Fabrication, AFM Imaging and Applications of Si-Nanowire FET Aptamer Biosensors

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Statement of Purpose: Aptamers are single-stranded oligonucleotides which can specifically bind to target proteins [1]. Biosensor is an analytical device which converts a biological response into an electrical signal. Among various biosensors, Si-nanowire (Si-NW) field effect transistor (FET) systems attracted a lot of research interests since Si-NW has extremely high surface-to-volume ratio and sensitivity of the carrier mobility, which make ultra-sensitive label free detection [2]. In this work, a novel aptamer biosensor gate was successfully prepared. Furthermore, The Si-NW aptamer biosensor was successfully applied for the real-time detection of electronic signals during and after protein binding.

Methods:

Surface modification: Si-NW was surface modified with 3-aminopropyl diethoxysilane (3-APDES) and succinic anhydride to introduce amine and carboxyl groups on Si-NW according to the protocol reported by Gao *et al.* [2] and Hong *et al.* [3].

Aptamer binding: Anti-thrombin DNA aptamer was dissolved in phosphate buffer (PB, pH 8). Carboxylated Si-NWs were immersed in PB and activated with 1-ethyl-3-(dimethylaminopropyl)carbodiimide hydrochloride and N-hydroxysulfosuccinimide. The aptamers were surface immobilized on Si-NWs by the amide bond formation between amine groups of aptamers and the activated carboxyl groups on Si-NWs. Atomic force microscopic (AFM) analysis in a tapping mode was carried out to characterize the aptamers immobilized on Si-NWs.

Real-time electrical bio-sensing: After surface immobilization of aptamers on *p*-type Si-NW channels, we measured the real time current of Si-NW FET functionalized with aptamers during the injection of thrombin samples at a constant bias voltage (V_b). First, acetate buffer (pH 5.4) was dropped as a control solution on the aptamer-modified Si-NW FETs through the electrochemical cell while V_b was maintained at a value of 100 mV. Then, thrombin and blood samples were injected in a same way. We waited to settle down the fluctuating conductance of FETs to a constant value after each sample injection.

Results: Figures 1 shows AFM height and cross-sectional images before and after binding of aptamers on Si-NW. The images showed the morphology of aptamers attached to Si-NW. The height of the aptamers was *ca.* 4 nm being immobilized at a certain interval. In addition, aptamer/thrombin complexes immobilized on Si-NW, after incubation with thrombin samples, appeared to have a mean height of 8 nm. The results visualized and confirmed the successful immobilization of aptamer on Si-NW. The ability of aptamers to strongly bind to target protein has been exploited for biosensor applications.

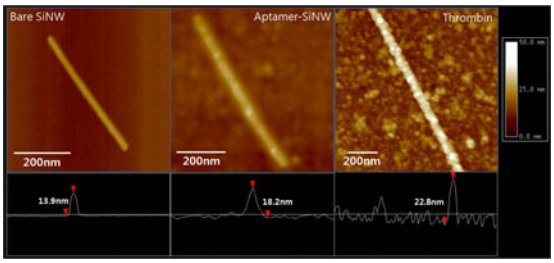

Figure 1. AFM height and cross-sectional images for bare Si-NW, anti-thrombin DNA aptamers on Si-NW, and thrombin bound anti-thrombin DNA aptamers on Si-NW.

Figure 2 shows the current variation in time according to the injections of thrombin samples. Since the isoelectric point (pI) of thrombin is 7.0, the samples are positively charged under the experimental condition of pH 5.4, which is much lower than the pI of thrombin. These positively charged samples can effectively screen the negative charges of aptamers and act as positive point charges on the gate dielectrics, resulting in the charge depletion in the NW channel and the decrease in conductance in case of the *p*-type Si-NW biosensors.

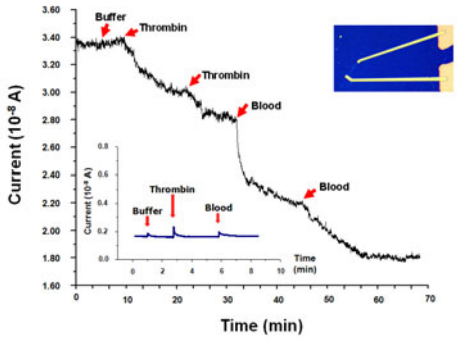

Figure 2. Real-time detection of electronic signals (current) from anti-thrombin DNA aptamer functionalized *p*-type Si-NW FET biosensor during and after thrombin binding. Inset shows the response of an as-prepared Si-NW FET without the aptamers.

Conclusions: Surface modifications of Si-NWs were successfully carried out with 3-APDES and succinic anhydride to introduce amine groups and carboxyl groups. AFM analysis confirmed the immobilization of anti-thrombin aptamers on the surface modified Si-NWs. The Si-NW FET based aptamer biosensor gate could be successfully applied for the real-time detection of electronic signals during and after thrombin binding.

References:

[1] Tuerk C. Science 1990; 249: 505-510.
[2] Gao Z. Anal. Chem. 2007; 79: 3291-3297.
[3] Hong BJ. Langmuir 2003; 19: 2357-2365.
[4] Lee HS. Biosens. Bioelectron. 2008; In press.