A sensing system of EDCs based on cell membrane model

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Objectives

The endocrine disrupting chemicals (EDCs) are believed to have ability to pass through cell membrane and to mimic or antagonize the effects of endogenous hormones or to disrupt the synthesis and metabolism of endogenous hormones and hormone receptors. In this study we aimed to establish a simple sensing system of EDCs. We focus on the EDCs' ability to pass through cell membrane and propose a novel sensing system of EDCs based on cell membrane model as recognition element. In this study, we immobilized cell membrane model complex on the surface of QCM and investigated frequency shift of the QCM/complex by nonylphenol which is one of well-known EDCs. As a cell membrane model, bR-liposome complex was utilized. Negatively charged. bR and positively charged liposome formed complex by an electrostatic force. The size of the complex was 1-10 μ m in diameter. bR-liposome complex proved to be degraded by EDCs and the phenomena showed the concentration dependence on EDCs. It was detected by OCM.

Immobilization of the complex on QCM

The complex was immobilized to alkanethiols by an electrostatic force. Three alcanethiols; mercaptopropionic acid (3C), 7-calboxy-1- heptanethiol (7C), 8-Amino-1-octanethiol, hydrocloride (8A) were used for immobilization of the complex on QCM. A QCM was immersed in alkanethiol solution for 1h. The QCM modified with alkanethiol was immersed in complex solution for 1h to immobilize the complex on QCM. It was estimated that the complex was immobilized on QCM, which was modified with 3C, 7C, and 8A, and the mass shift was calculated by the frequency shift. Average of the mass shift were 2000 to 4000 ng however the difference was not significant. We used 3C to immobilize the complex on QCM because QCM/3C/complex was the most stable and reproducible.

QCM/3C/complex responds to EDCs

Then we investigated frequency shift of the QCM/complex in the solution of nonylphenol. Nonylphenol was used as EDCs. The response of QCM/complex showed the apparent dependence on the concentration of nonylphenol. Linear relationship between the frequency shift and the concentration of nonylphenol was observed. And at ppb level, apparent dependence of the frequency shift was shown on the concentration of nonylphenol. The sensor responds at the range of 1.0ppb-1.0ppm nonylphenol. And the response time of the sensor was 40min- 2h.

Selectivity

To investigate the selectivity of the sensor, detergents that are not certified as EDCs such as lauric acid and TritonX were used (fig.3). The sensor QCM/complex did not respond to the detergents. The frequency shifts caused by these detergents were not increase but decrease and did not depend on the concentration of the detergents. The nonylphenol was shown that the QCM/complex respond to nonylphenol selectively.

Thus the QCM/complex is promising sensor for EDCs.

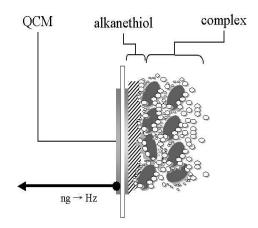


fig1. The construction of the sensor Gold electrode of QCM was modified with alkanethiol. And complex were immobilized onto the ionic groups of alkanethiol by an electrostatic force.

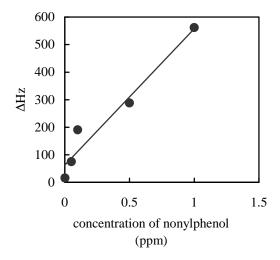


fig.2 The calibration curve of nonylphenol (ppm) with the QCM/3C/complex. The QCM was immersed in 1mM 3C solution for 1h at 20° C. The QCM decorated with alkanethiol was immersed in complex solution for 1h to immobilize the complex on QCM.

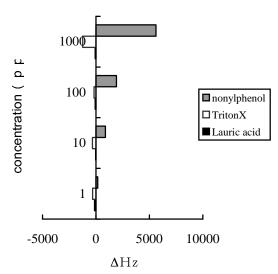


Fig.3 The comparison of frequency shift of nonylphenol with the detergents (TritonX and lauric acid).