高感度ダニアレルゲン計測のための沈殿出力増幅式 SAW 免疫センサ

Precipitates-assisted SAW immunosensor for sensitive monitoring of mite allergen 医科歯科大¹, 文京学院大², 日本無線³

^Oワノタヤンスィティナット¹, 大石 紘希¹, 加藤 実里², 倉田 香菜子², 當麻 浩司¹, 吉村 直之³, 荒川 貴博¹, 谷津田 博美³, 金森 きよ子², 三林 浩二¹

Tokyo Medical and Dental Univ.¹, Bunkyo Gakuin Univ.², Japan Radio Co. Ltd.³,

^oWanotayan Sittinadh¹, Koki Oishi¹, Misato Kato², Kanako Kurata², Koji Toma¹,

Naoyuki Yoshimura³, Takahiro Arakawa¹, Hiromi Yatsuda³, Kiyoko Kanamori², Kohji Mitsubayashi¹ E-mail: m.bdi@tmd.ac.jp

[Introduction]

Inhalation of airborne HDM (house dust mite) allergens causes development of allergic diseases. A potentially effective method to avoid exposure to such airborne is an on-site monitoring system. As the developed allergen sampler had shown a low allergen transport efficiency to a sensor in the system, the sensor sensitivity became a subject to be solved. This study aims to enhance the sensitivity of a surface acoustic wave (SAW) immunosensor by utilizing precipitates while maintaining an ability of repeated measurement.

[Experimental]

As a SAW device detects viscosity and mass change nearby the sensing surface, it is expected that the sensor output is improved by increasing a total mass with precipitates, which is produced through a horseradish peroxidase (HRP)-catalyzed reaction. The sensor performances—signal amplification and reproducibility—are studied with three candidate substrates as the precipitate precursor, 4-chloro-1-naphthol (4CN), 3,3-diaminobenzidine and 3,3',5'5-tetramethylbenzidine. The SAW immunosensor was fabricated by a self-assembled monolayer of ORLA85 protein and polyethylene glycol (PEG)-thiol, followed by

immobilization of capture antibody (cAb). In the measurement of a mite allergen, *Dermatophagoides farina* (*Der f* 2), production of precipitates was induced by injecting the substrate and H_2O_2 to the sensor surface where HRP-conjugated detection antibody (HRP-dAb) bound to capture *Der f* 2. Finally, the sensor surface was regenerated by pH change with HCl (pH 1) or NaOH (pH 13) solution.

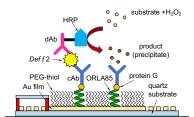


Fig. Schematic of a precipitate-assisted SAW immunosensor for *Der f2*.

[Results & Discussion]

Although all substrates showed poor regeneration with HCl, 4CN exhibited the highest reproducibility and a high regeneration rate with NaOH; therefore, 4CN was selected for the sensor development. The limit of detection (LOD) of the precipitate-assisted SAW immunosensor was 35 pg/mL, which was about 200-fold better than the previous SAW immunosensor without a precipitate. These results demonstrate that exploiting 4CN under the optimal conditions in the SAW immunosensor realizes sensitive monitoring of HDM allergens.