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**Photocatalysis-Induced Renewable Field-Effect Transistor
for Protein Detection**

*Changliang Zhang¹, Jia-Quan Xu², Yu-Tao Li¹, Le Huang³, Dai-Wen Pang², Yong
Ning¹, Wei-Hua Huang^{2,*}, Zhiyong Zhang^{3,*}, Guo-Jun Zhang^{1,*}*

¹School of Laboratory Medicine, Hubei University of Chinese Medicine, 1 Huangjia
Lake West Road, Wuhan 430065, P.R. China

²Key Laboratory of Analytical Chemistry for Biology and Medicine (Ministry of Edu
cation), College of Chemistry and Molecular Sciences, Wuhan University,
Wuhan 430072, P.R.China

³Key Laboratory for the Physics and Chemistry of Nanodevices, Department of
Electronics, Peking University, No.5 Yiheyuan Road Haidian District, Beijing 100871,
P.R. China

*Corresponding author: Tel: +86-27-68890259, Fax: +86-27-68890259

E-mail: zhanggj@hbtcu.edu.cn; whhuang@whu.edu.cn; zyzhang@pku.edu.cn

Abstract

Field-effect transistor (FET) biosensor has attracted extensive attentions, due to its unique features in detecting various biomolecules with high sensitivity and selectivity. However, currently used FET biosensors obtaining from expensive and elaborate micro/nano-fabrication are always disposable because the analyte cannot be efficiently removed after detection. In this work, we established a photocatalysis-induced renewable graphene-FET (G-FET) biosensor for protein detection, by layer-to-layer assembling reduced graphene oxide (RGO) and RGO-encapsulated TiO_2 composites to form a sandwiching $\text{RGO}@\text{TiO}_2$ structure on a pre-fabricated FET sensor surface. After immobilization of anti-D-Dimer on the graphene surface, sensitive detection of D-Dimer was achieved with the detection limits of 10 pg/mL in PBS and 100 pg/mL in serum, respectively. Notably, renewal of the FET biosensor for recycling measurements was significantly realized by photocatalytically cleaning the substances on the graphene surface. This work demonstrates for the first time the development and application of photocatalytically renewable G-FET biosensor, paving a new way for G-FET sensor towards a plethora of diverse applications.

Keywords: Graphene field effect transistor; Photocatalytic cleaning; Renewable biosensor; Protein detection

Introduction

During the past several decades, nanomaterials-based field-effect transistor (FET) biosensors, such as Si nanowire,¹⁻⁴ carbon nanotube⁵⁻⁷ and graphene,⁸⁻¹² have attracted extensive attentions, due to their potential applications in detecting various biomolecules with high sensitivity and selectivity. A successful FET biosensor significantly depends on the fabrication of a chip composed of nanoscale nanomaterials as sensing channels. At this stage, the fabrication mainly has two approaches, top-down and bottom-up, respectively. Both of the methods, however, have to involve expensive and elaborate micro- or nano-fabrication tools, resulting in a high cost. However, the currently fabricated FET biosensors usually suffer from one time use, since the analyte adsorbed or bound on the biosensor cannot be removed efficiently after detection. Therefore, renewable nanoscale FET biosensors capable of reducing the use-cost are highly desirable.¹³ In addition, for practical use, the regeneration of the FET chip shows more advantageous over the regeneration based on molecular association and dissociation as non-specific adsorption in real samples is inevitable.

Inspired by the photodegradation of contaminate with photocatalyst in environment area, nanophotocatalyst could possibly be used to construct a renewable FET sensor regenerated by UV light after detection. As an important semiconductor photocatalyst, titanium dioxide (TiO₂) has been widely used to decompose water, manufacture hydrogen and degrade the organic molecule,¹⁴⁻¹⁸ due to its low cost, high chemical stability, non-toxicity, exceptional optoelectronic property, strong oxidizing

power, resistance to photocorrosion and good performance of photosynthesis.¹⁹⁻²¹ Nevertheless, TiO_2 is not a suitable sensing material for FET sensor due to its low electron mobility. Thus, preparation of a TiO_2 composite with excellent FET sensing material is conceivably a significant strategy to construct the renewable FET sensor. Comparing with other nanomaterials, graphene not only has better electrical conductivity and higher electron mobility, but also owns unexceptionable mechanical flexibility, large surface area, and higher stability.^{22, 23} These unique features promote rapid emergence of graphene field-effect transistor (G-FET) biosensors.²⁴⁻²⁹ To this end, graphene is an excellent candidate to combine with TiO_2 for construction of a high-performance and renewable G-FET sensor. Very recently, Huang's group reported a photocatalytically renewable micro-electrochemical sensor for real-time monitoring of cells by employing the sandwiching $\text{RGO}@\text{TiO}_2$ structure.³⁰

Although it seems that other ways, such as UV-ozone and plasma, are able to clean the molecules on the surface of graphene, the performance of graphene between the source and drain electrodes can severely be degraded by these two tools.^{31, 32} In this paper, we combine excellent electrical conductivity and high electron mobility of graphene with superior photocatalysis of TiO_2 to prepare the renewable FET biosensor for highly sensitive detection of D-Dimer, an important biomarker for the diagnosis of venous thromboembolism.³³ We fabricate the G-FET biosensor through a simple method by assembling RGO and RGO/TiO_2 nanocomposites to form a sandwich structure on the channel between the source and drain electrodes. The immobilization of anti-D-Dimer on the graphene surface enables detection of

D-Dimer with high specificity and sensitivity. Notably, the encapsulated TiO_2 make the FET biosensor reusable for detection of other proteins after irradiation of the chip with UV-light to photocatalytically clean the substances on the graphene surface. The developed method has the following merits: 1) Cost-effective. The currently fabricated graphene field-effect transistor biosensors usually suffer from one time use, leading to a high cost of applications. A renewable graphene FET is carried out by the presented approach; 2) Anti-fouling. It is still a great challenge to completely and efficiently remove the contaminant from device surface without changing the surface structure and electrical performance; 3) Most importantly, the electrical properties are retained after photocatalysis by using the fabricated graphene FET. As far as we know, this is the first report to fabricate the renewable G-FET biosensor by combining the features of label-free immune recognition with photocatalysis, representing an important tendency of FET toward practical application in disease diagnostics.

