Nano Energy 30 (2016) 77–83

Contents lists available at ScienceDirect

Nano Energy

journal homepage: www.elsevier.com/locate/nanoen

Self-powered triboelectric aptasensor for label-free highly specific thrombin detection

Yun Kyung Jung[a,b], Kyeong Nam Kim[c], Jeong Min Baik[c,*], Byeong-Su Kim[b,d,**]

[a] School of Biomedical Engineering, Inje University, Gimhae, Gyeongnam 50834, Republic of Korea
[b] Department of Energy Engineering, Ulsan National Institute of Science and Technology (UNIST), Ulsan 44919, Republic of Korea
[c] School of Materials Science and Engineering, KIST-UNIST-Ulsan Center for Convergent Materials, Ulsan National Institute of Science and Technology (UNIST), Ulsan 44919, Republic of Korea
[d] Department of Chemistry, Ulsan National Institute of Science and Technology (UNIST), Ulsan 44919, Republic of Korea

ARTICLE INFO

Keywords:
Triboelectric
Biosensor
Gold nanoparticle
Thrombin
Aptamer

ABSTRACT

An aptamer-based triboelectric biosensor is developed for a highly specific, label-free and self-powered detection of thrombin. For the first time, intermolecular recognition interactions are used to develop a selective nanosensor based on triboelectric effect. Positively charged Au nanoparticles (Au NPs^{+}) with large difference in triboelectric polarity and work function are assembled onto Al film to increase the electrical output of triboelectric nanogenerators (TENG). Modification of anti-thrombin aptamers on the Au NPs^{+}-assembled TENG affords the triboelectric nanosensor highly selective toward thrombin, even in clinical samples because of specific binding affinity between aptamers and thrombin unlike random DNA-modified TENGs with undetectable response. A 0.41 nM limit of detection is achieved, which is directly demonstrated by the number of commercial LED lights without any supporting equipment such as power source and electrometer. Our study demonstrates an innovative and unique approach toward the self-powered and label-free detection of thrombin for rapid and simple in-field analysis.

1. Introduction

Detection of disease-related proteins holds significant applications in clinical diagnosis [1–3]. Thrombin, a serine protease involved in the coagulant cascade has great importance in physiological and pathological processes [4,5], such as regulation of tumour growth, metastasis, and angiogenesis [6]. The concentration of thrombin in blood depends on the physical condition of the subject. Thrombin can be almost absent in the blood of healthy subjects, but it can vary from low nanomolar to micromolar concentrations during the coagulation process [7]. Taking advantage of high specificity of intermolecular recognition reactions between thrombin and thrombin-specific ligands such as antibody or aptamer, significant efforts have been made to develop biosensors to directly detect thrombin.

Among various biological recognition elements which determine the degree of selectivity of the biosensor, nucleic acid-based aptamers have been recognized as very promising bioreceptors because of their outstanding selectivity, sensitivity, and stability [8]. Notably, thrombin binding to its aptamer is the most commonly and intensively used model system to demonstrate aptamer-based affinity assays in the clinical area [9–17]. Optical and electrochemical approaches have primarily been applied to detect the thrombin [1–3,10–17]. For example, colorimetry and fluorometry are the most widely used due to their convenience, high sensitivity, and application to point-of-care testing [1–3,11–17]. However, there are still significant challenges including complicated and time-consuming probe-labelling steps, high background noise, and difficulties in inferring accurate and quantitative results from the collected optical signals. In contrast, electrochemical approaches are advantageous in label-free and quantitative measurement capability as well as being practicable for in-field lab-on-a-chip devices, yet they have some limitations related to relatively low detection sensitivity, electrode fouling, electrochemical stability of reagents, side electrochemical reactions, and most critically, the requirement for an external power supply [2,3,10,12].

