C_{60} as novel all-carbon π-electron system has increasingly invited exploration of its outstanding new physical and chemical properties[1,2]. Among various particular properties, its electrochemical behavior is special. C_{60} can be cathodically reduced in aprotic electrolyte solution towards C_{60}^{-}, C_{60}^{2-}, …, C_{60}^{6-} in six reversible one electron steps[3], while thin solid films of C_{60} exhibit irreversible reductions, with a reconstruction of the film structure[4,5].

Electrochemical studies of fullerene films have been very rich in nonaqueous solution[6-11], however, the electrochemistry of fullerene films has been very limited in an aqueous solution[12,13], and that both the reduction and the oxidation were completely irreversible[14]. To solve the problem, the electrochemistry of supramolecular complex films of fullerenes with cyclodextrins and calixarenes in solution containing water was investigated[15-21], and a series of reversible electroreduction waves were obtained. On the other ways, C_{60} was embedded in cationic surfactant lipids, such as tetraoctylphosphonium bromide, tetraoctylammonium bromide, ditetradecyldimethylammonium poly(styrene sulfonate), and didodecyldimethylammonium bromide, etc., and these composite films also exhibit reversible electroreduction character in an aqueous solution[22].

Semiconductor NPs (nanoparticles) have been extensively studied in the past two decades due to unique optical and electronic properties which are not available in either discrete or in bulk solids[23, 24]. These NPs exhibit quantum size effects such as a blue shift of absorption onset, a change of electrochemical potential of band edge, and an enhancement of photocatalytic activities with decreasing crystallite size. Among the NPs, CdS is the most promising material for detecting visible radiation due to its primary band gap of 2.42eV at the room temperature. Meanwhile, CdS behaved as n-type

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semiconductor, and it can be used as an electron acceptor. These electrons can transfer to the surface of particles rapidly because of the quantum size effect of CdS NPs, resulting in higher charge detaching efficiency[25]. At present, there is a keen interest in the composite films of fullerenes and semiconductor NPs from both fundamental and practical points of views[26,27]. Our research is focused on the electrochemistry of films formed of fullerenes and semiconductor in the acetonitrile solution and their biological activities in an aqueous solution in order to produce electrochemically stable fullerene composites for further application of biosensors.

In this paper, we report on the creation of a C_{60}-CdS composite film by cyclic voltammetry. Electrochemistry of the film made from CdS NPs containing C_{60} on a glassy carbon electrode was investigated in the acetonitrile solution. It was found that the composite film exhibited quasi-reversible multiple-step electron-transfer reactions. To explore the possible biological application, electrocatalysis of Hb at C_{60}-CdS films was also studied. Subsequently, the C_{60}-CdS/Hb film was demonstrated to exhibit preferable catalytic activity towards hydrogen peroxide(H_2O_2).

EXPERIMENTAL

C_{60}(>99.9%) was purchased from Peking University. CdS NPs were synthesized according to literature but with some improvement[28]. In a typical procedure, 0.25g sublimed sulfur(0.007812mol) was dissolved in 200ml DMSO at 100°C for about 1 h, then increased to 150°C. At this temperature, a preheated solution of 2.5g cadmium acetate(9.398mmol) in 200mL DMSO was added. The reaction solution was kept in 150^0C about one hour and then cooled to ambient temperature. Adding 100mL acetone to the solution, the precipitant was collected by centrifuging, and then washed with acetone two times. After dried under vacuum at room temperature, the CdS powders were obtained. The prepared CdS NPs were stored by keeping away from light. $TBAPF_6$(Aldrich) was dried for 6 h in vacuum before use. Acetonitrile(CH_3CN)(HPLC grade, Labscan Asia Co., Thailand) was used as purchased. Toluene($C_6H_5CH_3$)(Park Co. Dublin, Ireland) was dried with sodium, refluxed for 6h, and then distilled. The purified toluene was stored in the presence of sodium. Hb was purchased from Sigma. Other used reagents were analytical grade. Water was doubly distilled from an all-quartz still.