EXPERIMENTAL SECTION

Reagents and Materials. 1-Pyrenebutanoic acid succinimidyl ester (PASE) and ethanolamine (EA) were purchased from Sigma-Aldrich. KMnO_4 , NaCl , 98% hydrazine, concentrated H_2SO_4 , 35 wt % H_2O_2 , 3-aminopropyltrimethoxysilane (APTMS) and other chemicals were purchased from Generay Biotech Co. Ltd. (Shanghai, China). Graphite powder (99.99995%, 325 mesh) was purchased from Alfa Aesar Co. Ltd. (Tianjin, China). TiO_2 was purchased from Evonik Degussa (Germany). Human serum albumin (HSA), anti-BSA and BSA (Bull Serum Albumin),

anti-D-Dimer and D-Dimer were purchased from Thalys Co. Ltd. (Wuhan, China). BNP (brain natriuretic peptide) and anti-BNP were purchased from Abcam Co. Ltd. (Shanghai, China).

Synthesis of RGO. Firstly, graphene oxide (GO) was manufactured from graphite power by a modified Hummers method.³⁴ Afterwards, the GO was reduced by a method of chemical reduction.³⁵ Briefly, 15 mg of GO was added into 15 mL of 98% hydrazine, and the mixture was sonicated for 10 min, and subsequently placed for one week.²⁹ Ultimately, the RGO could be obtained and it would not aggregate after a few months.

Synthesis of RGO/TiO₂ nanoparticles composites. After GO was prepared by the above-mentioned method, the RGO/TiO₂ nanocomposites were produced by the following processes. Concretely, 0.1 g of TiO₂ nanoparticles were dispersed into 20 mL of ethanol. Subsequently, 1 mL of APTMS was added, and refluxed at 80 °C for 4 h. Then, 5 mg of APTMS-treated TiO₂ nanoparticles were added into 0.05 mg/mL GO suspension for 10 min, and the produced GO/TiO₂ were separated from GO suspension by centrifugation.³⁶ For reducing the GO/TiO₂, 1 mg of GO/TiO₂ nanocomposites were added into 1 mL of 98% hydrazine and the mixture was sonicated for 10 min. A week later, the RGO/TiO₂ nanocomposites were obtained.

Fabrication of RGO/TiO₂/RGO FET devices. The FET chips (6×4.5 mm) used in our study was fabricated by using a standard semiconductor technology, as described early.²¹ The patterned Au source and drain electrodes (50 nm thick) with 4 μm gaps were fabricated on SiO₂ (285 nm)/Si substrate through the traditional

macro-nano processing technologies including electron-beam evaporation and photolithography. The synthetic RGO (0.25 mg/mL) was drop-casted onto the gaps between the source and drain electrodes and thermally annealed at 150 °C in order to enhance the contact between the RGO and the electrodes. Soon afterwards, the Piranha solution was used to make the few-layer RGO in hyper acoustic environment for 30 s. At last, the RGO/TiO₂ nanocomposites suspension was further drop-casted on the RGO FET devices and heated at 80 °C in a vacuum oven for 1 h. The RGO/TiO₂ FET devices were accomplished after rinsed three times using the ultrapure water.

Immobilization of anti-D-Dimer on the FET devices. Firstly, 10 mM PASE in N, N-Dimethylformamide (DMF) was dropped on the graphene surface for 2 h at room temperature and redundant PASE solution was washed by pure DMF and water, successively. Secondly, the device was incubated with 20 µL of 100 µg/mL anti-D-Dimer in 1×PBS at PH 7.4 for 2 h. The un-bound antibody was removed by washing the chip with PBS for 3 times. The anti- D-Dimer-immobilized FET device was then treated with 100 mM EA for 1 h to prevent the non-specific adsorption of biomolecules.

Immunoreaction. The D-Dimer was prepared with different concentrations (10 pg/mL, 100 pg/mL, 1 ng/mL, 10 ng/mL and 100 ng/mL) by dissolving it in 1×PBS. Soon afterwards, the D-Dimer solution was dropped on the sensing surface and incubated for 10 min, where the anti-D-Dimer was immobilized. Then the chip was rinsed by 1×PBS and water, respectively, to wash off the un-bound D-Dimer and

dried by N₂.

Photocatalysis. After binding of the D-Dimer protein with the immobilized anti-D-Dimer, the chip was immersed in the pure water under a UV environment for 90 min for the purpose of photocatalysis. Then, the chip was got out of the water and washed by the DI water and dried by nitrogen. Subsequently, the chip was placed in the vacuum chamber overnight.

Measurements. The measurement system consisting of Keithley 4200-SCS and EverBeing BD (probe station) was used. All the measurement regarding the electrical characterization of the FET devices was conducted with a liquid-gate at room temperature. When the different concentrations of D-Dimer from 1 pg/mL to 100 ng/mL were conjugated with the anti-D-Dimer on the chip, the electrical transfer curves including I_d - V_g (I_d is drain current, V_g is gate voltage) and I_d - V_d (V_d is drain voltage) were recorded.

