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THE STUDY ON NOVEL MICROELECTRODE ARRAY CHIPS FOR THE DETECTION OF HEAVY METALS IN WATER POLLUTION

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Qualitative and quantitative analysis of trace heavy metals in aqueous environment are rapidly assuming significance along with the rapid development of industry. In this paper, gold microelectrode array (MEA) plated with mercury film was used for simultaneous voltammetric detection of zinc, cadmium, lead and copper ions in water. The electrochemical behavior and the actual surface area of the MEA were investigated by cyclic voltammetry in $K_3[Fe(CN)_6]$. Electrochemical impedance spectrum (EIS) was utilized to examine the deposition of mercury on the electrode surface. Based on anodic stripping voltammetry, mercury film−Au MEA was applied to the detection of heavy metals in artificial analyte, where good calibrate linearity was obtained for cadmium, lead and copper ions, but with zinc exhibiting poor linearity.

Keywords: Microelectrode array (MEA) chip; heavy metals detection; water pollution.

1. Introduction

Contamination of heavy metals is a serious problem in recent years and is caused by mining, exhaust emission, waste water irrigation, and use of heavy metal products, etc. Along with the rapid development of the economy and large-scale industry, the extent of heavy metal pollution has increased in air, soil and water, with the contamination of water particularly assuming great importance. The usual concentration ranges for zinc, lead, cadmium and copper present in water are $10-500\,\mu g/L$, $0.5-15\,\mu g/L$, $0.5-50\,\mu g/L$ and $2-50\,\mu g/L$, respectively. These concentrations can be harmful to human health because of the processes of methylation, bioaccumulation and biomagnification. Thus, it is crucial to develop the ability to detect heavy metals in aqueous environment.[1−3]

Traditional heavy metals monitoring approaches are based upon discrete sampling methods followed by laboratory analysis. The most common approach is the spectroscopic analytical technique such as atomic absorption (AA), induced coupled plasma (ICP) and fluorimetry.[4,5] These approaches do not improve our understanding of the natural processes of the chemical species behavior, such as diffusion and migration. At the same time, the detection error caused by the long-term storage of water samples is

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also questionable. Furthermore, the instruments are expensive, time-consuming and require significant maintenance and operator expertise. Most importantly, these approaches do not provide higher resolution data compared to the electrochemical method. In view of the limitations of discrete sample collection and subsequent laboratory analysis, electrochemical sensors are efficient techniques for fast, low-cost, real-time heavy metal ions detection.[6,7]

With the rapid development in microfabrication technology, microelectrode arrays (MEA) are normally used as the sensitive device in electrochemical analysis. The radius of a microelectrode always ranged from several micrometers to hundreds of micrometers, facilitating the size requirement for equipment. Compared to the traditional electrodes, microelectrodes possess attractive features, such as high mass transport, great current density, small time constant, low ohmic drop, which are advantageous for electrochemical analysis performed in high resistive solution or even without supporting electrolyte.[8−14] However, the extremely low current values can be problematic. This is overcome by the use of an array of microelectrodes, yielding enhanced current signal while at the same time maintaining the electrochemical characteristics as the individual microelectrode.

In our past studies, zinc, cadmium, lead and copper ions were simultaneously detected by large traditional glassy carbon electrode (diameter 3 mm), and preliminary study was undertaken to determine lead and copper ions using gold nanoband electrode without mercury plating.[15] The size of the glassy carbon electrode is big, and the sensitivity is low compared to microelectrode; moreover, the gold nanoband electrode without the mercury film cannot detect zinc and cadmium ions. This paper proposed mercury film−Au MEA, thereby realizing the simultaneous detection of zinc, cadmium, lead and copper ions in water. The electrochemical behavior of the microelectrode, the extent of mercury plating and the long-term stability were analyzed, and heavy metals were detected by stripping voltammetry.

2. Experimental Procedure

2.1. *Apparatus and reagents*

CHI1030 electrochemical analyzer was used for all electrochemical studies. All experiments were performed in a three-electrode system, where the working electrode was the Au-MEA, and the auxiliary electrode was a platinum wire, with an external Ag/AgCl reference electrode. A ground electrode was used to minimize the electrical noise as much as possible.