Immediately before use, a glassy carbon electrode (3mm diameter) was polished to a mirror finished with emery paper and alumina slurry (1.0 and 0.3μm), ultrasonically cleaning in water for ~5 min, and dried in a high-purity nitrogen stream. C_{60} was put into toluene to create a saturated solution. At the same time, CdS NPs were added into ethylene tetrachloride to create a solution with the concentration of 0.1M($mol \cdot L^{-1}$). Then, 0.1ml CdS NPs(0.1M) solution were dispersed in 0.75ml ethanol, added into 0.03ml C_{60} saturated solution, and then ultrasonicated for 10?15min to create a suspension. To prepare the C_{60}/CdS mixture film, 15μl of the suspension was directly cast on a glassy carbon(GC) electrode surface and the solvent was allowed to evaporate at the room temperature.

UV-Vis spectrum was done on a Cintra 10e UV-Vis Spectrometer(GBC, Australia). TEM(transmission electron microscopy) measurements were conducted on Philips CM20 and JEOL 2010F transmission electron microscopes with an accelerating voltage of 200kV and SEM(scanning electron microscopy) was carried out with a KYKY2000 SEM instrument in the secondary electron emission mode. Cyclic voltammetry scan(CHI610B, Inc., Austin, USA) was performed with a three-electrode configuration .The modified GC electrodes were used as the working electrode. A Pt wire electrode was served as the auxiliary electrode, and a Ag/AgCl electrode was used as the reference. All electrochemical experiments were performed in a high purity N_2 atmosphere at ambient temperature.

RESULTS AND DISCUSSION

The cyclic voltammetric (CV) scans were carried out on the freshly cast films of C_{60}/CdS on the GC electrode in the acetonitrile solution of 0.1M $TBAPF_6$ between 0 and -1.8V. After two or three scans, a typical CV profile of the C_{60}-CdS composite film is obtained as shown in figure 1. Some difference between the initial scans(see figure S1 in supporting information) and subsequent scans was observed. The current response increased a little and the peak shape was well-defined with increasing CV cycles. Notice that a distinction is

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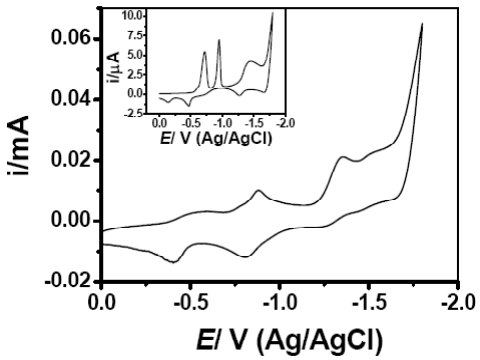

Figure 1 : Cyclic voltammogram of C_{60}-CdS composite film on GC electrode in acetonitrile solution containing 0.1mol l^{-1} $TBAPF_6$, and (inset) C_{60} film on GC electrode in acetonitrile solution containing 0.1M $TBAPF_6$. Scan rate: 0.1V s^{-1}.

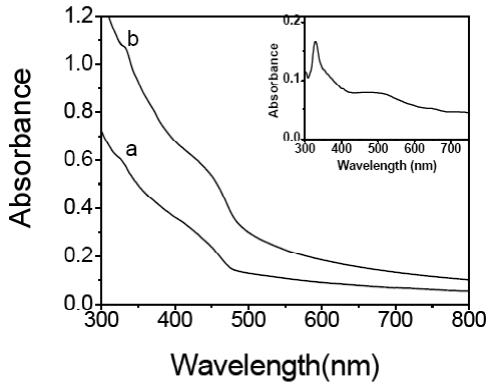

Figure 2 : UV-Vis spectra of: (a) C_{60}/CdS mixture film, (b) C_{60}-CdS composite film, and inset C_{60} film

