



# Piezoelectric immunosensor with gold nanoparticles enhanced competitive immunoreaction technique for 2,4-dichlorophenoxyacetic acid quantification

Junhui Ding<sup>a</sup>, Zhe Lu<sup>a</sup>, Ruozhong Wang<sup>a</sup>, Guoli Shen<sup>b</sup>, Langtao Xiao<sup>a,\*</sup>

<sup>a</sup> Hunan Provincial Key Laboratory of Phytohormones and Growth Development, College of Bioscience and Biotechnology, Hunan Agricultural University, Changsha 410128, China

<sup>b</sup> State Key Laboratory of Chemo/Biosensing and Chemometrics, Hunan University, Changsha 410082, China

## ARTICLE INFO

### Article history:

Received 2 July 2013

Received in revised form

18 November 2013

Accepted 24 November 2013

Available online 1 December 2013

### Keywords:

Quartz crystal microbalance

2,4-Dichlorophenoxyacetic acid

Competitive immunoreaction

Gold nanoparticles

Herbicide

## ABSTRACT

The competitive immunoreaction technique for the detection of 2,4-dichlorophenoxyacetic acid (2,4-D) was described in this paper based on quartz crystal microbalance (QCM) with gold nanoparticles enhanced surfaces. The results showed the response of the sensor has a good linear relationship with 2,4-D concentrations in the range of 13.3–666.7 ng/mL with a detection limit at about 13.0 ng/mL. This work also provides a promising alternative approach for immuno-detection of other small molecules. The sensor could be regenerated under mild conditions simply by immersing the sensor into glycine buffer solution to break the antibody-antigen linkage. It was found that the proposed sensor could be reused at least nine runs without obvious loss of sensitivity.

© 2013 Elsevier B.V. All rights reserved.

## 1. Introduction

Phytohormones, trace signaling molecules synthesized in plants, play a significant part in the overall course of plant growth and development. About 10 categories of auxin, gibberellins, cytokinins, abscisic acid, ethylene, brassinosteroids, jasmonic acid, salicylic acid and strigolactone were found as naturally occurring phytohormones [1]. Phytohormones and plant growth regulators have long been a hotspot in biology and agriculture, with the research achievement in both fields contributed greatly to agriculture [2]. Because of the low content in plant (1–100 ng/g fresh tissue) [3], decomposability with heat, light and oxygen, it is difficult to analyze trace phytohormones efficiently and accurately. On the other hand, with the growing concerns about plant growth regulator residue in agricultural products, it is urgent to develop new techniques to detect plant growth regulators with high sensitivity and efficiency for food inspection [4–6].

2,4-dichlorophenoxyacetic acid (2,4-D) is an artificial plant growth regulator structurally and functionally similar to auxin [7]. It is a component of plant tissue culture medium at low concentrations ( $0.5 \times 10^{-6}$ – $1.0 \times 10^{-6}$  g/L); however, it can also

be used as herbicide for broadleaf weeds at high concentrations ( $1000 \times 10^{-6}$  g/L). In the last two decades, 2,4-D-centric artificial auxin like chemicals have been widely applied in agriculture. However, 2,4-D belongs to a group of chemicals known as phenoxy compounds, which are potentially toxic to humans, and easy to be accumulated in the human body [8,9]. Furthermore, it is of potential carcinogenicity and mutagenicity [10]. As a result, the development of new analytical methods for the determination of 2,4-D in different samples is currently a high-interest research area.

Several techniques for 2,4-D detection have been developed, such as gas chromatography (GC) [11], high performance liquid chromatography (HPLC) [12], gas chromatography mass spectrometry (GC/MS) [13], liquid chromatography mass spectrometry (HPLC/MS). However, these techniques are of flaws of complex operation, long time, low sensitivity (the detection limit of 2,4-D are about 200–400 ng/mL) and expensive apparatus [14,12,15]. Consequently, it is pressing to develop rapid and sensitive, simple and inexpensive detection method for 2,4-D. Immunosensor has been extensively applied in biotechnology, clinical diagnosis, environmental monitoring and food industry because of its high sensitivity, simple apparatus and easy operation. At present, the modern methods for 2,4-D detection involve electrochemical sensor [14], immunosensor [16] and biomimetic sensor [17]. Of the various kinds of immunosensors, great interest is attached to

\* Corresponding author. Tel.: +86 73184635261.

E-mail address: [langtaoxiao@phytohormones.com](mailto:langtaoxiao@phytohormones.com) (L. Xiao).

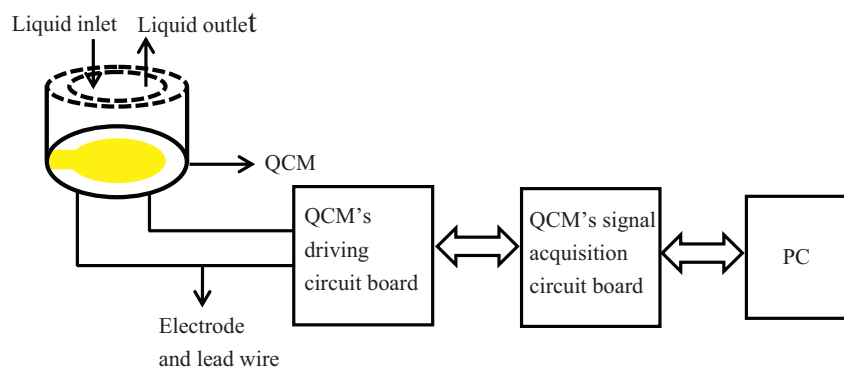


Fig. 1. Experimental schematic.