Solutions were prepared using Milli-Q (Millipore, USA) water (18.2 MΩ resistivity). Standard solutions of Zn, Cd, Pb and Cu were prepared by appropriate dilution of the stock solutions (1000 mg/L, atomic absorption standard solutions, Aldrich) using 0.5 M KCl electrolyte, and acidified to pH 4 with HCl. The concentration of the mercury solution was 30 mg/L, dissolved in 1% HNO_3. KCl, HCl, HNO_3, H_2SO_4 and PBS were all guaranteed reagents.

2.2. *Electrode preparation*

The gold MEA was fabricated by standard microfabrication technology. The substrate was 4-inch silicon wafer (N-type, $\langle 100 \rangle$, 250 μm thickness, ∼7−20 Ω·cm resistivity), a layer of SiO_2 was growing onto the surface of the wafer by dry oxidation. Then gold (250 nm thickness) was sputtered after a chromium adhesion layer (100 nm) deposited. Polyimide was used as the insulate layer and then the photoresist was spun and patterned. Following fabrication, the wafers were diced and the electrode arrays were packaged on printed circuit boards (PCB) with silver epoxy. The whole chip of Au-MEA was $2 \times 2\,cm^2$, consisted of 4×4 individual microelectrodes, with the single electrode radius 10 μm separated by a 45 μm gap. Figure 1(a) depicts Au-MEA under optical microscope and Fig. 1(b) is a photograph of a packaged die. Before using, the residual photoresist on the electrode surface must be washed with acetone.

2.3. *Measurement of actual surface area*

The determination of the real surface area of the electrode is very important for calculation of various parameters characterizing electrochemical processes. According to the Gouy−Chapman theory, the differential charge of the redox peak in the cyclic linear sweep was used to calculate the actual surface area of each microelectrode, for further estimating the roughness factor of the electrode.[16−18] Cyclic voltammograms were made between −0.4 and −0.8 V in 2 mM $K_3[Fe(CN)_6]$ in 1 M KCl electrolyte at 10 mV/s.
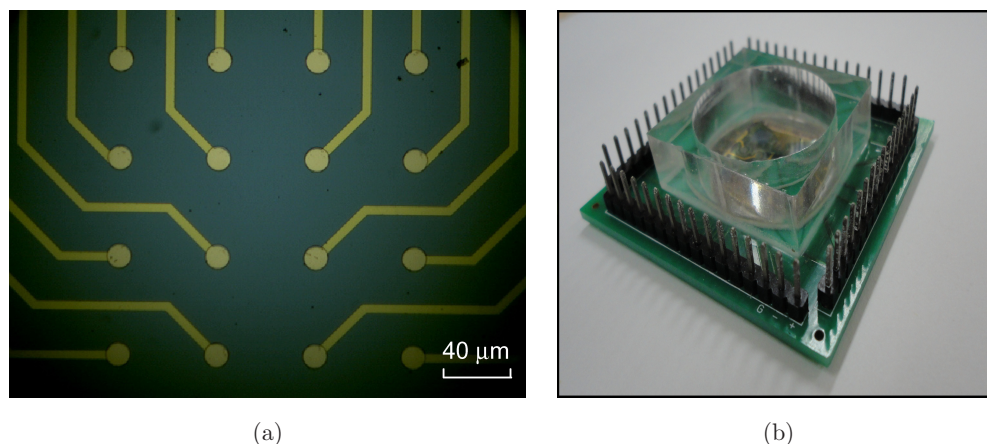
Fig. 1. (a) The picture of Au microelectrode array under optical microscope and (b) photograph of a packaged die.

2.4. *Mercury plating and analysis*

Before the detection of heavy metals in water, the surface of the electrode needs to be plated with mercury. Linear scanning voltammetry was applied to plate mercury, with the potential changing from -1.0 to -0.2 V, and the deposition potential was -1.0 V, deposition time was 60 s. Electrochemical impedance spectrum (EIS) was utilized to observe the surface of the electrode before and after plating mercury. The analysis was made in PBS solution with pH $= 4$, with the scan frequency changing from 1 KHz to 100 KHz, and the amplitude was 0.005 V.