Recently, the concept of self-powered sensors based on the triboelectric nanogenerator (TENG), which converts mechanical energy from the environment into electricity, has received considerable attention because no battery is needed to power the device [18–23]. Considering the operational mechanism of self-powered device, the electric output signal can be critically determined by molecules

* Corresponding author.
** Corresponding author at: Department of Energy Engineering, Ulsan National Institute of Science and Technology (UNIST), Ulsan 44919, Republic of Korea.
E-mail addresses: jbaik@unist.ac.kr (J.M. Baik), bskim19@unist.ac.kr (B.-S. Kim).

http://dx.doi.org/10.1016/j.nanoen.2016.09.039
Received 12 July 2016; Received in revised form 22 September 2016; Accepted 24 September 2016
Available online 28 September 2016

adsorbed on an electrically active surface. Although Wang and co-workers have reported the early examples of sensing by TENG for mercury ions and glucose [18,19], their system was focused on the marker specific to mercury ions or the TENG was used as a power source for the sensor. There has been no report on the use of selective binding events which have specific energy states in this type of sensors to date. Thus, specific, label-free and self-powered methods are highly desirable for rapid and simple in-field analysis.

Herein, we describe the development of an aptamer-based triboelectric biosensor for the highly specific, label-free and self-powered detection of thrombin. For the first time, specific aptamer-protein interactions are used to develop a fully integrated, stand-alone and self-powered nanosensor. With respect to previous works in the field, our approach is unique in a number of attributes. First, to maximize the charge density on both surfaces, the materials with the largest difference in triboelectric polarity [24] and work function [24] are chosen. In that sense, positively charged Au nanoparticles (Au NPs^+) with large surface area [25] were assembled onto the Al film to improve the performance of the TENG [26]. These assembled Au NPs^+ not only act as steady gaps between the two plates at the strain-free condition, but also increase the electrical output of the TENG by enlarging the contact area of the two plates. Second, through modification of thiol-modified anti-thrombin aptamers on the assembled Au NPs^+, the nanogenerator became a highly selective nanosensor toward thrombin detection because of the strong binding affinity between them. On the basis of this unique structure, the output voltage and current of the Au NPs^+-assembled triboelectric nanosensor significantly decreased after decoration with negatively charged aptamers. Under optimum conditions, this aptamer-modified TENG sensor showed the enhanced electrical signals only after incubation with thrombin, with a detection limit of 0.41 nM. The random DNA-modified TENG did not show any response to thrombin. Third, the electricity generated by the interactions with thrombin directly lit up commercial green LEDs, which showed great potential as a simple detection systems as well as facilitating label-free sensing without complex labelling of expensive dyes. Our study demonstrates a unique advanced approach toward the self-powered detection of thrombin for rapid and simple in-field analysis compared to the widely used colorimetric or fluorometric methods (Table S1).

2. Experimental section

2.1. Materials

Thiol-modified oxidized (S-S) TASSET aptamer sequences (5′-AGT CCG TGG TAG GGC AGG TTG GGG TGA CT-3′) and random DNAs (5′-CGT TAC AGT TGG GTA ACG GG-3′) were synthesized and purified by Integrated DNA Technologies (Coralville, IA, USA). Other proteins and chemicals including thrombin from human plasma, streptavidin (Str), lysozyme (Lyso), HEPES, and $MgCl_2$ were purchased from Sigma-Aldrich, Inc. (St. Louis, MO, USA) and used without further purification. Sample stock solutions were prepared by directly dissolving the proteins in HEPES buffer (1.0 mM HEPES, 1.0 mM $MgCl_2$, pH 7.26) and stored in a refrigerator at −20 °C. Polydimethylsiloxane (PDMS, Sylgard 184, Dow Corning) were used for the fabrication of TENG. The base monomer (Sylgard 184A) and curing agent (Sylgard 184B) were mixed in a mass ratio of 10:1, followed by vacuum drying to degas the PDMS mixture. After 30 min, 1.0 mL of mixture was coated onto the silicon wafer, and allowed to solidify into an amorphous free-standing film by heating on an oven at 90 °C for 5 min.

