Fabrication of Ultrasensitive Field Effect Transistor DNA Biosensors by A Directional Transfer Technique Based on CVD-Grown Graphene

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Abstract

Most of graphene field effect transistor (G-FET) biosensors are fabricated through routine process, in which graphene is transferred on Si/SiO_2 substrate and then devices are subsequently produced by micro manufacture processes. However, such a fabrication may introduce contamination to the graphene surface during the lithographic processes, resulting in interference for the following biosensing. In this work, we have developed a novel directional transfer technique to fabricate the G-FET biosensor based on chemical vapor deposition (CVD)-grown single layer graphene (SLG) and have applied this biosensor for sensitive detection of DNA. The FET device with six individual array sensors was first fabricated, and the SLG obtained from CVD-grown method was transferred to the sensor surface in a directional manner. Afterwards, peptide nucleic acid (PNA) was covalently immobilized on the graphene surface and DNA detection was realized by applying specific target DNA to the PNA-functionalized G-FET biosensor. The developed G-FET biosensor was able to detect target DNA as low as 10 fM, which is 1 order of magnitude lower than that reported in the previous work. In addition, the biosensor was capable of distinguishing the complementary DNA from the one-base mismatched DNA and the non-complementary DNA. The directional transfer technique for fabrication of the G-FET biosensor is simple and the as-constructed G-FET DNA biosensor shows ultrasensitivity and high specificity, indicating its potential applications in disease diagnostics as a point-of-care tool.

Keywords: graphene · field effect transistor biosensor · directional transfer technique · peptide nucleic acid-DNA hybridization · detection

Introduction

Graphene is a two-dimensional honeycomb lattice of carbon with sp^2 structure that owns amazing physical and chemical properties for biosensing, such as high intrinsic carrier mobility, ultrathin body, ambipolar effect, low intrinsic noise level, good biocompatibility and stability, etc.[1-3] Rapid and highly sensitive detection of DNA is quite important for disease diagnosis, environment monitoring and so on.[4-6] Conventional DNA detections including real-time PCR fall into tedious operation process and require labeling fluorescent dye. Recently, graphene-based optical and electrochemical transduction platforms have been developed,[7-8] in which fluorescence or electrochemical tags are still required. Differently, graphene field effect transistor (G-FET) biosensors hold particularly advantages for detecting nucleic acids, because they do not require fluorescent label or electrochemical tags but achieve high sensitivity, specificity and rapid measurement.[2, 9-10] So there is a significant need for rapid and cost-effective production of high quality G-FET devices.

Rapid and economical method of preparing high quality graphene is a very important process for the fabrication of graphene devices. Currently, there are three major kinds of methods for preparing graphene, which has subsequently been applied in FET biosensor fabrication. These methods include production of graphene flakes obtained by micromechanical exfoliation from graphite,[1, 11-12] production of reduced graphene oxide (RGO) from reduction of liquid exfoliation of graphite,[13-15] and production of chemical vapor deposition (CVD)-grown graphene.[16-18] In case of micromechanical exfoliation, the pristine graphene is obtained by micromechanical cleavage of graphite with tape, which demonstrates the best electronic and structural qualities. However, the approach is time-consuming and yields incontrollable graphene devices in terms of shape, size and location. Drop-casting[10] or spin coating

[19-20] technique provides a potential for assembling RGO-based FET biosensors, respectively. Nevertheless, the size and layer of the resulting RGO are not yet controllable, so it is difficult to obtain a single layer of RGO. Because all oxygen groups on the GO surface can not completely be removed during the reduction process, the mobility and conductivity of the resulting RGO are still lower than those of the CVD-grown graphene. CVD method prepares large-scale high quality monolayer graphene and is an economical and promising way. It is believed that the high-yield and high-quality CVD graphene enables the mass production of electronic sensors with high sensitivity and high reproducibility.

Many efforts have recently been made on fabrication of CVD-grown graphene FET biosensors for label-free DNA detection with high sensitivity. For example, Guo et al.[21] reported a label-free DNA FET biosensor based on CVD-grown graphene, achieving a detection limit down to 1 nM. Dong et al.[22] reported a liquid gated FET biosensor based on few layer CVD-grown graphene for label-free DNA detection, in which the detection limit for complementary DNA could be as low as 10 pM. Chen et al.[23] developed a gold transfer technique to fabricate CVD-grown monolayer graphene FET biosensor, in which DNA detection could be down to 1 pM. In that work, although the fabrication process can be rapid due to usage of silver paste as electrodes, the contact between graphene and silver paste, including the adhesion strength and contact resistant, is yet uncontrollable. Recently, Xu et al.[24] adopted an amplification strategy combined with CVD-grown graphene to realize 100 fM DNA detection. However, it is well-known that the CVD-grown graphene FET devices are usually fabricated by the conventional techniques.[25-26] The flow diagram of the conventional G-FET fabrication process is shown in Scheme S1. As seen, the fabrication process of each single chip is complex. For one run, only one G-FET chip

can be fabricated by the whole fabrication process. As a result, the manufacturing flow suffers from time-consuming and costly fabrication. On the other hand, the cleanliness of graphene surface is extremely important, because residual surface contamination on the graphene surface affects functionalization of biological species and intrinsic properties of graphene.[2, 27] Very recently, Lerner et al.[28] proposed a transfer method of patterning a graphene nanoribbon for scalable production of nanosensors in order to detect proteins with a high sensitivity. However, this method still suffers from complex operation processes, and requirements of professional technical personnel.