piezoelectric immunosensor due to its high sensitivity of mass-basis response and high specificity of immunoreaction. Furthermore, it is convenient, time saving, highly accurate, and easy to collect and analyze data online without impairing or contaminating the sample [18]. The quartz crystal microbalance (QCM), as a highly sensitive sensor, can detect ng-level changes in surface quality, but can not meet directly the inspection requirements for analyzing ultra-trace biochemical substances [19,20]. The transducer of the piezoelectric sensor is an oscillating quartz crystal device whose resonance frequency changes with the mass change according to the Sauerbrey equation [21]:

$$\Delta F = -2.26 \times 10^{-6} \frac{2F_q^2 \Delta M}{A}$$

where  $\Delta F$  is the change in frequency of the crystal,  $F_q$  is the fundamental resonant frequency of the crystal,  $\Delta M$  is the mass deposited on the electrode surface and  $A$  is the area of the coated crystal. In this experiment, QCM was prepared by competition in the immune response and nano-gold amplification to detect 2,4-D. The test result indicated that it is a new type of quick, simple and reusable piezoelectric immunosensor for 2,4-D detection. Fig. 1 is a schematic of the whole experiment.

## 2. Experimental

### 2.1. Reagents and apparatus

Cysteamine hydrochloride (Cys) was product of ACROS. Glutaric dialdehyde was purchased from Sigma. BSA-2,4-D conjugate was made by Hunan Agricultural University (Hunan, China). Anti-2,4-D antibody (mouse IgG, mIgG) was purchased from Beijing Biosynthesis Biotechnology Co., LTD (Beijing, China). Goat anti-mouse IgG (G-Anti-MsIgG) and bovine serum albumin (BSA) were products of Beijing Dingguo Biotechnology Company (Beijing, China). Chloroauric acid and trisodium citrate solution were purchased from Shanghai Chemical Reagents (Shanghai, China). Phosphate

buffer solution (PBS) of various pH was prepared by mixing 0.01 mol/L  $\text{Na}_2\text{HPO}_4$  and 0.01 mol/L  $\text{KH}_2\text{PO}_4$ . The piranha solution was a mixture of 98%  $\text{H}_2\text{SO}_4$  and 30%  $\text{H}_2\text{O}_2$  in a volume ratio of 7:3. Other chemicals were all of analytical grade. Doubly distilled water was used throughout the experiments.

The QCM (AT-cut, 9 MHz, gold electrodes) was obtained from Chenxing Radio Equipments (Beijing, China). To keep a steady oscillation in the solution, the QCM was sealed on one side with an O-trapped-ring of silicone rubber covered by a plastic plate that formed an air chamber isolated from aqueous solution. The detection was performed by setting the piezoelectric sensor in a laboratory-made reaction cell containing 300 mL sample solution. The resonance frequency was monitored with a quartz crystal analyzer (QCA922, The Princeton Applications Institute, U.S.A.), which is shown in Fig. 2; The experimental temperature was controlled with a thermostat (model CSS501, Chongqing Experimental Equipments, China). TCL-16A table-type high speed refrigerated centrifuge (Changsha Pingfan Instrument and Meter Co., Ltd.) and scanning electron microscope (SEM, JSM 6700F, JEOL Ltd., Japan) were also used.

### 2.2. Preparation of GNP-labeled secondary antibody

Gold nanoparticle (GNP) suspensions of average 8 nm in diameter were synthesized according to the reported method [22] with slight modifications. Glassware used in the experiment had been soaked in aqua regia, and then rinsed by ultrapure water (UPW). Briefly, 4.12 mL of 0.1%  $\text{HAuCl}_4$  solution mixed with 96 mL of UPW was heated to boil with gentle stirring in a 250 mL round flask. After 5 mL of 2% trisodium citrate solution was added and the color of the solution changed into blue in 2 min, and changed into claret-red in another 2 min, boiling was pursued for an additional 15 min. Then, the solution was stirred continuously until it was cooled to room temperature and was stored at 4 °C for gold nanoparticles-labeled goat anti-mouse IgG.

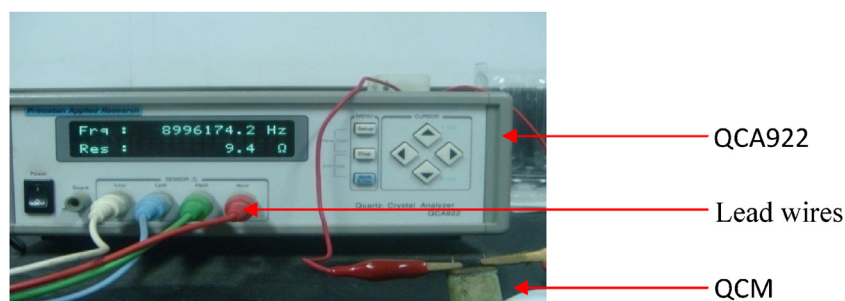


Fig. 2. Piezoelectric analyzer (QCA922).

Download English Version:

<https://daneshyari.com/en/article/740111>

Download Persian Version:

<https://daneshyari.com/article/740111>

[Daneshyari.com](https://daneshyari.com)