2.2. Fabrication of TENGs

An aqueous solution of 0.40 mL of 4-(dimethylamino)pyridine (DMAP)-Au NPs (Au NP^+) [26] and carboxylic acid-modified Au NPs (Au NP^-) [27–30] (conc. of 60 mg/L) was casted on Al bottom electrode (1 cm×1 cm) and dried for 2 h at room temperature. One hundred μm-thick pure PDMS film was attached on the Al top electrode by the double sided polyimide tape. A triboelectric nanogenerator was prepared by stacking two pieces of the structures and mechanically triggered by a linear motor after optimizing the conditions for impact force and frequency (Fig. S1). As the input force and the frequency increase, the output current of the Au NP^+-assembled TENG increases and reaches around 35 μA in both cases. A compressive force of 50 N and a frequency of 10 Hz were employed in this study to generate the high and reliable electrical signals by the TENG. The long-term stability of the Au NP^+-assembled TENG was also evaluated by using a pushing tester for 24 h. As shown in Fig. S2, consistent output voltage of the TENG was maintained, demonstrating the successful integration of Au NPs onto the Al film substrate and its stability during the operation of

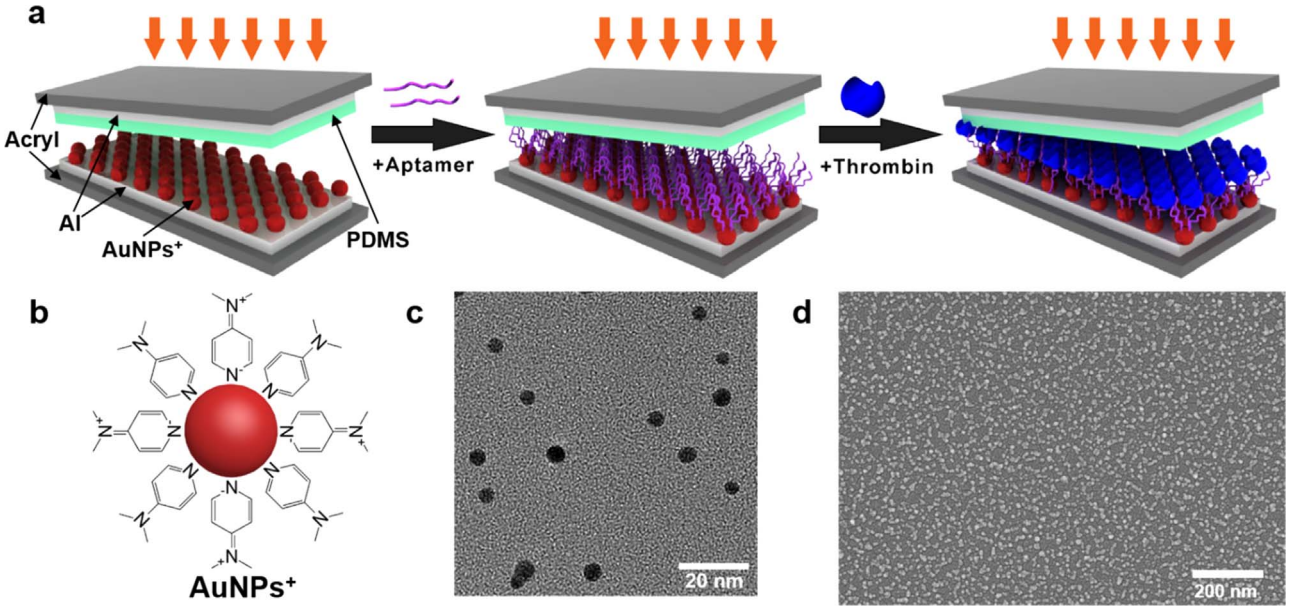

Fig. 1. (a) Schematic illustration of thrombin detection by triboelectric biosensors containing DNA aptamer-decorated Au NPs assembled onto the surface of Al film. (b) Positively charged Au NPs (Au NPs^+) decorated with 4-(dimethylamino)pyridine (DMAP). (c) TEM image of Au NPs^+ and (d) SEM image of Au NPs^+ on Al film.