In this work, we have developed an ultrasensitive CVD-grown single layer graphene (SLG) FET DNA biosensor by a novel directional transfer technique, in which the CVD-grown monolayer graphene is transferred onto the pre-fabricated sensor array surface in a directional manner (Scheme 1). A 4-inch wafer containing 110 devices is fabricated at one time by the micro fabrication process, after which the G-FET device is obtained by the directional transfer technique. To our knowledge, we have demonstrated the first example of the directional transfer technique to fabricate peptide nucleid acid (PNA)-functionalized graphene FET biosensor for DNA detection. Compared to the conventional techniques, the directional transfer technique presents the outstanding advantages as follows: (1) High yield, 160 G-FET devices are fabricated and 144 of them work normally (Yield=90%) in this work, (2) Simple operation and reduced time, one G-FET chip can be fabricated for the whole one run in the conventional fabrication process vs. graphene can be transferred to the pre-fabricated wafer-scale chips for fabrication of the G-FET chips by the directional transfer technique, (3) Cost-effective process, e.g. a 1×1 cm^2 SLG/copper could be used for fabrication of about 100 devices, (4) Clean surface of graphene, polymethyl

methacrylate (PMMA) is only used at a time, while three-time usage of PMMA is required for the conventional approach.

Experimental Methods

Materials

The 22-mer PNA probe sequence was synthesized by Bio-Synthesis, Inc. (Lewisville, Texas). The 22-mer DNAs were purchased from Takara Biotechnology Co. Ltd. (Dalian, China), and were purified by HPLC. All PNAs and DNAs were dissolved in 1×PBS buffer solution. The sequence of the PNA probe is N-AACCACACAACCTACTACCTCA-C. The hybridization sequences of DNAs in use were as follows: 5'-TGAGGTAGTAGGTTGTGTGGTT-3' (complementary, cDNA), 5'-TGAGGTAGTAGGATGTGTGGTT-3' (one-base mismatched), 5'-ATGCATGCATGCATGCATGCAA-3' (non-complementary, non-DNA). 1-Pyrenebutanoic acid succinimidyl ester (PASE) and ethanolamine (EA) were obtained from Sigma Aldrich. Ultrapure water obtained from a Millipore water purification system (18.2 MΩ resistivity, Milli-Q Direct 8) was used throughout the research.

Growth of graphene on the copper foil surface by chemical vapor deposition

Graphene films were grown by atmospheric pressure CVD on 25 μm copper foils (Alfa Asear, purity 99.8%), and the grown method has been described anywhere.[29] Copper foil was placed in tube furnace and heated at 1070 □ under a flow of 300 sccm hydrogen for 50 min to remove the native oxide of copper, and then a mixture of 5 sccm methane and 700 sccm hydrogen were introduced into the quartz tube to grow graphene for 30 min. Subsequently, the quartz tube was pulled out from the heating region of the furnace so that the copper foil was rapidly cooled for monolayer graphene formation.

Fabrication of G-FETs by the directional transfer technique

In order to make it convenient for transferring graphene onto the biosensor array, the biosensor array was fabricated on the edge of the chip surface. By the conventional micro fabrication technology, the source and drain electrodes were well fabricated on 285 nm SiO_2/Si substrate by photolithography followed by electron beam evaporation deposition of 5 nm/45 nm Ti/Au, To strengthen adhesion between Au and the Si wafer, a 5 nm thickness Ti layer was used.

In order to transfer graphene from copper substrates to the FET chip substrates, the PMMA solution was spin-coated on SLG/copper foils at 4000 rpm for 30 s and dried in air. The 1×1 cm^2 PMMA/SLG/copper was cut into about 100 small ones by scissor (approximately 1×1 mm^2 for one cut graphene sheet). Copper substrate was etched away by Cu etchant (a mixture of copper sulfate, hydrochloric acid and DI water) for about 15 min. It's interesting that the directional transfer technique process required a shorter Cu etching time (ca. 15 min) than the conventional transfer technique did (ca. 2 h). That's mainly benefitted from the small size of the cut PMMA/SLG/Copper. Isopropyl alcohol (IPA)/DI water (1:10) was used to wash the chip three times to remove the etchant residue.[30-31] The PMMA/SLG was directionally transferred onto the top of the biosensor array surface of the pre-fabricated electrodes devices by aligning the small size of PMMA/SLG with the array surface in solution. Compared to the conventional method, the alignment could be realized in solution, simplifying the fabrication process. Prior to the transfer process, the FET channel surface was functionalized with 3-aminopropyltriethoxysilane (APTES),[29, 32] which is extremely important to make the transferred graphene completely stable on the SiO_2 surface due to the strong interaction between amino groups and graphene. Then, The stack PMMA/SLG/SiO_2 was annealed at 220 °C for about 5 min on a hot plate,[33]

which was performed to minimize the wrinkles that inevitably form during transfer process, and enhance the contact between the SLG and SiO_2/Si substrate. Finally, in order to make graphene surface as clean as possible, the PMMA layer was removed using hot acetone (60 °C) overnight, and then the SLG/SiO_2/Si substrate was annealed in vacuum at 200 °C for 2 h to decompose the polymers.[23, 34] Since the PMMA polymer was used only for one time, the residual polymer on the graphene surface was easily removed.

