DETECTION OF DNA HYBRIDIZATION USING PROTEIN A MODIFIED ION SENSITIVE FIELD EFFECT TRANSISTOR

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ABSTRACT

A DNA sensor based Ion Sensitive Field Effect Transistor (ISFET) with protein A modification, for the detection of DNA hybridization, was investigated. In this experiment, the surface of the ISFET was first modified with protein A via physical adsorption. Protein A has a sticky property and can bind specifically to the fragment crystallizable (Fc) portion of the antibody leading to the uniform orientation for effectively binding to the antigen. In order to detect DNA hybridization, an anti-biotin antibody was immobilized over protein A and a single-strand biotinylated DNA probe was added to bind to a specific anti-biotin antibody. The voltage shift of DNA/DNA hybridization was observed right after the complementary DNA target was added over the immobilized probe. The non-complementary DNA target was also tested as a negative control. The saturated concentrations for all steps were as follows: 1mg/ml for protein A, 1 mg/ml for anti-biotin antibody, 0.5 μM for biotinylated probe, and 0.5 μM for synthetic DNA target. As high amount of protein A and anti-biotin may barricade the hybridization signal detection; therefore, in this experiment 0.1 mg/ml of both protein A and anti-biotin were tested instead of their saturated concentrations. Using this strategy, DNA hybridization of the synthetic DNA target was successfully detected with the detection limit approximately at 0.08 μM as well as differentiated complementary DNA from non-complementary DNA target.

KEY WORDS: ISFET / PROTEIN A / DNA HYBRIDIZATION

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