

## Daily Amperometric Monitoring of Immunoglobulin E in a mouse whole blood: model of Ovalbumin induced asthma

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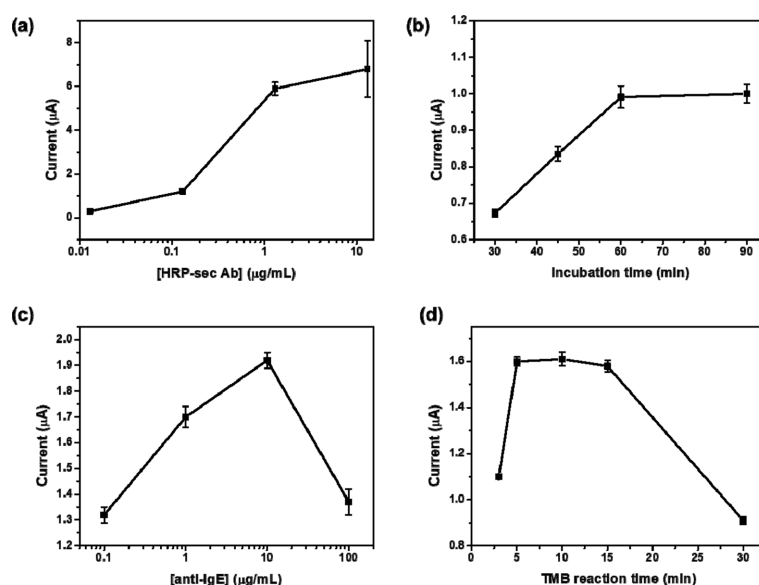
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**Figure S1.** Optimization of the step in the preparation of immunosensors for IgE (a) HRP secondary antibody concentration. (b) HRP secondary antibody incubation time. (c) capture antibody concentration. (d) TMB reaction time.

In case of HRP secondary antibody as shown in Figure S1, 1 μg/mL signal is similar of 10 μg/mL secondary antibody concentration. We assumed that HRP sec ab is saturated adsorbed to the electrode surface, so we choose 1 μg/ml concentration. The effect of incubation time is critical of the immunosensor, and we choose 60 min to optimize immunosensor. As shown the figure (b), 60 min is saturated so 60 min signal is similar with 90 min signal. The effect of anti-IgE (Capture antibody) is critical and we choose 10 μg/ml concentration. The signal is increased with increasing capture antibody concentrations. However, high concentration of capture antibody affects to decrease the electric signal. We assumed that excess amount of antibody is randomly adsorbed to the electrode surface and to hinder antigens binding site. Therefore, antigen cannot bind to the antibody and it leads to decrease the sensitivity (Hook effect). TMB reaction time is fixed to 5 min and signal is decreased with increasing reaction time. TMB is light sensitive and they lost their activity when light is exposed.

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