Functionalization of G-FET biosensor

PASE was used as a cross linker to immobilize PNA on the graphene surface. Firstly, the chip was immersed in PASE in dimethylsulfoxide (DMSO) at room temperature for 1 h, followed by rinsed with DMSO, ethanol and DI water in sequence. Subsequently, 10 μM PNA was dropped onto the chip surface and incubated at room temperature for 2 h. In order to remove un-reacted probe, 1×PBS containing 0.2% SDS, 1×PBS, and DI water were successively used. PASE is only able to crosslink PNA to the graphene, because the π-π stacking interaction between PASE and graphene surface occurs, while there is no π-π stacking interaction between the chip surface (SiO_2 interface) and PASE. Finally, the chip was treated with 100 mM ethanolamine (EA) solution (pH 9.0) for 1 h to avoid possible nonspecific adsorption on the graphene surface and then rinsed by DI water.

PNA-DNA Hybridization

The complementary probe DNA was added onto the top of the biosensor array surface after PNA functionalization, and the incubation was allowed for 1 h to ensure the sufficient PNA-DNA hybridization reaction. Subsequently, the chip was rinsed with 1×PBS containing 0.2% SDS, 1×PBS, and DI successively and then dried with

N_2. The same operation process was applied for the one-base mismatched and the noncomplementary DNA sequences for hybridization with PNA probe, respectively.

Electrical measurements

All the electrical measurements were obtained by keithley 4200 semiconductor characterization system combined with a probe station (EverBeing BD-6). For transfer curve measurements of the FET biosensor, the solution-gate experiments were carried out by using silver wire reference electrodes as a gate electrode immersed in buffer solution and a constant bias V_{ds}=0.1 V (the drain-source voltage) was applied. Output curves were measured at assigned V_g (the gate voltage) value.

Characterizations of monolayer graphene

The transferred SLG was characterized using both optical microscopy (ZEISS Axio scope A1) and Raman microscopy (LabRAM HR 800). Raman spectrum of the samples was taken with the Raman microscopy system equipped with 633 nm laser. JSM-6510 LV scanning electron microscopy (SEM) was used for SEM characterization of the device. The X-ray photoelectron spectroscopy was performed by an ESCALAB 250 Xi XPS system (Thermo Fisher Scientific) equipped with monochromatic Al K α source radiation.

Results and Discussion

Figure 1(a, b) shows the optical images of the FET devices fabricated on a 4-inch wafer containing 110 devices and an individual FET device, respectively. Figure 1(c) shows the SEM images of the 6 sensor arrays of the individual FET device. The size of the whole device was 6×4.5 mm, and the width of the sensing channel was 4 μm between the two electrodes.

Figure 2(a) shows the optical images of the transferred graphene between two electrodes, indicating that monolayer graphene is covered on the whole channel.[35] The graphene after directional transfer was further characterized by Raman spectrum microscopy. Raman spectrum for the as-transferred graphene sample showed high quality monolayer graphene, for it was found that a high I_{2D}/I_G ratio was 2.43 (Figure 2(b)).[18, 36] The results demonstrate that the graphene transferred to the fabricated FET sensor chip is a monolayer.

It is notable that there was no effect on the measurement although the contact lines of the chips were not passivated. As seen from Figure S3, it was observed that drain current was three orders of magnitude higher than gate leakage. So the effect is negligible even if the contact lines are not passivated. Electrical measurement was obtained before and after annealing treatment. As seen in Figure 3(a), the right shift of the charge neutrality point (V_{CNP}) indicated that the carrier density of graphene increased (p-doped) after thermal annealing.[23] From the I_{ds}-V_g curve in Figure 3(b), the FET with the as-transferred graphene showed a Dirac point voltage of 0.4 V and a very small hysteresis of 2 mV, pointing to a fact that the fabrication process is clean and harmless to graphene. The p-type doping is attributed to graphene exposed to ambient environment and subsequently thermal annealing.[23, 37-38] The output characteristics in Figure S4 showed that the drain current decreased when the gate voltage increased from -0.3 V to 0.3 V, again indicating the device is p-type doped. The results are consistent with the previous results and the drain response is sensitive to the gate voltage change.

As known, it is important to wash PMMA away and make the transferred graphene clean. To do so, PMMA was washed away with hot acetone and annealing in the work. There are three evidences supporting the clean graphene surface. Firstly,

Raman spectrum is a potential method for tracing the residual PMMA of the graphene surface. Figure S1 showed a typical Raman spectrum of PMMA residue on graphene. Two peaks at 1450 and 1530 cm^{-1}, respectively, could be attributed to the effect of PMMA residue on graphene surface (Figure S1). On the contrary, the two additional peaks disappeared after PMMA was washed away, as shown in Figure 2(b), indicating that the graphene surface is clean. Secondly, transfer characteristics curve (Figure 3(b)) showed a very small hysteresis of 2 mV, further proving that graphene surface is clean. Last but not the least, X-ray photoelectron spectroscopy (XPS) for tracing PMMA residue on graphene was further conducted. Figure S2 showed the evolution of C1s core-level spectra of PMMA residue on graphene and clean graphene, respectively. The black line represents the raw spectrum. In order to fit the C1s XPS spectra, the sp^2 component of C-C bonding was obtained by using asymmetric Gaussian-Lorentzian formula. The main peak (red line) was attributed to the graphene sp^2 components of the post-transfer C1s spectrum. Compared to XPS spectra of clean graphene (Figure S2(b)), both C-O peak (magenta line) and C=O peak (purple line) in the spectra appeared (Figure S2(a)), corresponding to the binding energy of species of PMMA residue on graphene in addition to the main peak. So it's clear that PMMA is completely removed from the graphene surface by hot acetone and annealing. Based on the above-mentioned evidences, the G-FET biosensors fabricated by the directional transfer technique are clean.

