Bioassay of Target Proteins Using A NiCr Strain Gauge-Cantilever Liposome Biosensor with Droplet-Sealed Structure

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Abstract

We have developed Si micro-cantilever biosensor with embedded NiCr thin film strain gauge and DPPC liposome immobilized on its surface. This time, a PDMS-based sealed reservoir structure was newly fabricated and added on the sensor to keep the protein solution stable, where a long-time stable detection was achieved. The resistance of the cantilever sensor increased with time in carbonic anhydrase from bovine (CAB) or lysozyme aqueous solution, and the characteristic of chronological resistance change varied with the kind and concentration of proteins. We expect that this micro-cantilever sensor with droplet sealed structure becomes an effective device for bioassay of proteins.

1. Introduction

We have reported that the micro-cantilever sensor with thermally stable NiCr strain gauge was fabricated through surface micromachining and its surface was covered with a self-assembled monolayer (SAM), where DPPC liposomes were immobilized as sensing biomolecules [1]. The deflection of cantilever, caused by liposome-protein interaction [2], was measured through the resistance change of strain gauge. The sensor showed chronological detection of target proteins successfully, but the target protein solution lacked stability because of droplet evaporation, resulting in a short-time detection dependent on the droplet size, so that some protein solutions with low activity could not be accurately detected. Herein we developed a PDMS-based sealed structure to sustain the liquid condition and adopted the detection of different kind of target proteins as a parameter of concentration.

2. General Instructions

Figure 1 exhibits that the NiCr thin film strain gauge was formed on the micro-cantilever, where the composition of Ni:Cr was adjusted 8:2 to optimize the sensitivity as resistance change this time. Figure 2 shows an AFM image of DPPC liposomes immobilized on the SAM/Au layer. It is found that the spherical liposome is immobilized intact on the SAM and its diameter is 50-100 nm, which agrees well with the filter mesh size when extruding the liposome suspension through the filter as a final lipo-

some preparation process.

Fig. 1 An optical microscopic image of the micro-cantilever with NiCr thin film strain gauge.

Fig. 2 An AFM image of DPPC liposomes with diameters of 50-100 nm immobilized on the SAM/Au layer.

The newly fabricated droplet sealed structure is illustrated in Figure 3. The reservoir was covered with a cover glass to prevent the evaporation of solution during measurement. In Figure 4 is plotted the resistance change rate (ΔR/R0) of the micro-cantilever sensor in lysozyme aqueous solutions against time. The resistance is almost stable in water but increases with time in lysozyme solutions. Moreover, a long-time stable detection more than 30 minutes has been achieved by using the droplet sealed
structure, and the resistance change tendency are clearly observed in every solution. Figure 5 shows the resistance change rate of the micro-cantilever sensor vs. protein concentration, after filling the reservoir with protein solutions for 30 min. The resistance increases in both CAB and lysozyme solutions, and the resistance change rate increases with increasing the concentration of either protein. By comparing the resistance changes in CAB and lysozyme solutions with the same concentration (e.g., 800 µM), it is especially found that the increasing rate in CAB solution is much larger than that in lysozyme solution. These results indicate that this micro-cantilever sensor with droplet sealed structure is excellent at detecting the kind and concentration of proteins.

3. Conclusions
The droplet-sealed structure on the cantilever-based liposome biosensor brings about a long-time stable detection of proteins, and this biosensor is confirmed to be excellent at detecting the kind and concentration of proteins.

Fig. 3 A cross-sectional view (a) and a surface photograph (b) of the droplet sealed structure.

Fig. 4 Resistance change rate ($\Delta R/R_0$) of the micro-cantilever sensor in deionized water and lysozyme aqueous solutions against time.

Fig. 5 Resistance change rate ($\Delta R/R_0$) of the micro-cantilever sensor vs. protein concentration after filling the reservoir with CAB or lysozyme aqueous solutions for 30 min

Acknowledgements
This research was partly supported by a Grant-in-Aid for Scientific Research (KAKENHI Grant No. 25249048) from the Japan Society for the Promotion of Science (JSPS).

References