2.5. *Voltammetry detection of heavy metals*

Each measurement consisted of three steps: Conditioning of the electrode and an analytical and a background scan. The background was subtracted from the analytical scan. Differential pulse anodic stripping voltammetry (DPASV) measurements were made in the sample solutions with 0.5 M KCl as the supporting electrolyte. The deposition time was 120 s, while the preconcentration potential was -1.55 V; the analysis then began from -1.4 V to -0.1 V with the step potential 0.004 V.

3. Results and Discussion

3.1. *Surface area of the electrode*

Figure 2 showed the cyclic voltammogram of an individual Au microelectrode in 2 mM $K_3[Fe(CN)_6]$ in 1 M KCl solution at 10 mV/s. Oxidized current reached the maximum value i_P^{ox} at about 0.3 V, and
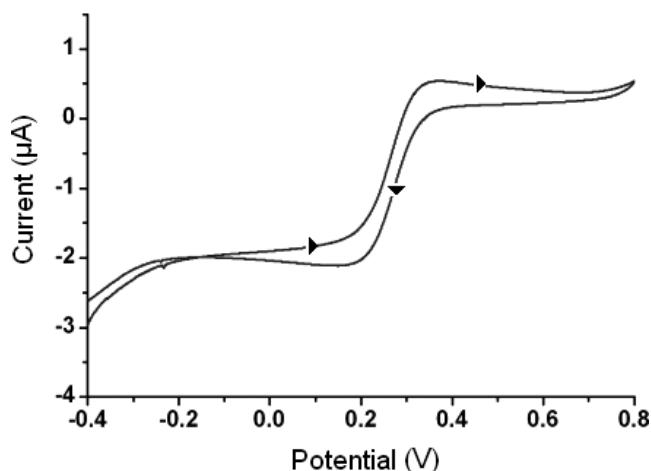

Fig. 2. Cyclic voltammogram of single Au electrode in 2 mM $K_3[Fe(CN)_6]$ in 1 M KCl solution at 10 mV/s.

i_P^{ox} was 1.6 μA. In the reverse sweep, the reduction current peak i_P^{red} appeared at 0.2 V, and the peak current was 1.5 μA. So $|i_P^{ox}/i_P^{red}| = 1.07$, this value was close to those expected for an electrochemically reversible process. For an reversible linear scan, the peak current i_P can be obtained by the Randles–Sevcik equation[19]:

$$i_P = 2.68 \times 10^5 n^{2/3} A D^{1/2} v^{1/2} c^0, \qquad (1)$$

where n is the electrons transferred, A is the actual surface of the electrode, D is the diffusion coefficient (the value was 0.76×10^{-5} cm^2/s), v is the scan rate, c^0 is the concentration of the reactant. The actual surface area of the electrode A_{real} can be calculated from Eq. (1), and the ratio A_{real}/A_{geom} is the roughness factor R_f, where A_{geom} is the geometrical area of the electrode. The roughness of our individual gold microelectrode was 1.18, which showed that the

actual surface area of the electrode was larger than the geometrical area. Smooth electrodes have a higher ratio of faradaic to charging current because the faradic current is directly proportional to the geometrical area while the charging current is directly related to the actual surface area. Our electrode required further polishing procedure to make the surface smooth enough.

3.2. *EIS analysis*

Figure 3 illustrated the electrochemical impedance spectrograms of Au-MEA before and after plating mercury and used for five times. The impedance of the electrode after plating mercury was smaller than that of before plating mercury, which may be caused by the increased surface area. The impedance remained stable after using five times, which showed that the mercury film might adhere to the electrode surface tightly, and the mercury film-Au MEA can be used reproducibly. After each measurement, $+0.5$ V potential was applied to clean the residual mercury film.