The principle of the SLG FET biosensor for DNA detection is same as that reported previously.[10] Shortly, the G-FET arrays are functionalized with PNA by a linker molecule PASE through π-π stacking interaction between the pyrene group and the graphene surface. The specific target DNA is then hybridized with the immobilized PNA. For electrical measurement, 0.01×PBS buffer solution is dropped

to the biosensor and a silver wire as liquid gate is immersed in buffer solution to offer gate voltage. The negative shift in V_{CNP} of graphene upon PNA/DNA hybridization has been observed and the electron-rich nucleobases in DNA strand imposed n-doping to graphene is considered as detection mechanism.[2, 39-40] The specific detection of target DNA using the CVD-grown SLG FET biosensor was investigated by applying the various target DNA concentrations to the PNA-functionalized FET biosensor. To exclude the possibility of nonspecific adsorption and false signals, a series of control experiments were carried out. Firstly, it's necessary to measure the influence of 1×PBS on the biosensing because target DNA was dissolved in 1×PBS. The transfer characteristics curves were found to have negligible change before and after 1×PBS buffer solution was incubated with the PNA-functionalized FET biosensor (Figure 4(a)). Secondly, it is known from the previous reports that graphene oxide category-based biosensors usually present obvious nonspecific adsorption.[41] In this research, as observed in Figure 4(b), transfer curves exhibited rare change after the FET biosensor without PNA modification was incubated with 10 fM cDNA, 1 pM cDNA, and 100 pM cDNA, respectively. Compared to RGO, CVD-grown SLG graphene does not contain functional groups, which is inert to the biomolecules. Figure 4(c) shows transfer characteristics curves of the PNA-immobilized CVD-grown SLG FET biosensors incubated with 10 fM non-DNA, 1 pM non-DNA, 100 pM non-DNA, respectively. As illustrated, the shift of V_{CNP} was also negligible, indicating there is no hybridization between PNA and non-DNA. Figure 4(d) shows that the shift of V_{CNP} of the PNA-immobilized SLG FET biosensors incubated with 10 fM one-base mismatched DNA, 1 pM one-base mismatched DNA, 100 pM one-base mismatched DNA, respectively. At very low concentrations of one-base mismatched DNA, the shift of V_{CNP} was hardly observed. However, the shift of V_{CNP} at 100 pM was found to

be obvious and much larger than that for non-DNA. For one-base mismatch, the duplex between the one-base mismatched DNA and the probe PNA can still be formed, while the duplex is unstable and most of duplexs may be unzipped during the washing steps. Therefore, compared to complementary and noncomplementary DNA, the signal is either lower or larger, respectively.

Next, to investigate the sensitivity of the CVD-grown SLG FET biosensor, various concentrations of target DNA were sequentially incubated with the PNA-immobilized SLG FET biosensors. As shown in Figure 5(a), the V_{CNP} of the device shifted toward negative gate voltage while the concentrations of cDNA increased from 1 fM to 100 pM. The larger the shift, the higher the concentration. This result is in good agreement with that reported by other literatures.[2, 23, 39] Figure 5(b) summarizes the V_{CNP} shift versus cDNA, non-DNA, one-base mismatched DNA, blank control PBS buffer, respectively. These data confirm that the developed FET biosensors can well distinguish complementary DNA from one-base mismatched DNA and non-complementary DNA, and they can certainly be used for SNP (single nucleotide polymorphism) detection. Therefore, the CVD-grown SLG FET biosensors present high specificity for DNA detection. The detection limit of the FET biosensor was as low as 10 fM for DNA detection based on the signals that surpass the background by 3-fold. The detection limit is 2 orders of magnitude lower than that reported by Chen et al.[23], 1 order of magnitude lower than that reported by Cai et al.[10] The high sensitivity can be attributed to usage of monolayer graphene as channel material as well as PNA as a probe. As mentioned early, since PNA has a non-ionic backbone,[42-43] electrostatic repulsion between PNA-DNA hybridized strands is reduced, thereby lowering the background signal and improving detection sensitivity.[44] On the other hand, the quality of graphene basically affects the