Characterizations. A field emission-scanning electron microscope (FE-SEM) (Zeiss SIGMA, Germany) was used for SEM characterization of the sensor surface after RGO and the RGO/TiO₂ nanocomposites were assembled on the FET devices, respectively. Transmission electron microscopy (TEM) (JEM-2100, Japan) was used for observing the morphology of TiO₂ and RGO/TiO₂ nanocomposites. X-ray photoelectron spectroscopy (XPS) spectra of the sensing interface of the biosensors were obtained by an ESCALAB 250Xi XPS (Thermo Fisher, America) and the source gun types were Al and Ka.

RESULTS AND DISCUSSION

Working principle. The principle of the photocatalysis-induced reusable RGO@TiO₂ FET biosensor for protein detection is schematically illustrated in Figure 1. At first, the TiO₂ nanopartciles functionalized with APTMS are coated with thin layers of GO through the electrostatic interaction between the positively charged TiO₂ nanoparticles and the negatively charged GO, followed by reduction with hydrazine to form RGO/TiO₂ composites. Meanwhile, the FET chip prepared by a traditional microfabrication process is made into G-FET by drop-casting RGO²⁹ and the synthesized RGO/TiO₂ to the chip surface between the source and drain electrodes as conductive channels. After these processes, PASE is used as a linker molecule to bind antibody on the graphene surface.^{37, 38} The interaction between antibody and antigen is monitored by recording the electrical transfer curves before and after antibody-antigen reaction. Finally, the regeneration of the chip is realized by irradiating with ultraviolet ray, through which all the substances on the graphene surface are removed and the chip can be regenerated for the next run of detection.

As known, TiO₂ is wildly used as photocatalysis-induced cleaning material because it is capable of decomposing the organic molecules by active oxygen radicals generated under ultraviolet irradiation without altering the surface morphology and structure. However, TiO₂ is inappropriate to be directly used as a sensing material because it does not have good conductivity. Therefore, combination of graphene with TiO₂ not only improves the sensing performance, but also enhances the photocatalytic efficiency. In the process of protein detection, the main contaminant on the FET

sensor surface is organic molecules. Hence, the unique sandwich structure of RGO and RGO/TiO₂ nanocomposites can be employed to construct photo-induced renewable G-FET biosensor.

Characterizations. To view the sensing channels, the fabricated FET chip was imaged by an optical microscope. It was observed that the FET sensor was composed of six individual sensors connected with the corresponding electrical lines (Figure 2a). Inset image shows that the individual sensing channel was located between the source and drain electrodes, and the width of the channel was 4 μm . To prove that the RGO/TiO₂ composites were formed, TEM observation was conducted. As shown in Figure 2b, the size of the TiO₂ nanoparticles was about 30 nm (Inset) and the nanoparticles were well shaped. After formation of the RGO/TiO₂ nanocomposites, TiO₂ nanoparticles were clearly observed to be encapsulated by the sheet-shaped RGO. The TEM images indicate that the RGO/TiO₂ nanocomposites have been synthesized successfully. To make a satisfactory contact, two layers of RGO and the RGO/TiO₂ nanocomposites were assembled to the sensing channels, respectively. The FE-SEM image of the RGO FET device was illustrated in Figure 2c. It was clearly seen that the RGO sheet spanned across a pair of Au electrodes. After depositing the RGO/TiO₂ nanocomposites on the RGO surface, the sensing region was observed again by FE-SEM. As shown in Figure 2d, it was verified that the nanocomposites were deposited on the surface of RGO. The SEM images demonstrate that layer-to-layer assembly of RGO and the RGO/TiO₂ nanocomposites to the sensing channel is realized as anticipated and the F-FET device is successfully

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4 fabricated.