3.3. *Trace heavy metals detection*

Figure 4 was the differential pulse stripping voltammograms of Zn, Cd, Pb and Cu ions. According to the Nernst−Planck equation, the corresponding stripping current of each heavy metal is linear with its concentration. The concentration of these heavy metals in the artificial solution was $50 \, \mu$g/L, $5 \, \mu$g/L, $10 \, \mu$g/L and $10 \, \mu$g/L, respectively, and the volume
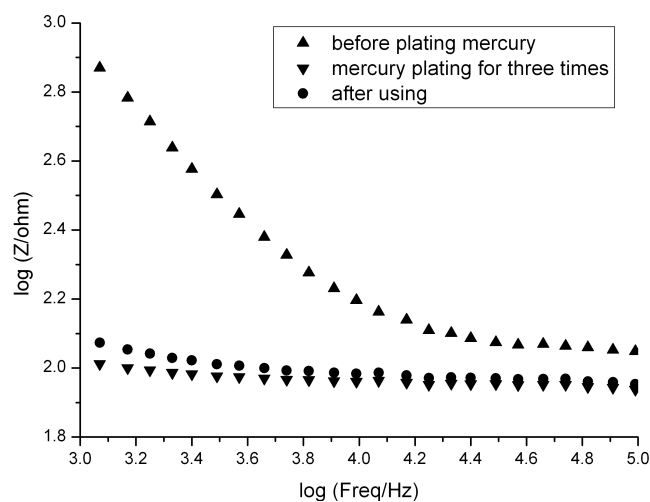

Fig. 3. Ac impedance spectrograms of Au-MEA before and after plating mercury and after using five times.
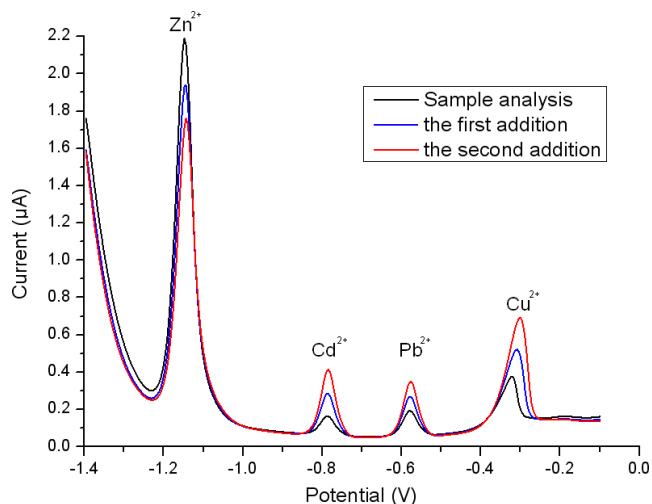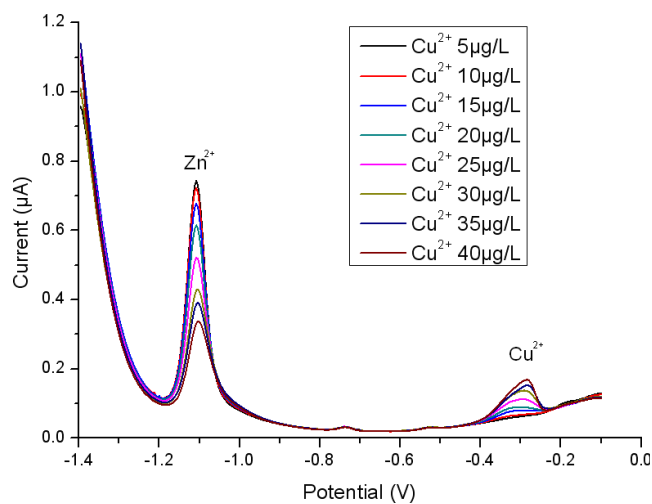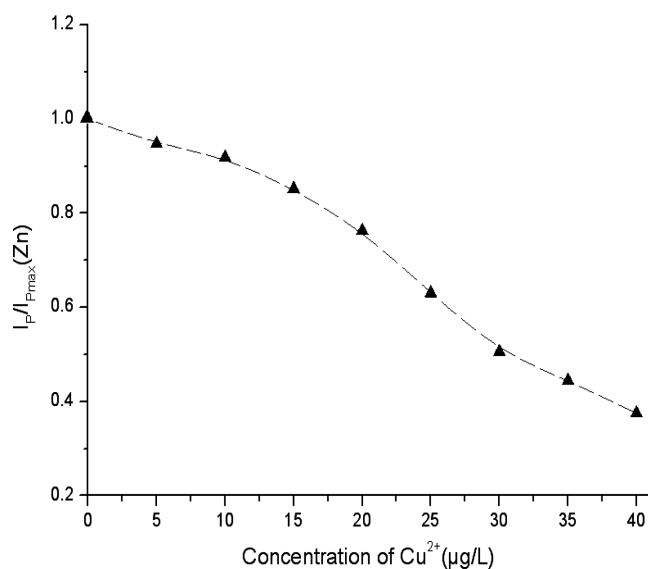