sensitivity of the FET biosensor. The graphene transferred to the FET device is a monolayer, possessing a high surface-to-volume ratio and thereby a high detection sensitivity. The carrier mobility is an important parameter for determining the graphene quality although there is no direct relation between mobility and the shift of Dirac point. In the paper, we obtained the mobility of graphene by the directional transfer technique based on the following equation without considering the contact resistance: $\mu = 1/C_{ox} \cdot L/W \cdot g_m/V_{ds}$, in which $g_m = \Delta I_{DS}/\Delta V_G = W/L \cdot \mu C_{ox} V_{ds}$, g_m is the peak transconductance, and L is the length of the device between source and drain, W is the width of the device, and C_{ox} is the insulating layer capacitance between the gate insulator and the conducting graphene channel. The mobility was extracted to be 605 $cm^2V^{-1}s^{-1}$, which is much higher than that of the RGO-based FET biosensors,[45-46] but lower than that of the CVD-grown graphene nanoribbon FET biosensor.[28] Dong et al.[45] reported a label-free DNA FET biosensor based on RGO and gold nanoparticles-modified RGO, in which the mobility of RGO and Au-RGO was about 0.015 $cm^2V^{-1}s^{-1}$ and 9 $cm^2V^{-1}s^{-1}$, respectively. However, the sensitivity in this work was not reported yet. Hasegawa et al.[46] fabricated a RGO-based FET biosensor for IgE detection, in which the mobility of RGO was around 6 $cm^2V^{-1}s^{-1}$ and the detection limit of IgE detection was 8.1 ng/ml. Lerner et al.[28] proposed a transfer method of patterning a CVD-grown graphene nanoribbon for scalable production of nanosensors in order to detect naltrexone with a high sensitivity (10 pg/mL), in which the mobility of the CVD-grown graphene nanoribbon was on average 1500 $cm^2V^{-1}s^{-1}$. This clearly shows that the transferred SLG has a high quality and the developed SLG FET biosensor is capable of achieving a high sensitivity.

The reproducibility of DNA sensing was investigated by calculating relative standard deviation (RSD) from the independent SLG FET biosensors. The relative

standard deviation (RSD) value was found to be 8.5%, demonstrating a satisfactory chip to chip variation.

Finally, the reusability of the SLG FET biosensor was also investigated (Figure S5). The experimental process was performed as mentioned in the previous report.[10] 1 pM complementary DNA was chosen as target. After three times' cycle of denaturation and re-hybridization, the hybridization signal percentage of the second time and the third time hybridization was found to be 86% and 76% of the first time hybridization signal, respectively, indicating that the SLG FET biosensor has a good reusability and can be reused multiple times.

Conclusions

In conclusion, we have developed a novel FET nanobiosensor based on CVD-grown monolayer graphene transferred to the device by the directional technique and high-affinity PNA-DNA hybridization for ultrasensitive, label-free, and highly specific detection of DNA. Rapid production of the CVD-grown monolayer graphene FET biosensor was achieved by the simple directional transfer technique. Compared to the previously published work, the fabricated CVD-grown SLG FET biosensor showed a great sequence-specific affinity to target DNA and achieved an excellent DNA detection sensitivity as high as 10 fM. What's more, this FET nanobiosensor could also discriminate complementary DNA from one-base mismatched DNA and non-complementary DNA with high specificity. Meanwhile, the SLG FET biosensor shows satisfactory reproducibility and stability. This report may further promote development in graphene transfer techniques and pave a progressive avenue to design and fabricate a novel electronic nanobiosensor for ultrasensitive and highly specific detection of DNA in disease diagnostics.

ASSOCIATED CONTENT:

Supporting information. Detailed descriptions of schematic illustration of the conventional transfer technique process for fabrication of a G-FET biosensor, Raman and XPS characterization of graphene surface, electrical properties of G-FET, reusability of the SLG G-FET biosensors. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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Figure captions:

Scheme 1. Schematic illustration of the directional transfer technique process for fabrication of a graphene-based FET biosensor.

a. Single layer graphene (SLG) is grown on 1×1 cm^2 copper by CVD.

b. PMMA is coated on SLG/copper by a spin-coater.

c. PMMA/SLG/copper is cut into small pieces by scissor, as desired.

d. The small piece of PMMA/SLG/copper is immersed into etchant for removing copper.

e. PMMA/SLG is directionally transferred onto the FET chip, where the sensing array is located.

f. The PMMA/SLG is transferred to the sensing array surface by the directional transfer technique.

g. The G-FET is obtained by treatment with hot acetone (60°C) overnight and subsequently thermal annealing at 200°C in vacuum for decomposing PMMA.

Figure 1. (a) The optical image of the FET devices fabricated on a 4-inch wafer. (b) The optical images of an individual FET device. (c) The SEM images of the sensor arrays of an individual FET device.

