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## Basic study on Peptide Aptamer-modified Single-Walled Carbon Nanotube Field-Effect Transistor (SWCNT FET) for biomolecule analysis

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In this work, single-walled carbon nanotubes (SWCNTs) were synthesized by using ethanol chemical vapour deposition (CVD) method, then proceeding to fabricate SWCNT field-effect transistors (SWCNT FETs) applied for biosensing application. Peptide aptamers that contain biotin were immobilized onto the SWCNT channel of SWCNT FETs for streptavidin detection in order to investigate the bioreceptor function of peptide aptamers. Peptide aptamers showed promising candidate in terms of FET biosensors owing to their short size with small binding footprint and 20 types of amino acids. As a result, peptide aptamers were successfully immobilized on SWCNT FET that functionalized the detection of biomolecules by FET inside sensitive region of Debye length.

Keywords: peptide aptamer, SWCNT FET, streptavidin

#### 1. Introduction

Single-walled carbon nanotube field transistors (SWCNT FETs) have drawn much attention biosensing application thanking quasi-one-dimensional SWCNT channel which is very small compared to the biomolecule dimension. However, short Debye- screening is a very severe drawback of SWCNT FET, which makes probe molecules that must be shorter than the Debye length. This issue could be resolved by using small receptor like DNA aptamers [1, 2]. In this study, peptide aptamers were used as bioreceptor. In comparison with DNA aptarmers, peptide aptamers are relatively shorter in size with smaller binding footprint that allows more thorough and precise capture of target. Therefore, with the same surface area, peptide aptamers provide higher binding-site density and lower background signal that arises from the nonspecific binding of target analyte [3]. Additionally, DNA aptamers are limited to 4 types of nitrogenous base (A, T, C, G) while peptide aptamers may contain 20 kinds of amino acids. Therefore, peptide aptamer-based biosensors can access to a wide such variety of unreachable targets, low-molecular-weight biomarkers.

The binding of biotin and streptavidin is one of the strongest non-covalent interactions known in nature. In this work, peptide aptamer receptor that contains biotin was immobilized on to the SWCNT channel of SWCNT FET. Then streptavidin was introduced for specific recognition in order to investigate the bioreceptor function of immobilized peptide aptamers.

#### 2. Experimental

# 2.1. SWCNTs synthesis by ethanol chemical vapour deposition (CVD) method.

SWCNTs were synthesized directly between two catalytic cobalt patterns on SiO<sub>2</sub> (100nm)/Si substrate at desirable ethanol pressure and temperature for 30 min by using ethanol CVD method. In this work, two important parameters of the CVD process including growth temperature and pressure were investigated for

optimizing SWCNT growth. In case of temperature dependence, CNTs growth was implemented at pressure of 1200 Pa for 30 minutes with variant temperatures. In case of pressure dependence, SWCNT growth was carried out at 850°C for 30 minutes with variant pressures. The as-grown SWCNTs were investigated by Raman spectroscopy and scanning electron microscopy (SEM) image.

#### 2.2. SWCNT FET fabrication.

After growing SWCNT, the Ti/Au (2 nm/45 nm) Source and Drain electrode pads were formed by using photolithography and lift-off technology. The fabricated SWCNT FETs were characterized with back-gated measurement setup by using Agilent 4156C Precision Semiconductor Parameter Analyzer (Figure 1).

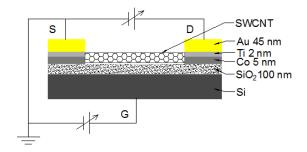


Fig. 1: Back-gated schematic circuit for measuring the electrical characteristics of fabricated SWCNT FET.

# 2.3. Immobilization of peptide aptamers onto SWCNT FETs.

Peptide contains aptamer that biotin were immobilized onto **SWCNT** channel 1-pyrenebutanoic acid succinimidyl ester (PBASE) linker (Figure 2). Firstly, 6 mM of PBASE in dimethylformamide (DMF) was dropped onto the SWCNT channel and kept for 1 hour at room temperature followed by rinsing with pure DMF and drying with N<sub>2</sub> gas. Then 0.5 mM peptide-aptamer that contains biotin was dropped and kept for 18 hour at room temperature followed by rinsing with pure water and drying with  $N_2$  gas. The PBASE can make noncovalent  $\pi\text{-stacking}$  with SWCNTs in aromatic ring tail and covalent amide bond with peptide aptamers in another tail. Then the peptide aptamer-modified SWCNT FET was treated with 100 mM ethanolamine for 30 minutes to block the non-specific binding site followed by rinsing with pure water and drying with  $N_2$  gas.



Fig. 2: Biofunctionalization of SWCNT channel with peptide aptamer containing biotin.