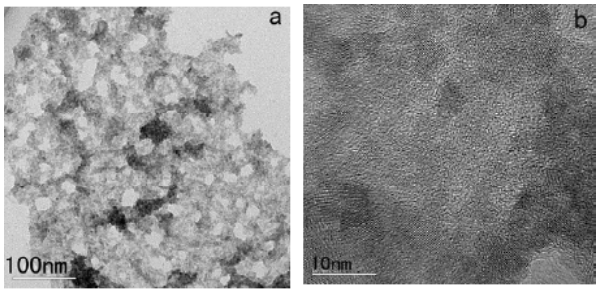

Figure 3 : TEM images of (a) C_{60}/CdS mixture films and (b) C_{60}-CdS composite films

made here between the C_{60}/CdS mixture and the C_{60}-CdS composites: the former is characterized by feeble physical adsorption, whereas the latter involves strong interactions between C_{60} and CdS induced by the CV scans. Namely, the composite film exhibits quasi-reversible multiple-step electron-transfer reactions corresponding to C_{60}/C_{60}^{-},C_{60}^{-}/C_{60}^{2-} and C_{60}^{2-}/C_{60}^{3-}[29], which resemble those of C_{60} dissolved in an acetonitrile/toluene solvent mixture on a GC working electrode (see Figure 2 in supporting information). This is in contrast with the reduction waves of C_{60} film in an acetonitrile solution containing 0.1 M $TBAPF_6$, where there are large splitting between the first two reduction waves and corresponding reoxidation waves (see inset in figure 1)[30]. These indicated that electrochemical response of C_{60} embedded in cast films of CdS NPs on GC electrode was different from that of the pristine C_{60} film.

The ethanol suspension of a C_{60}-CdS composite film obtained after a number of CV scans together with the C_{60}/CdS mixture film and C_{60} film were characterized by UV-Vis absorption spectroscopy(see figure 2). Compared with the typical absorption peak of C_{60} at 326 nm(see inset in figure 2), besides the absorption peak of CdS between 420 and 510 nm, the absorption peak of C_{60} in C_{60}/CdS mixture film is unchanged(curve a), which is in comparison with a red shift of about 8nm for C_{60}-CdS composite film(curve b). The phenomenon indicates the interaction between C_{60} and CdS NPs in C_{60}-CdS composite film after the inducement of CV scans. Similar observations were reported in the functionalization of C_{60} previously, but here the functional substance is CdS NPs. It follows that, in the C_{60}-CdS composite film prepared by CV, C_{60} is most probably attached to the CdS by covalent interaction, which is in line with the existing literature reports[31,32].

In order to understand the role of CV scans in forming the C_{60}-CdS composite film, TEM was employed to examine the surface morphologies of C_{60}/CdS mixture film and C_{60}-CdS composite film (Figure 3). For the C_{60}/CdS mixture film, crystal structured CdS NPs and black NPs of C_{60} exist individually. Many aggregates of C_{60} are also observed (Figure 3a). However, the TEM image of the C_{60}-CdS composite film obtained after several CV scans shows that the nanoparticles of C_{60} are uniformly dispersed on the CdS NPs (Figure 3b). It is likely that the C_{60}-CdS composite films are formed in the following way. CdS is an n-type semiconductor, which can be used as an electron acceptor. To begin with, C_{60} molecules are adsorbed on the surface of CdS NPs by electron affinity. However, because C_{60} moieties are reactive, the additional free C_{60} molecules will tend to aggregate with the already adsorbed C_{60} on CdS NPs to form C_{60} clusters. Conceivably, the CV scans to negative potentials pro-