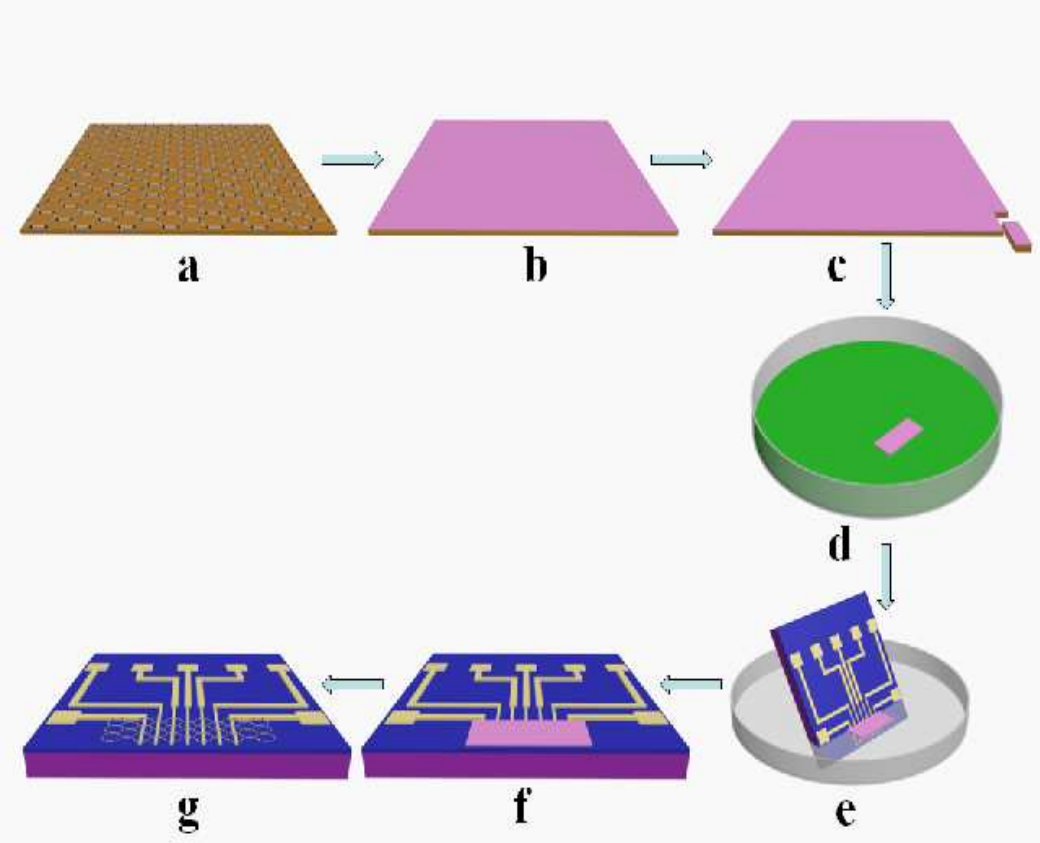
Figure 2. (a) Metallurgical microscopy images showing 6 individual sensor channels coated with the CVD SLG spanning across Au electrodes. (b) Raman spectrum of the CVD SLG prepared by the directional transfer technique, indicating that the transferred graphene is a single layer.

Figure 3. (a) Transfer characteristics of a solution-gate SLG FET before and after thermal annealing. (b) The I_{ds}-V_g transfer curves of the CVD-grown SLG FET device with a constant V_{ds} of 0.1 V in solution. The arrows refer to the direction of the gate

voltage sweep. The black lines and red lines represent the transfer characteristics curves when the gate bias is swept forward and backward between 0 V and 1 V.

Figure 4. (a) The transfer characteristics of the PNA-functionalized CVD SLG FET biosensor incubated with 1 × PBS buffer solution, as a blank control. (b) The transfer characteristics of the CVD SLG FET biosensor without PNA modification hybridized with 1 × PBS, 10 fM, 1 pM, 100 pM complementary target DNA (cDNA), respectively. (c) The V_{CNP} shift of the PNA-immobilized CVD SLG FET biosensors hybridized with different concentrations of non-complementary DNA. (d) The V_{CNP} shift of the PNA-immobilized CVD SLG FET biosensors hybridized with different concentrations of one-base mismatch DNA target.

Figure 5. (a) The V_{CNP} shift of the PNA-immobilized CVD SLG FET biosensors hybridized with complementary DNA target with concentrations ranging from 1 fM to 100 pM. (b) A summary of V_{CNP} shift of the CVD SLG FET biosensor: the FET biosensor without PNA modification upon hybridization with various concentrations of complementary DNA target, and the PNA-immobilized FET biosensor upon hybridization with various concentrations of complementary, non-complementary, one-base mismatched DNA targets, respectively.



