

Special Article - Biosensor Elements

Kinetic Analysis of Biosensor

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Introduction

Recent researches of biosensor considering kinetics

Limit of Detection (LOD) is highly related to affinity constant. In case of direct method (target molecule directly reacts to the recognition element), Singh et al. [1] and Lazcka et. al. [2] analyzed the associate constant (k_a) and dissociate constant (k_d). Affinity constant (K) is calculated from the ratio of k_a/k_d . They discussed the LOD of pathogen detection.

Researches of Surface Plasmon Resonance (SPR) biosensor usually reported the kinetic analysis, because the well-organized book [3] and BiaCoRE provides the assay handbook [4]. Those books introduce the calculation method using the slope of time-course measurement.

On the other hands, Kim [5] proposed the mathematical modeling of protein adsorption onto the solid interface based on the kinetics. He compared the Langmuir adsorption isotherm and the Random Sequential Adsorption (RSA) model to fit the mathematical model to the real sensing system.

Kinetic analysis of direct method was also introduced by Khodami et al. [6]. They originally developed long-range surface plasmon waveguides and proposed the kinetic analysis of the binding event of Bovine Serum Albumin (BSA) and anti-BSA antibody. Further, they reported the kinetic analysis of sandwich and inhibition assay [7]. Song et al. also investigated the biosensor using fluorescence dye by using the first-order kinetics [8]. We think that those calculation methods are not useful for only their biosensors but also other sensors.

Kinetic analysis of biosensor

The basis of kinetic analysis of the biosensor is Langmuir adsorption isotherm. It is an expression expressing adsorption by chemisorption, and is an expression theoretically derived assuming

Abstract

Most of researches of biosensor generally report new sensor surfaces and new target molecules. It is expected that the sensitivity of biosensor could be improved more, if the researchers consider the design of the reaction with kinetic analysis. However, it is believed that the kinetic analysis is difficult to understand the mathematical treatment. Further, the kinetic analysis requests the sensor gram with respect to time (time-course measurement) to calculate the associate constant and the dissociate constant from the slope. However, the most of biosensors cannot obtain the sensor gram of time-course measurement. Therefore, researcher avoids analyzing the results by using kinetics.

First, this review article summarizes the recent researches considering with kinetic analysis, and then, it is reported here the calculation method of affinity constant by using the calibration plot only. This information is very useful to design the reaction scheme toward to the improvement of the sensing performance.

Keywords: Surface plasmon resonance; Random sequential adsorption

that all reaction sites are covered with a monolayer. Consider a target molecule that adsorbs to a fixed surface at a constant temperature.

It is expressed as follows.



First, the molar concentration of analyte [A] and the molar concentration of the reaction site on the substrate [S] are defined. And then, the affinity constant (K) can be written;

$$K = \frac{[AS]}{[A][S]} \quad (2)$$

Second, [S] is simply considered as the subtraction of the maximum molar concentration of adsorption site ($[AS]_{max}$) to [AS];

$$[S] = [AS]_{max} - [AS] \quad (3)$$

Eq. (3) is substituted to Eq. (2), and then the equation can be rewritten as;

$$\frac{[A]}{[AS]} = \frac{[A]}{[AS]_{max}} + \frac{1}{K[AS]_{max}} \quad (4)$$

Here, $[AS]_{max}$ and K can be obtained from the intercept and slope of the plot $[A]/[AS]$ with respect to [A].

This method is simply applied to direct and sandwich assays.

Basic reaction of enzyme reaction and competitive assays is illustrated as:



Reporter (B) is initially associated with substrate (S), and B is replaced to A in case of competitive assay. The affinity constant K is depicted as;

$$K = \frac{[B][AS]}{[A][BS]} \quad (6)$$

Since molar concentration of B ([B]) is subtraction of [BS]-[AS],

Eq (6) can be rewritten as;

$$K = \frac{([BS]-[AS])[AS]}{[A][BS]} \quad (7)$$

This equation can be reformed to;

$$[AS][BS] = K[A][BS]+[AS]^2 \quad (8)$$

K can be obtained from the slope of the plot $[AS]$ $[BS]$ with respect to $[A]$.

This calculation procedure is useful for the biosensor using the reporter such as fluorescence or color dye.

In summary, the recent researches on kinetic analysis of biosensor were introduced, and then the kinetic analysis based on Langmuir isotherm was proposed in this article. In fact, the Freundlich adsorption isotherm also needs to be considered. But we introduced only the simple system of the homogeneous reaction, because there is the limitation of number of words in the article, and we want to propose a use of kinetics analysis toward to the strategy to design the system of biosensor.

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