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6 **Electrical property of the FET device.** As known, TiO_2 nanoparticles have
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8 the property of semi-conductive materials. However, the conductivity of the
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10 RGO/ TiO_2 nanocomposites FET device was found to be bad if the RGO/ TiO_2
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12 nanocomposites were deposited onto the sensing channels (Figure S1). This is
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14 probably caused by the reason that a physical interface may occur between the
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16 RGO/ TiO_2 nanocomposites and the FET substrate, resulting in a non-ideal contact. To
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18 solve this issue, RGO was deposited prior to assembly of the RGO/ TiO_2
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20 nanocomposites because the RGO FET device has good conductive properties as
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22 described previously.²⁹ Figure 3a shows the transfer curve of the RGO@ TiO_2 FET
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24 device compared with that of the bare RGO FET device. The I_d - V_g curves of both the
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26 RGO FET and the RGO@ TiO_2 FET device revealed an obvious ambipolar
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28 characteristic, and a right-shift Dirac voltage under air environment was clearly seen.
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30 TiO_2 , as a kind of semiconducting metal oxides, is not like metal. For metal, the
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32 electrons are easily transferred to the coterminous conducting materials. While for
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34 metal oxides, the electrons are hardly moved and easily polarized, meaning that the
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36 electrons and the holes are separate.^{36, 39, 40} When the RGO/ TiO_2 nanocomposites
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38 were deposited on the surface of RGO, the I_d - V_g curve of the RGO@ TiO_2 FET device
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40 would shift to left compared to the curve of the RGO FET device. This is because the
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42 Fermi energy level of TiO_2 (-4.0 eV) is higher than that of graphene (-4.43 eV) and
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44 the electrons on the polarized surface of TiO_2 can be transferred to graphene easily.
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46 N-doping subsequently occurred on the bare RGO FET after it was deposited with the
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3 RGO/ TiO₂ nanocomposites, which is in good agreement with the previous report.³³
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6 However, the n-doped RGO@TiO₂ FET device is easily converted to a p-type
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8 semiconductor in air because the device is exposed to oxygen or water, as described
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10 earlier.^{26, 41} As shown in the Figure 3b, the I_d-V_d curves were recorded at different V_g,
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12 and I_d increased along with the rise of V_g, suggesting a p-type behavior of the
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14 RGO@TiO₂ FET device.
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19 After anti-D-Dimer was immobilized on the RGO@TiO₂ FET surface, the
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21 I_d-V_g curves of the biosensors were recorded to compare with those of the bare
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23 RGO/TiO₂/RGO FET in Figure 3c. The isoelectric point (pI) of the anti-D-Dimer is
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25 approximate 8.0, so the anti-D-Dimer is positively charged at PBS solution (PH=7.4).
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27 The contribution of an increased net carrier density to the FET device would generate
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29 an n-doping effect, and the curve would shift to right, which is in good agreement
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31 with the results of other works.^{26, 27} On the contrary, the pI of D-Dimer is about 4.0,
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33 and the D-Dimer is negatively charged in PBS solution. After interaction of D-Dimer
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35 with anti-D-Dimer, a decreased net carrier density was contributed to the FET device,
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37 leading to a p-doping effect. The curve thus shifted to left, as shown in Figure 3d. The
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39 results demonstrate that anti-D-Dimer has been immobilized on the RGO@TiO₂ FET
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41 biosensor and D-Dimer has also been bound to the immobilized anti-D-Dimer.
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49 **Specificity of the biosensor.** The specificity of the biosensor was measured by
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51 using 1×PBS, 100 ng/mL HSA, 10 ng/mL D-Dimer and 100 ng/mL D-Dimer,
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53 respectively. In this experiment, 1×PBS acted as the blank control and HSA worked
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55 as the non-specific control. When HSA solution was dropped on the chip, HSA could
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not bind with the anti-D-Dimer on the biosensor because HSA is not specific to the antibody. As shown in Figure 4a, the shift of I_d - V_g curve was hardly observed after $1\times$ PBS and HSA solutions, respectively, were incubated with the anti-D-Dimer-immobilized FET biosensor. On the contrary, the curves shifted obviously after 10 ng/mL and 100 ng/mL D-Dimer solutions, respectively, were dropped on the biosensor surface. The specificity of the biosensor was summarized in Figure 4b. The ΔV_{CNP} (the gate voltage corresponding to the minimum conductance) of $1\times$ PBS, 100 ng/mL HSA, 10 ng/mL and 100 ng/mL D-Dimer were -7mV, -11mV, -69mV and -84mV, respectively. The ΔV_{CNP} of $1\times$ PBS was the noise level. The ΔV_{CNP} of 100 ng/mL HSA was larger than that of $1\times$ PBS. This is mainly caused by some non-specific adsorption. Though the ΔV_{CNP} of 100 ng/mL HSA was larger than that of blank control, the ΔV_{CNP} of 10 ng/mL D-Dimer, having a lower concentration than 100 ng/mL HSA, obviously exceeded that of 100 ng/mL HSA. It was very clear that the ΔV_{CNP} of 100 ng/mL D-Dimer was much higher than that of 10 ng/mL D-Dimer because higher concentrations of proteins usually have more electrons contribution to the FET device. These results indicate that the RGO@TiO₂ FET biosensor has a satisfactory specificity.

Sensitivity of the biosensor. To test the sensitivity of the RGO@TiO₂ FET biosensor in PBS solution, a series of concentrations of D-Dimer solution from 1 pg/mL to 100 ng/mL were chosen, and the results were shown in Figure 5a and 5b. As shown in Figure 5a, it was seen that the V_{CNP} shifted to left after D-Dimer was bound to the immobilized anti-D-Dimer. The shifts increased along with the increased

D-Dimer concentrations, and this result is in consistent with that described by other researches.²¹ Figure 5b shows the ΔV_{CNP} (i.e. the shift of V_{CNP} after interacting with different concentrations of D-Dimer relative to V_{CNP} of anti-D-Dimer) as a function of the concentrations of the D-Dimer, which summarizes the various shift values of V_{CNP} corresponding to the different concentrations of D-Dimer. The dashed line in Figure 4b revealed the noise level (about -7 mV), which was obtained from a blank control. The (LOD) of 10 pg/ml rather than 1 pg/ml was obtained based on the signal/noise ratio ≥ 3 .

In order to further explore the performance of the biosensor for practical application, the different concentrations of D-Dimer in human serum sample were also conducted. Figure 5c shows the ΔV_{CNP} as function of different concentrations of 10 pg/mL, 100 pg/mL, 1 ng/mL, 10 ng/mL and 100 ng/mL D-Dimer in human serum sample, respectively. Similar with the above-mentioned results in Figure 5a, the transfer curves shifted to left when the increased concentrations of D-Dimer from 10 pg/mL to 100 ng/mL in the human serum samples were applied. A summary of the various shift values of V_{CNP} corresponding to the different concentrations of D-Dimer in serum was shown in Figure 5d. It was found that the LOD in serum was 100 pg/ml, which is 1 order magnitude higher than that of the FET biosensor tested in buffer solution. As known, serum is a more complex environment and abundant in proteins. These noncognate proteins commonly interfere with the detection of target proteins. Hence, the binding efficiency of D-Dimer with anti-D-Dimer in serum is lower than that in PBS, resulting in less electron contribution to the G-FET. Even so, compared

with the classical method of detecting D-Dimer, like ELISA (threshold: 500 ng/mL), the G-FET biosensor still has a high sensitivity and it would be applied in the practical applications.