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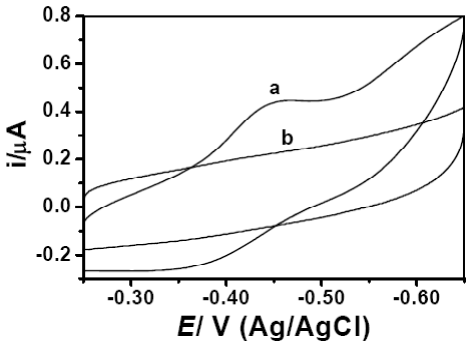

Figure 4 : Cyclic voltammograms for (a) C_{60}-CdS composite film and (b) CdS film on GC electrode in 0.1 M PBS

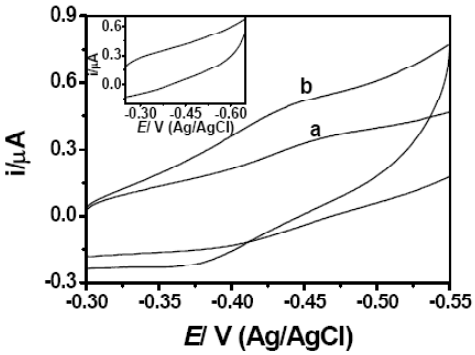

Figure 5 : Cyclic voltammograms of C_{60}-CdS films in the absence (a) and presence (b) of 1.0×10^{-5} mol·L^{-1} Hb in 0.1mol L^{-1} PBS. Inset: cyclic voltammogram of CdS film in 1.0×10^{-5}mol·L^{-1} Hb in 0.1mol l^{-1} PBS, Scan rate: 0.03V·s^{-1}

vide an electron-rich environment on the CdS surfaces, for nucleophilic addition to the electro-deficient C_{60} to form a covalent bond. This leads to uniform dispersion of C_{60} on CdS surfaces. Furthermore, it is found that the resulting C_{60}-CdS composite film was stable. Although more than 50 complete CV cycles were performed, no significant change in cathodic and anodic peak potentials and currents was observed (as shown in figure 1).

The stable C_{60}-CdS composite film promises the potential application as new material. To explore its biological application, the electrochemical properties in an aqueous solution were investigated. When the potential was scanned between -0.25 and -0.65 V, the C_{60}-CdS composite film showed a couple of reduction/reoxidetion peaks positioned at E_{pc}=-0.443V, and E_{pa}=-0.383V in 0.1M phosphate buffer solution(PBS) in figure 4 (curve a), while CdS NPs film showed no peaks under the same condition(curve b). Namely, the composite film exhibits quasi-reversible electron-transfer reaction corresponding to C_{60}/C_{60}^-. The CVs for the C_{60}-CdS composite films at different scan rate in 0.1M PBS are shown in figure S3 in supporting Information. Furthermore, the anodic currents for redox couples have a linear relationship with the scan rate in the range of 0.01-0.05Vs^{-1} (see inset of figure S3), as expected to a thin-layer electrochemical behavior and a typical surface-controlled quasi-reversible process.

We have reported that C_{60} in C_{60}-MWCNT (Multiple-walled carbon nanotube) composite films could be as an excellent electron-transfer mediator for the heterogeneous electron transfer of Hb[33]. As a result, the voltammetric behavior of Hb at C_{60}-CdS film was also investigated. CVs obtained with C_{60}-CdS composite films between -0.3 and -0.6V in 0.1MPBS with (curve α) and without(curve b) added Hb are shown in figure 5. The C_{60}-CdS composite film showed an increase in the height of the reduction current and the oxidation current after added Hb and a small peak-potential shift towards positive direction was also observed(cvrve b). In contrast, CVs for the CdS film on GC electrode in 0.1 M PBS with added Hb shows no any redox peaks(see inset in figure 5)[34]. These results indicate that the C_{60}-CdS composite film should have catalytic effect with Hb. Furthermore, the catalytic reduction peak current for C_{60} was linearly proportional to the concentrations of Hb in the range from 2.0×10^{-6} mol·L^{-1} to 1.0×10^{-5}mol·L^{-1}, the linear regression equation is expressed as I/μA=0.355+0.627c/(10^{-5}mol L^{-1}) (R=0.9993, N=5). These are characteristic of catalytic reduction of Hb by C_{60} in the films, indicating that the C_{60}-CdS composite films are capable of mediating the electron-transfer rate of Hb and the embedded C_{60} is an electron-transfer mediator. Then a possible mechanism for the electrocatalytic reaction was proposed as follows:

