

291c Physical and Chemical Modification of Aluminum Substrate to Improve the Sensitivity of Impedimetric Immunosensor for Detection of *Staphylococcus Aureus* Enterotoxin B

Changhoon Chai and Paul Takhistov

The physical and chemical properties of substrate for impedimetric immunosensors are crucial factors to determine the sensitivity for toxin detection since the substrate mediates antibody-antigen reaction to electrical signal. In this study, commercial aluminum, which is feasible for physical and chemical modification, was utilized as the substrate of impedimetric immunosensor for detection of *Staphylococcus aureus* enterotoxin B (SEB) instead of gold or silicon, currently used as the substrate. The technique to stabilize immunoglobulin (IgG), which is commonly used as the template for commercial antibody, onto aluminum surface was established and the improvement of sensitivity by nano-porous fabrication of aluminum substrate was demonstrated. Aluminum substrate was silanized with 3-aminopropyltriethoxysilane (APTES) to cross-link IgG to aluminum surface. The formation of APTES monolayer on aluminum surface was achieved by heating aluminum substrate at 65°C for 8hrs in 0.25% APTES solution. Well immobilized FITC conjugated IgG (FITC-IgG) could be seen through fluorescence microscope after immobilization of FITC-IgG. Then this technique was applied to immobilize anti-SEB on aluminum surface. Immobilized anti-SEB was quite stable to the electrical stress, thus impedance analysis of anti-SEB immobilized aluminum substrate was reproducible. Anti-SEB and SEB reaction on immunosensor surface resulted in the changes of impedance. The changes were dependent on the function of time and electrical frequency. At 31 kHz, the specific change of impedance to anti-SEB and SEB reaction was observed. The sensitivity of immunosensor was improved significantly by changing the physical properties of aluminum substrate. Aluminum substrate was anodized at 40V in 0.3% oxalic acid and well ordered nano-porous structures were observed through atomic force microscopy (AFM). The developed anti-SEB immunosensor, based on nano-porous aluminum substrate, could detect as less as 0.01ng/ml of SEB in 20 minutes.