Regeneration of the G-FET biosensor by Photocatalysis. To demonstrate the reusability of the G-FET biosensor via photocatalysis, the D-Dimer-bound RGO@TiO₂ FET biosensor underwent self-cleaning by UV irradiation. Afterwards, the regenerated FET chip was treated with PASE, immobilized with anti-D-Dimer, and bound with D-Dimer once more. 3 cycles of self-cleaning and protein-protein interactions were performed. The changes of the V_{CNP} including the immobilization of anti-D-Dimer, the immunobinding of D-Dimer, and the degradation of macromolecule on the FET surfaces via photocatalytic cleaning were recorded and compared. Figure 6a shows the changes of I_d - V_g curve after the FET biosensors underwent one cycle of the above-mentioned process. As described early, the V_{CNP} shifted to right after the anti-D-Dimer was immobilized, while the V_{CNP} shifted to left after D-Dimer was bound to the chip. However, the V_{CNP} shifted to left after the FET device was irradiated under UV-light. TiO₂ on the devices can catalyze the surrounding water moisture and oxygen molecules, and give birth to reactive intermediate species, e.g., $\cdot\text{OH}$, O_2^- , and H_2O_2 , which would selectively oxidize the organic molecules on the sensing surface.^{42, 43} The organic molecules in the close vicinity are finally degraded by these reactive intermediates, by which the RGO@TiO₂ FET biosensor can be self-cleaned and regenerated for the next run, as illustrated in Fig. S2. When the devices underwent the process of photocatalysis, the electrical curves shifted to

left and the V_{CNP} was retained to the initial level. In order to further illustrate the effect of photocatalysis and investigate the reversible ability of the FET biosensors, three sequential cycles of the above-mentioned experiments were conducted and the experimental results are shown in Figure 6b. The V_{CNP} was restored near the primal level after UV irradiation every time.

For the sake of proving that photocatalysis occurred as anticipated, XPS was employed to characterize the 4 different interfaces of one cycle. Figure 6c shows wide energy XPS spectra after each functionalized step including the RGO@TiO₂ surface (1), the anti-D-Dimer modified surface (2), the D-Dimer-bound surface (3), and the RGO@TiO₂ surface after photocatalysis (4). The RGO@TiO₂ surface gave rise to C1s, Ti2p, and O1s peaks, indicating that the assembly of the two-layer nanomaterials is successful. After modification with antibody and interaction of antibody-antigen, C1s, Ti2p, and O1s peaks were still visible, while N1s obviously appeared in both cases because protein has peptide backbone and contains abundant nitrogen element. After UV irradiation, the proteins were degraded by photocatalysis and the N1s peak returned to the initial level. Narrow energy spectra comparing various N1s peaks among the 4 surfaces were shown in the Fig. S3. It was clearly seen that N1s peaks became very remarkable before the UV-light irradiation. The N1s peak of the D-Dimer-bound surface was much higher than that of the anti-D-Dimer-immobilized surface, mainly because more nitrogen was contributed to the surface after interaction of antibody-antigen. These results confirm that the device can be used repeatedly through photocatalysis for multiple times.

It is of great significance to investigate the universality of the G-FET biosensor by employing one device for detection of different proteins regenerated via photocatalysis. To do so, the G-FET biosensor was first used to detect BSA after anti-BSA was immobilized on the sensing surface. After UV irradiation, the regenerated G-FET biosensor was then employed to detect D-Dimer and BNP in sequence after different antibodies including anti-D-Dimer and anti-BNP, were immobilized on the sensing surface. The changes of V_{CNP} corresponding to the three cycle processes for detecting three proteins were shown in Figure 6d. As shown, when 100 pg/mL BSA was bound to the immobilized anti-BSA, the V_{CNP} change was found to be changed from 191 mV to 150 mV. The V_{CNP} turned to be 141 mV after UV exposure. Afterwards, the same chip was regenerated to detect the same concentrations of D-Dimer and BNP (both in 100 pg/mL), respectively, by immobilizing the corresponding antibodies on the same G-FET biosensor. It was obvious that both D-Dimer and BNP have been successfully detected. Not only can the G-FET device be recycled after photocatalysis, but also it has a consistent pattern for the three protein detection in a three-time cycle. These results show that the G-FET biosensors have a capability of universality and this feature can be applied in the future to reduce the cost of the FET biosensors greatly.

It is reported that other FET biosensors can also be regenerated. For instance, Lin et al. developed a renewable silicon nanowire FET biosensor to study protein-protein interactions by taking advantage of the reversible association-dissociation between glutathione (GSH) and glutathione S-transferase

(GST).⁴⁴ In addition, we also demonstrated a renewable graphene FET biosensor capable of detecting DNA by hybridizing and denaturizing the DNA duplex.²⁹ However, these methods can't thoroughly clear all the molecules away on the sensing surface since these receptors and targets have strong binding affinity, making it difficult to completely remove the analytes from the surface of FET biosensors after detection. More severely, these methods are inappropriate to be used for detection in real samples like serum since other nonspecific proteins still remain on the surface and are thus hard to remove. This is the reason why the vast majority of the FET biosensors could be used only for one single measurement and they are difficult to be regenerated in complex circumstances. Hence, the developed method provides a unique and potential approach to detecting the analytes in clinical samples by using the same FET sensor for multiple times.

Conclusion

In conclusion, we have developed a photocatalytically renewable FET biosensor capable of detecting proteins with high sensitivity and specificity. The fabricated G-FET biosensors were composed of RGO@TiO₂ sandwich architecture by making full use of RGO's excellent conductivity, high electron mobility, fine biocompatibility, and TiO₂ nanoparticles' superior photocatalysis capability. The biosensors achieved a commendable sensitivity of D-Dimer in PBS buffer (LOD: 10 pg/mL) and serum sample (LOD: 100 pg/mL), respectively. More importantly, the developed RGO@TiO₂ FET biosensors possessed excellent reusability, making the

FET chip renewable for multiple times detection. Furthermore, the G-FET showed a universal ability of detecting different proteins by using one chip. Such a versatile strategy greatly reduces the cost of the FET chip, thereby promoting practical application of FET in biomedical measurements.

