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Multichannel quartz crystal microbalance array: Fabrication, evaluation, application in biomarker detection

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ABSTRACT

A multichannel quartz crystal microbalance array (MQCM) with three pairs of gold electrodes was fabricated for detection of two biomarkers: acetone and nitric oxide (NO). The gold electrodes were deposited symmetrically on an AT-cut 10 MHz circular quartz plate using photolithography, sputtering, and lift-off technologies. The effect of gold layer thickness on MQCM performance was investigated and the optimized thickness was 101 nm. The simulation values of the electric parameters C_0, C_q, L_q, and R_q in the Butterworth–Van Dike equivalent circuit for the MQCM device were 97 pF, 1.3 pF, 1.05 mH, and 9.8 Ω, respectively. Simulation values were in the theoretical range, which indicated that the fabricated MQCM device had good resonance performance. Two types of nanocomposites, titanium dioxide–multiwalled carbon nanotubes and cobalt (II)phthalocyanine–silica, were synthesized as sensing materials. The sensing mechanism is based on coordination adsorption of target molecules onto the sensing material, resulting in a resonant frequency shift of modified QCM sensor. A linear range from 4.33 to 129.75 ppmv for acetone was obtained and one from 5.75 to 103.45 ppbv for NO.

Exhaled human breath contains a spectrum of endogenous chemicals and trace volatile organic compounds derived from metabolic products in blood by passive diffusion across the pulmonary alveolar membrane. Their concentrations are indicative of physiological conditions and respiratory diseases. For instance, breath acetone serves as a biomarker for diabetes and elevated levels of exhaled nitric oxide (NO) is related to various pulmonary diseases. Monitoring their trace level change in exhaled human breath is significant to early disease diagnosis. For diabetic patients under different conditions, acetone concentration is from 2.05 to 5.58 ppmv [1,2]. Here, ppmv means parts per million by volume. In healthy individuals, the value is in the range from 0.48 to 0.51 ppmv [3]. The correlation coefficient between breath acetone and blood sugar has been found to be in the range from 70% to 80% [4,5]. Meanwhile, the cut point of the fraction of exhaled NO (FE_{NO}) for asthma and other obstructive lung diseases is around 20–25 ppbv [6]. Here, ppbv means parts per billion by volume. Until now, existing noninvasive techniques for breath acetone and FE_{NO} analysis have been based on optical absorption spectrometry and gas chromatography–mass spectrometry [7–11], which are cumbersome, expensive, and not widely practiced in clinical settings. For early disease diagnosis, we need to develop a sensitive and noninvasive method for detecting breath acetone and FE_{NO} around 0.74 ppmv and 20 ppbv, respectively.

An array of electrodes fabricated on a single quartz wafer, which is called a multichannel monolithic quartz crystal microbalance (MQCM), has been developed and used in physical, chemical, and biological sensing since 1999 [12]. Because the size and spacing between QCMs are necessarily small, the miniaturization of QCM sensors is possible. MQCM device possesses many advantages, such as lower cost, less sample consumption, shorter diagnosis time, multiple analytes, and more portability. One advantage needs to be mentioned: that during measurement, potential effect factors such as temperature, humidity, and gas flow rate can be measured and

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subtracted from the signal QCM channel by using a blank QCM as a control at the same time. However, frequency interference between adjacent electrodes on a MQCM device needs to be considered. Several methods have been attempted to suppress the frequency interference [12,13]. For example, the MQCM can be designed to have different resonant frequencies using a deep reactive ion etching technique, which could make a single quartz plate with different quartz thicknesses [14]. Larger distances between channels of QCM sensor can also avoid strong interference. A simpler and more convenient method was to design the adjacent electrodes in appropriate positions on a single quartz crystal plate, which proved to produce negligible interference [15–17]. Only a few gas sensors based on MQCM have been reported. Zeng et al. [17] fabricated a symmetric MQCM device and modified QCMs with an assortment of sensing films: ionic liquids and poly(vinyl ferrocene). The modified MQCM was applied in the detection of ethanol, dichloromethane, and hexane. Palaniappan et al. [16] constructed a benzene and ethanol gas sensor based on a silica hybrid film that modified a four-channel quartz crystal microbalance array. Zampetti [18] reported a four-channel quartz crystal microbalance sensor array as a miniaturized electronic nose for analysis of several volatile organic compounds. In this work, a MQCM device was built up to sense biomarkers: acetone and nitric oxide. MQCM with three pairs of gold electrodes were fabricated in symmetric geometrical designs by photolithography, sputtering, and lift-off techniques. The sensing layers of titanium dioxide–multiwalled carbon nanotube (TiO_2–MWCNT) and cobalt phthalocyanine–silica (CoPc–silica) hybrid films were synthesized and characterized. They were deposited on respective QCM sensors to adsorb target gases at room temperature. During the adsorption process, shifts in the resonant frequency of the MQCM device were measured and analyzed. The mass sensitivity of a 10 MHz QCM is 27 ng/Hz. Combining the high sensitivity of MQCM with a room temperature sensing layer, a sensitive and noninvasive sensor for breath acetone and NO could be constructed at room temperature.