$$C_{60}\text{- CdS}+e^- \rightleftharpoons C_{60}^-\text{- CdS}$$

$$\text{HbFe(III) + C60}^-\text{ -CdS C60-CdS+Hbfe(II)}$$

$$\text{HbFe(II)} \rightleftharpoons \text{HbFe(III)}+e^-$$

where C_{60} in the C_{60}-CdS composite film on GC electrode get electron firstly, then, as a mediator, C_{60}^--CdS transfer the electron to HbFe(III) to form C_{60}-CdS and HbFe(II), respectively, followed by oxidizing HbFe(II) to HbFe(III), at reversible potential scan.

To attest that Hb molecules have penetrated into the C_{60}-CdS composite films, the morphology of a C_{60}-CdS/Hb composite film was characterized by SEM (see

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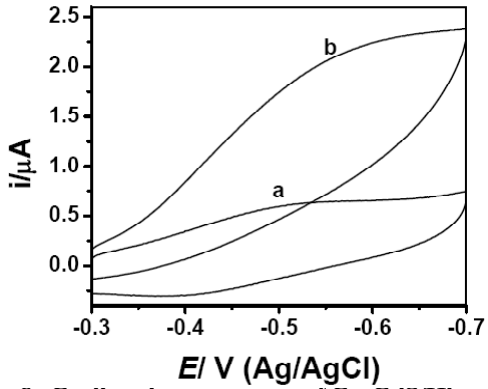

Figure 6 : Cyclic voltammograms of C_{60}-CdS/Hb composite film in the absence (a) and presence (b) of 15μm H_2O_2 in 0.1mol·L^{-1} PBS, Scan rate: 0.05V·s^{-1}

igure S4 in Supporting Information); The SEM shows some snowflake crystals in comparison with the morphology of C_{60}-CdS composite film (figure 3), suggesting that Hb molecules exist in the C_{60}-CdS films .

Further studies reveal that Hb also exhibits catalytic activity towards H_2O_2 after its interacting with C_{60}-CdS composite film. In blank PBS, the C_{60}-CdS/Hb film gave a pair of quasi-reversible anodic and cathodic waves (see figure 6a). When 15μMH_2O_2 was added into the solution, the cathodic peak current increased significantly and the oxidation peak current decreased (Figure 6b). The increase of cathodic peak current and a small peak-potential shift towards negative direction with the increase of H_2O_2 concentration were observed. These phenomena also indicated that Hb molecules exist in the C_{60}-CdS films and the C_{60}-CdS/Hb composite film modified electrode might be developed as a H_2O_2 sensor in the future.

SUMMARY AND CONCLUSION

In summary, uniform C_{60}-CdS composite films have been prepared by dispersing C_{60} on CdS NPs through CV scans in the acetonitrile solution of 0.1M $TBAPF_6$. The composite films present quasi-reversible redox behavior which is very different from the pristine C_{60} film. On the basis of the spectroscopic and TEM results obtained, it is presumed that these novel properties come from the interaction of C_{60} with the CdS NPs in a uniform fashion. To explore the biological activities of the C_{60}-CdS composites, the electrocatalysis of Hb at C_{60}-CdS composite film was also studied, which indicate that the embedded C_{60} is an electron-transfer mediator. Meanwhile, the C_{60}-CdS/Hb composite film can exhibit nice catalytic activity towards H_2O_2. The composite film might be developed as a H_2O_2 sensor in the future. Research is continuing in our laboratories on further applications of C_{60} composite films for developing biosensors.

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