AUTHOR INFORMATION

Corresponding Author

*G.-J. Zhang. Tel: +86-27-68890259. Fax: +86-27-68890259.

E-mail: zhanggj@hbtcm.edu.cn.

Notes

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ASSOCIATED CONTENT

Supporting Information

Typical I_{ds} - V_{ds} curves of the RGO/TiO₂ FET device, the schematic illustration for principle of photocatalysis, and XPS comparison of N1s on the different sensing surfaces of the FET devices. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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Figure captions:

Figure 1. Schematic illustration of the photocatalysis-induced renewable RGO@TiO₂ FET biosensor for protein detection.

Figure 2. (a) Optical microscope photographs of the sensing channels and the corresponding contact lines of one FET chip. The sensing channels are zoomed in Inset. (b) TEM image of RGO/TiO₂ nanocomposites and TEM image of the TiO₂ nanoparticles are shown in Inset (c) FE-SEM image of a single RGO FET device. (d) FE-SEM of the RGO@TiO₂ nanocomposites fabricated on the surface of the RGO spanning across Au electrodes.

Figure 3. (a) Typical I_d - V_g curves before and after the RGO/TiO₂ nanocomposites are deposited on the RGO FET device. (b) Typical I_d - V_d curves of the RGO@TiO₂ FET device. The gate voltage (V_g) is varied from 0 to 0.4 V with an interval of 0.1 V. (c) Typical I_d - V_g curves of the anti-D-Dimer-immobilized RGO@TiO₂ FET biosensor. (d) Typical I_d - V_g curves of the D-Dimer-bound RGO@TiO₂ FET biosensor.

Figure 4. (a) Transfer curves and (b) the shift of V_{CNP} of the anti-D-Dimer-immobilized RGO@TiO₂ FET biosensor incubated with 1×PBS, 100 ng/mL HSA, 10 ng/mL D-Dimer and 100 ng/mL D-Dimer, respectively.

Figure 5. (a) Transfer curves and (b) the shift of V_{CNP} of the anti-D-Dimer-immobilized RGO@TiO₂ FET biosensor interacted with D-Dimer at a series of concentrations (1 pg/mL, 10 pg /mL, 100 pg /mL, 1 ng/mL, 10 ng/mL and 100 ng/mL) in 1×PBS solution. (c) Transfer curves and (d) the shift of V_{CNP} of the anti-D-Dimer-immobilized RGO@TiO₂ FET biosensor interacted with D-Dimer at a

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series of concentrations (10 pg/mL, 100 pg/mL, 1 ng/mL, 10 ng/mL and 100 ng/mL) in serum sample. Dashed line represents the signal/noise ratio of 3 from the blank control test.

Figure 6. (a) Transfer curves of the RGO/TiO₂/RGO FET biosensor undergoing several steps including the immobilization of anti-D-Dimer, binding with D-Dimer and UV irradiation. (b) Changes of V_{CNP} of three sequential cycles of the above-mentioned 3 steps. (c) XPS spectra (survey scan) of the different sensing surfaces of the FET devices (1: bare RGO/TiO₂/RGO, 2: immobilization of anti-D-Dimer, 3: binding with D-Dimer, and 4: UV irradiation.). (d) Changes of V_{CNP} of three proteins' detection including BSA, D-Dimer and BNP by using one G-FET device regenerated via photocatalysis.