Scheme 1

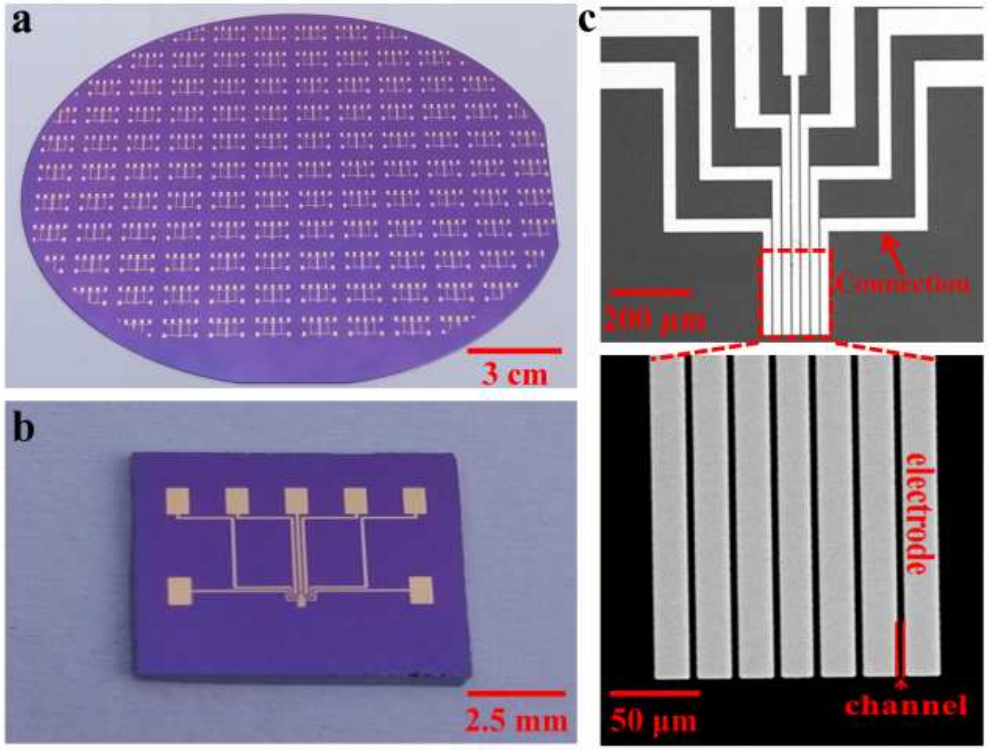


Figure 1

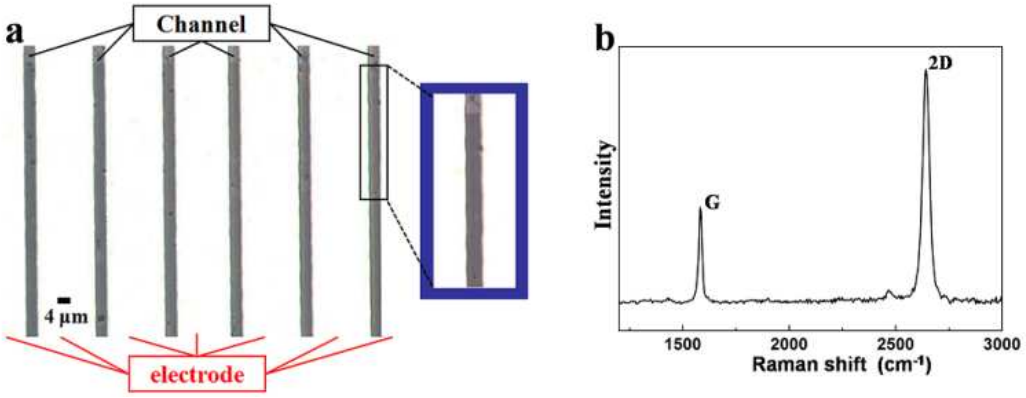

a
Channel
4 μm
electrode
b
2D
G
Intensity
1500
2000
2500
3000
Raman shift (cm⁻¹)

Figure 2

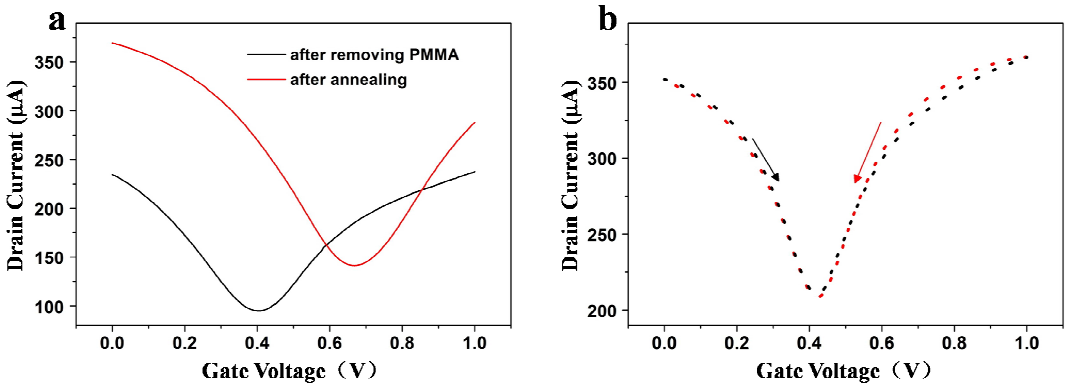

a
after removing PMMA
after annealing
Drain Current (μA)
Gate Voltage（V）
b
Drain Current (μA)
Gate Voltage（V）

