マイクロ流路システムを用いた MCC 法による生体高分子の計測

Biomacromolecule detection based on molecular charge contact method with micro-fluidics system

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1. Introduction

In demand of the recently aging society, the development of medical diagnostic technologies is becoming increasingly important. Among various biosensors, a field-effect transistor (FET)-based biosensor has a potential to realize next-generation diagnostics, owing to its unique feature of the potentiometric methodology. In fact, an ion-sensitive FET (ISFET) biosensor has been developed and applied to the detection of DNA complexation and cellular respiration [1, 2]. However, despite of more than 50 years of study, the detection of biomacromolecules such as proteins is still limited due to the limitation caused by the Debye length. The Debye length is based on the electrical double layer, which is the valid sensing area around the gate insulator/electrolyte solution interface less than 10 nm in general, depending on an ionic strength in a solution [3]. Nevertheless, the majority of target biomolecules in clinical applications include macromolecules with the size more than a few nm.

In our previous study, the molecular charge contact (MCC) method has been used to move biomacromolecular charges to enter the Debye Length without controlling the spatial distribution of beads [4]. In this study, a micro-fluidics system was combined with MCC method to avoid noises caused by the distribution and movement of magnetic beads.

2. Methods

A micro-fluidic system is used in this research to form an access for micro flowing. In this research, 1X PBS (Thermo Fisher Scientific, pH 7.4 (ca. 155 mM NaCl) was diluted 100 times and used as a measurement solution (0.01X PBS). Biotin-coated beads (PureCube Biotin MagBeads XL, Φ70 μm) were dropped on the gate surface of ISFET, and a magnet set under ISFET to adhere magnetic beads on gate surface. After the stabilization of surface potential (stage 1), the sample solution containing streptavidin (10 mg/mL) was added onto the gate surface of ISEFT. The next flow started after a surface potential became stable (stage 2), and a final potential was collected (stage 3).

3. Results and Discussion

The change in surface potential of the ISFET with biotin-coated beads on the gate was approximately 70 mV (=stage 2-stage 1) upon adding streptavidin. This is because streptavidin reacted with biotin-coated magnetic beads at the beads/gate interface. After 0.01X PBS was flowed in the system, the change in the surface potential decreased to 25 mV (=stage 3-stage 1). This flowing step got rid of the influence of unreacted streptavidin in the solution; therefore, the latter differential signal shows that streptavidin induced positive charges to the gate surface compared with biotin. The results will be discussed in detail in the conference.

References

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