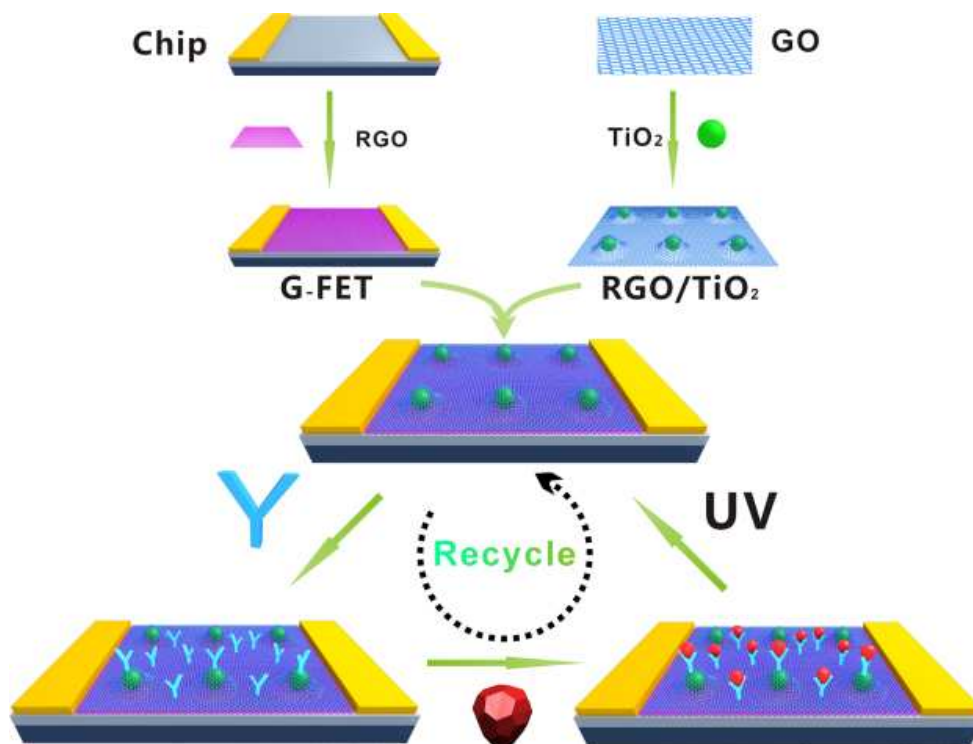


Figure 1

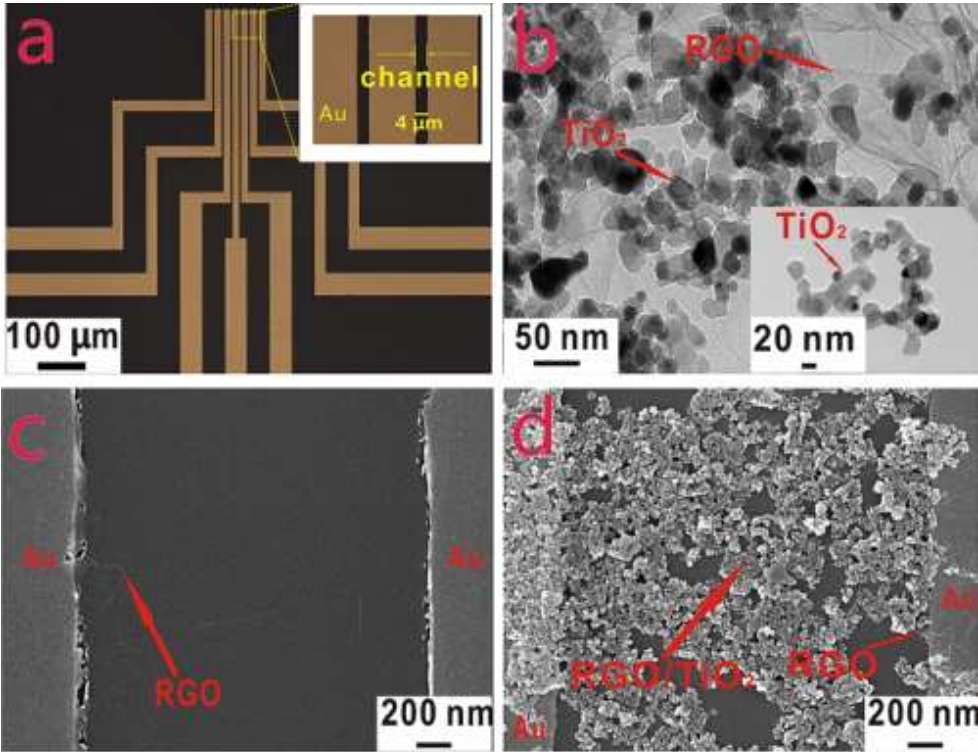


Figure 2

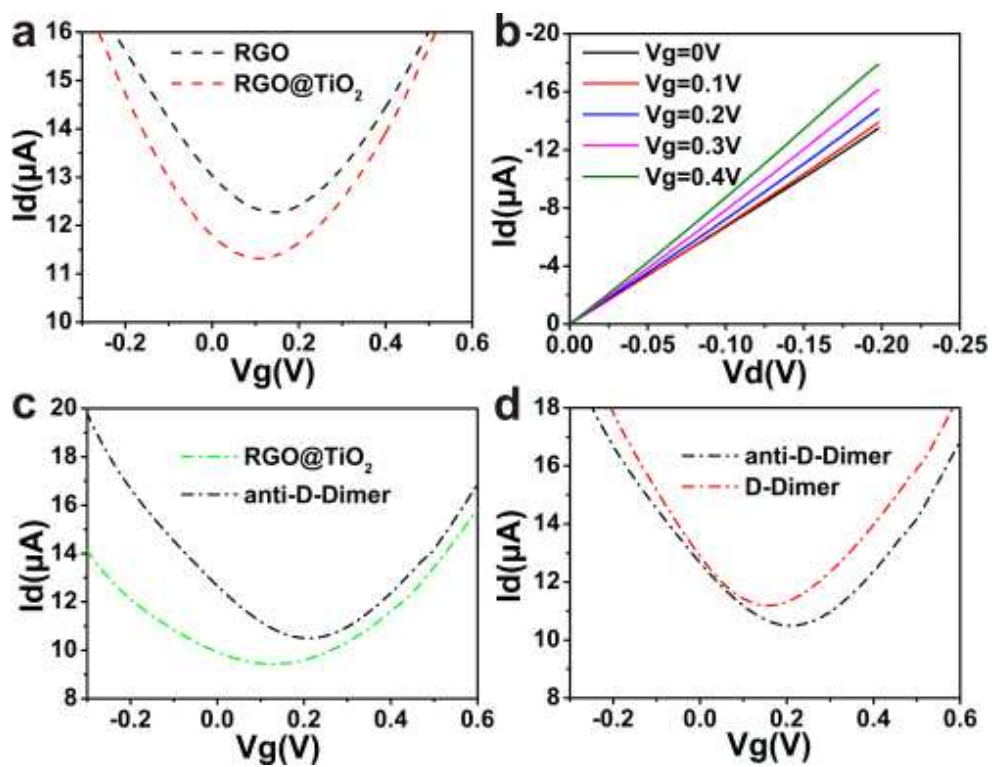


Figure 3