Experimental

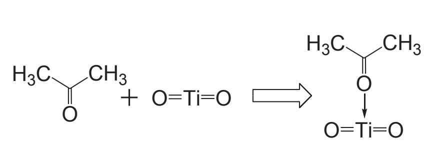
Sensing principle

Owing to piezoelectric effects, QCM resonate electromechanically in a thickness–shear mode. The shift in resonant frequency will result from external physical loading, including gravimetric and viscoelastic loading. In this work, two QCM sensors on a MQCM device were deposited with TiO_2–MWCNT nanocomposite and CoPc–silica hybrid film, respectively. The frequency shift (Δf_0) caused by gravimetric loading (Δm) obeys Sauerbrey's equation,

$$\Delta f_0 = -\frac{2f_0^2}{A\sqrt{\rho_Q \mu_Q}}\Delta m, \quad (1)$$

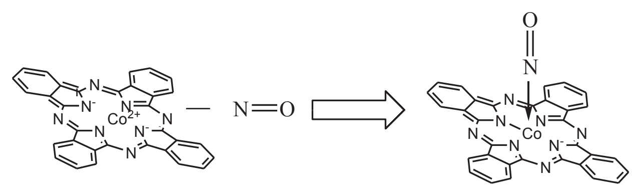
where f_0 is the resonant frequency, A is the piezoelectric active crystal area, and μ_Q and ρ_Q are the elastic modulus and density of quartz, respectively.

In TiO_2–MWCNT nanocomposite, there is chemical bonding between TiO_2 and –OH and –COOH groups on the surface of MWCNT, resulting in formation of Ti–OH bonds. So acetone adsorption on the TiO_2–MWCNT nanocomposite would depend on the coordination environment on the surface of the Ti site. Swelling of the nanocomposite caused by the adsorbed acetone molecules increases the hopping resistance among MWCNT. Coordination bonds will be formed between Ti atoms and oxygen atoms in acetone, as shown in



(2)

They are specific to oxygen. Cobalt phthalocyanine was adapted to adsorb NO by using an open d*z*2 shell configuration, which is one process of NO molecule coordination with the central cobalt atom with appreciable electron transfer from the central atom to the NO molecule [19]. The principle is shown in



(3)

After the sensing layer is adsorbed by the target gas molecules, the frequency shift of the signal QCM sensor can be obtained by subtraction from the control QCM sensor at the same time. The relationship between frequency shift and gas concentration can be set up.

Materials and chemicals

A 13 mm diameter, 166 μm thick, AT-cut 10 MHz circular quartz crystal was purchased from the International Crystal Manufacturing Company (Oklahoma City, USA). Carboxyl group functionalized multiwall carbon nanotubes (MWCNT, 95% purity) with outer diameter around 8 nm and length 0.5–2 μm were purchased from Nanostructure & Amorphous Materials Inc. (Houston, USA). Titanium tetra-isopropoxide, cobalt (II) phthalocyanine (β-form, CoPc), silica nanopowder with primary particle size 12 nm, and acetone were purchased from Sigma–Aldrich. All chemicals were of analytical grade and used without further purification.

MQCM device fabrication

Both sides of the quartz crystal plate were sputtered with a chromium (Cr) adhesion layer and a gold (Au) layer. The pattern schematic of the gold electrodes is shown in Fig. 1a. Three pairs of circular gold electrodes were placed in a symmetrical design. The electrode thickness is around 100 nm. The diameter for one gold electrode is 2.54 mm, and the center-to-center spacing between two electrodes is 6.22 mm. The MQCM device (Fig. 1b) was fabricated through processes of photolithography, sputtering, and lift-off, as shown in Fig. 1c.

Nanocomposite synthesis

Titanium dioxide (TiO_2) and TiO_2–MWCNT nanocomposite were prepared by a sol–gel method, using titanium tetra-isopropoxide [$Ti(C_3H_6OH)_4$] as the precursor, anhydrous ethanol as the solvent, and acetic acid (HAc) as the stabilizer. First, specific amounts of $Ti(C_3H_6OH)_4$, ethanol, and HAc (V/V = 10:30:8) were mixed at room temperature with vigorous stirring for 1 h. Second, a mixture of 7.5 ml ethanol and 0.7 ml H_2O was added drop by drop to the solution to initiate the hydrolysis reaction. The solution was then stirred continuously for 2 h to form a sol and aged 2 days to produce the corresponding gelatin. The preparation of TiO_2–MWCNT nanocomposites is basically that described for TiO_2. The different step was that prior to the addition of the titanium