#### 2.4. Detection of streptavidin.

The polydimethylsiloxane (PDMS) cavity was attached onto the device to contain the phosphate-buffered saline (PBS) 1 mM (pH 7.8). The current between FET's source and drain was measured with a constant liquid-gated and source-drain voltage of 0V and 200 mV, respectively. The reference electrode Ag/AgCl was used. The various streptavidin concentrations (from 1  $\mu M$  to 2 mM) were introduced for real-time measurement (Figure 3).

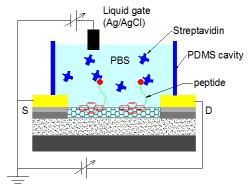


Figure 3: Schematic circuit of experimental setup for streptavidin detection.

#### 3. Results and discussion

# 3.1. Temperature and pressure dependence of SWCNTs growth.

Figure 4 shows the SEM images of CNTs growth as a function of growth temperatures at 1200 Pa for 30 minutes. The results indicated that SWCNTs started growing at 800°C with increases in density and length. At lower temperatures, introduced ethanol might not be pyrolyzed properly, leading to the formation of damaged graphene or single-walled nanohorn which was confirmed by Raman spectra. SWCNTs grown at 850°C appeared long enough to bridge between two cobalt patterns with highest density.

Figure 5 shows the SEM images of SWCNTs as a function of pressure at  $850^{\circ}$ C for 30 min. The flow rate

of ethanol introduction is proportional with chamber pressure. SWCNTs were sparse at 220 Pa due to less amount of ethanol source and denser at higher pressures because of larger amount of ethanol source.

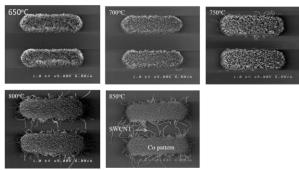


Figure 4: SEM image of temperature dependence of SWCNT growth.

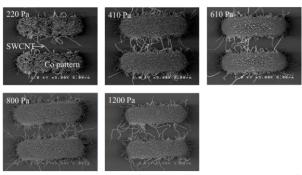


Figure 5: SEM image of the pressure dependence of SWCNT growth.

Figure 6 shows the Raman spectrum of SWCNT grown at 1200 Pa of ethanol, 850°C for 30 minutes. These results clearly indicated that SWCNTs were successfully synthesized by CVD method. The characteristic radial breathing mode (RBM) of SWCNT was observed at 170 cm<sup>-1</sup> corresponding to as-grown SWCNT's diameter of around 1.4 nm.

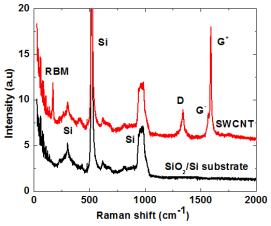


Figure 6: Raman spectrum of as-grown SWCNT.

## 3.2. Characterization of fabricated SWCNT FETs.

Figure 7 shows the electrical characteristics of the fabricated SWCNT FET using back-gated measurement scheme. The drain current decreased with increase of gate voltages. These results indicated that

the fabricated SWCNT FET exhibited typical p-type characteristic at room temperature in air ambient. The obtained "on/off" current ratio and subthreshold swing (S) factor were  $1.12 \times 10^4$  and 152 mV/decade, respectively. The smaller the S-factor, the higher the sensitivity. Our results suggested that the fabricated SWCNT FETs are suitable for biosensing applications.

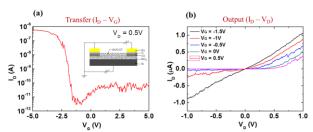


Figure 7: Electrical characteristics of fabricated SWCNT FET using back-gate measurement: (a) Transfer curve and (b) Output curve.

## 3.3. Biosensing application of fabricated SWCNT FETs.

Figure 8 shows the real-time detection of streptavidin using fabricated SWCNT FET biosensors. The drain current decreased with increases of streptavidin concentration. This indicates that partially positive charges from streptavidin were detected by the fabricated SWCNT FET. The initial result indicated that peptide aptamers can be immobilized actively on SWCNT FET, and functionalized the detection of biomolecules by FET inside sensitive region of Debye length.

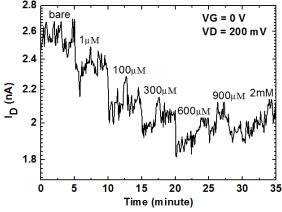


Figure 8: Real-time detection of streptavidin using peptide aptamer-modified SWCNT FET biosensor.

#### 4. Conclusion

The peptide aptamer-modified SWCNT FETs were studied for biomolecule analysis. SWCNTs were successfully synthesized and optimized by using the CVD method at 850°C, 1200 Pa for 30 min. The fabricated SWCNT FETs exhibited the typical p-type characteristic with the "on/off" current ratio and S-factor of  $1.12 \times 10^4$  and 152 mV/decade, respectively. The peptide aptamer receptors can be immobilized actively onto SWCNT FET, and functionalized

detection of biomolecules by FET inside sensitive region of Debye length. The proposed peptide aptamer-modified SWCNT FET system shows great promise for biosensing application.

#### 5. References

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