Figure 3

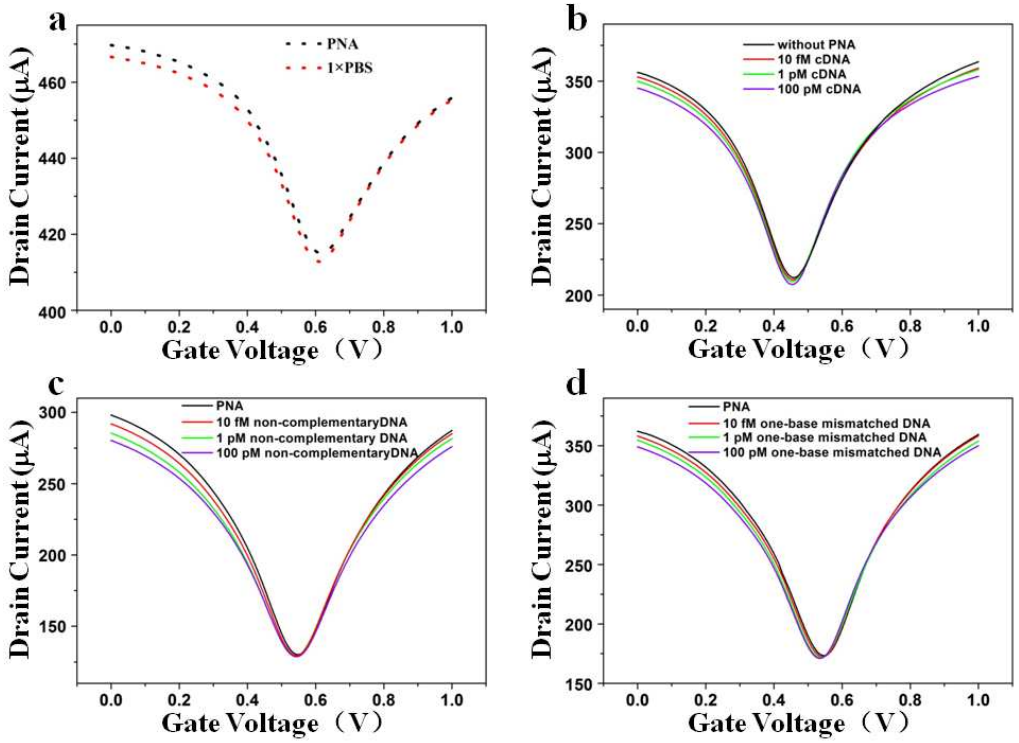

a
PNA
1×PBS
Drain Current (μA)
Gate Voltage（V）
b
without PNA
10 fM cDNA
1 pM cDNA
100 pM cDNA
Drain Current (μA)
Gate Voltage（V）
c
PNA
10 fM non-complementaryDNA
1 pM non-complementary DNA
100 pM non-complementaryDNA
Drain Current (μA)
Gate Voltage（V）
d
PNA
10 fM one-base mismatched DNA
1 pM one-base mismatched DNA
100 pM one-base mismatched DNA
Drain Current (μA)
Gate Voltage（V）

Figure 4

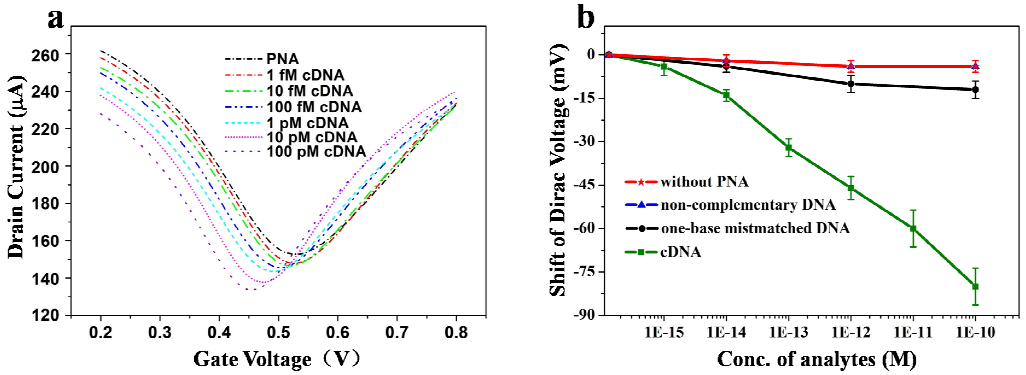

a
Drain Current (μA)
Gate Voltage（V）
PNA
1 fM cDNA
10 fM cDNA
100 fM cDNA
1 pM cDNA
10 pM cDNA
100 pM cDNA
b
Shift of Dirac Voltage (mV)
Conc. of analytes (M)
without PNA
non-complementary DNA
one-base mistmatched DNA
cDNA

Figure 5

"Table of Contents"

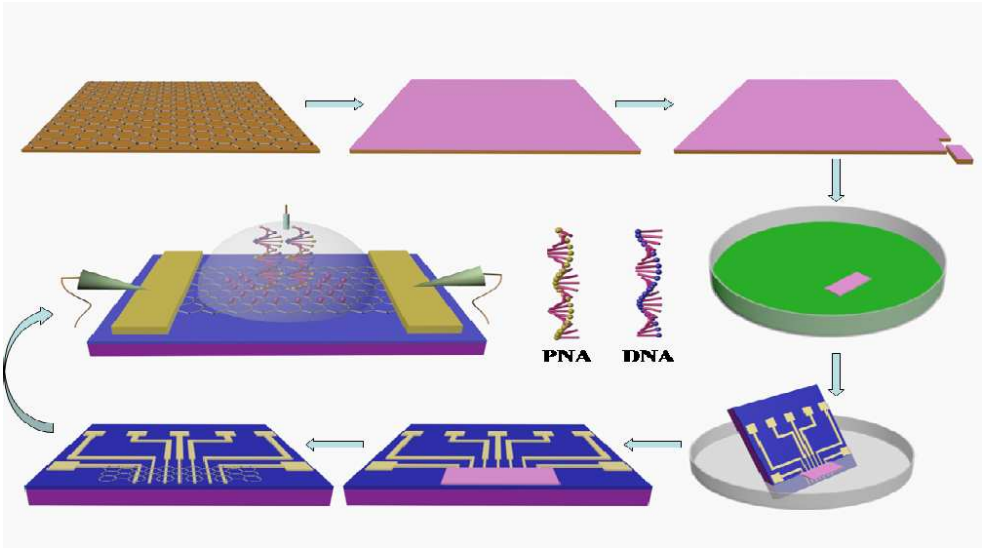


Table of Contents (TOC) graphic for manuscript entitled "Fabrication of Ultrasensitive Field Effect Transistor DNA Biosensors by A Directional Transfer Technique Based on CVD-Grown Graphene".