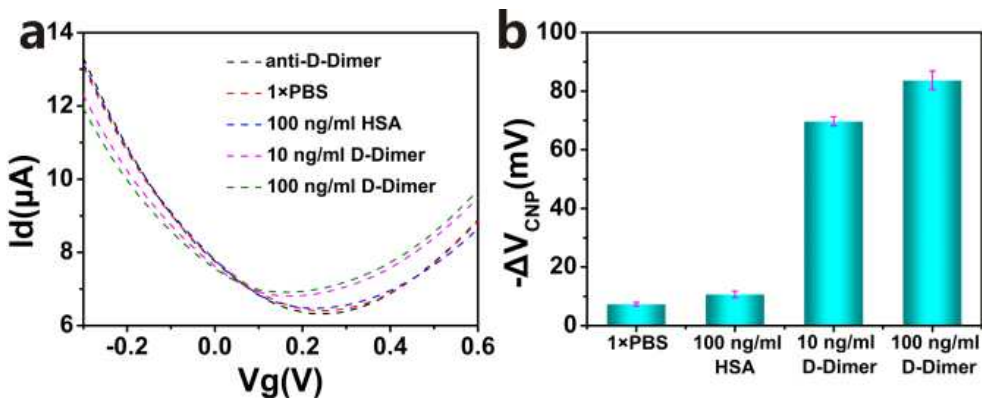


Figure 4

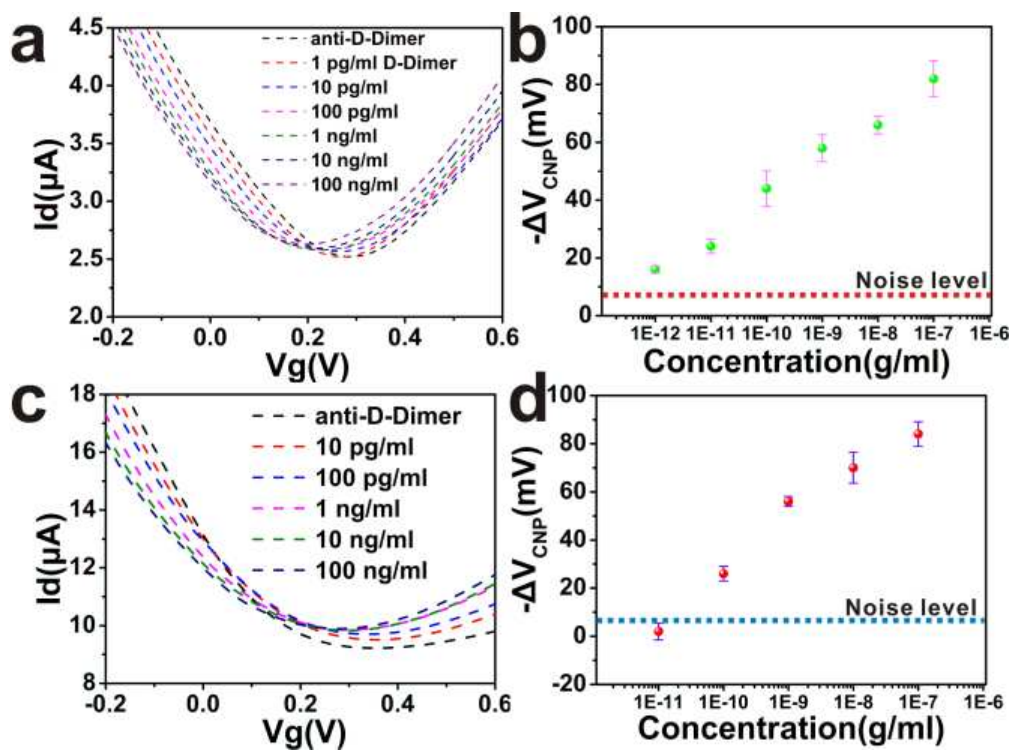


Figure 5

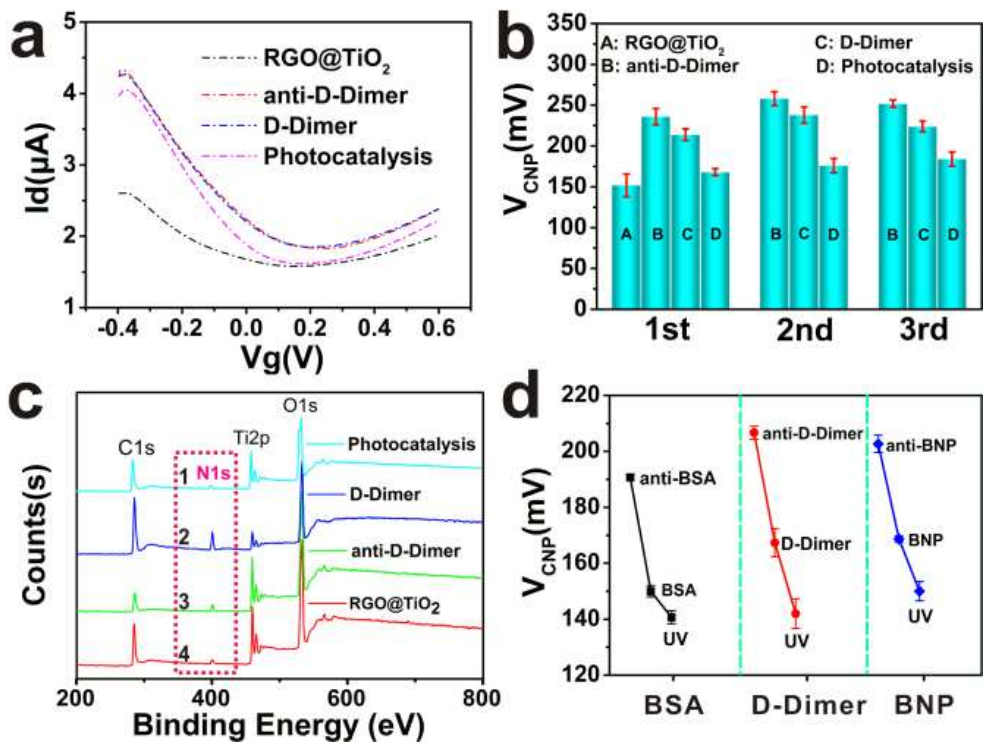


Figure 6

For TOC only