Fig. 4. Stripping voltammograms of Zn, Cd, Pb and Cu ions.

of the sample solution was 2 mL. The standard addition method was used to obtain the concentration gradient. The concentrations of the four heavy metals applied for standard addition were all 2 mg/L, and in each addition calibration, the addition volume were $10 \, \mu$L, $4 \, \mu$L, $5 \, \mu$L and $5 \, \mu$L, respectively. In these voltammograms, the background had been subtracted from the analytical curves. It could be seen that, Zn^{2+}, Cd^{2+}, Pb^{2+} and Cu^{2+} can be easily separated from each other in the voltammograms and the peaks of the four ions were stable and sharp. The concentration linearity of Cd^{2+}, Pb^{2+} and Cu^{2+} were well enough with the calibration curve correlation coefficient all greater than 0.95. The detection linear ranges for cadmium, lead and copper were $0.3−200 \, \mu$g/L, $1−200 \, \mu$g/L and $1−300 \, \mu$g/L, respectively. And the detection limits are $0.1 \, \mu$g/L, $0.5 \, \mu$g/L and $0.3 \, \mu$g/L, respectively. However, the peak currents of Zn^{2+} gradually decreased while the concentrations increased. This may be caused by the unchanging concentration of Zn^{2+} coming from the inaccurate addition while the baseline of the voltammograms drifted in each scanning, or may be affected by the formation of copper−zinc intermetallic compounds[20] along with the copper addition.

For further study of the peak currents of zinc, the voltammograms of zinc and copper in the same solution were utilized for the analysis of the copper−zinc intermetallic compounds. Figure 5(a) showed the stripping voltammograms of Zn and Cu ions in the same solution. The concentration of Zn was a constant value of $10 \, \mu$g/L, while the concentration of Cu gradually increased from 5 to $40 \, \mu$g/L.

(a)

(b)

Fig. 5. (a) Stripping voltammograms of Zn and Cu ions when the concentration of Zn was $10\,\mu g/L$, and the concentration of Cu increased from 5 to $40\,\mu g/L$, (b) depression of $I_p[\mathrm{Zn}^{2+}]$ caused by the addition of Cu^{2+} to the analyte solution.

The peak currents of zinc observed to decrease along with the addition of copper. Figure 5(b) illustrated the normalization of the depression of the peak current of zinc $I_p[\mathrm{Zn}^{2+}]$ caused by the addition of Cu^{2+} to the analyte solution. At a 1:1 zinc:copper ratio, I_p was reduced to 90% compared to its peak current $I_{p\,\mathrm{max}}$ without copper, and to one half at a 1:3 ratio. In Fig. 4, the initial zinc:copper ratio was 5:1, and the ratio changed to 7:2 after addition. The concentration of zinc was larger than that of copper, excluding the possibility of the Zn−Cu intermetallic

compounds effect. Besides, the concentration of zinc is several times that of copper in the natural water, so the problem rising from the copper−zinc intermetallic compounds can be neglected in the actual detection. Otherwise, this effect can be reduced by spiking the analyte solution with gallium ions,[21] and regression modeling methods such as partial least square regression or artificial neural network method[22] can also be used for calibration. Therefore, decrease in the stripping peak currents may be mostly caused by the inaccurate addition and the shift of the baseline.

3.4. *Long-term stability of the Au-MEA*

Stability of the electrode is important when it is to be used for long-term *in situ* monitoring in natural waters. After further use of the electrode, the baseline of the analytical voltammograms always shift and the width of the current peaks will become larger. Figure 6 showed the continuous measurements over 6 h of $30\,\mu g/L$ Cu^{2+} in the same solution detected by the same Au-MEA. The peak currents were stable with 1 h interval, and the standard deviation was 1.9%, which confirms that the stability of electrode is good enough for the long-term detection.
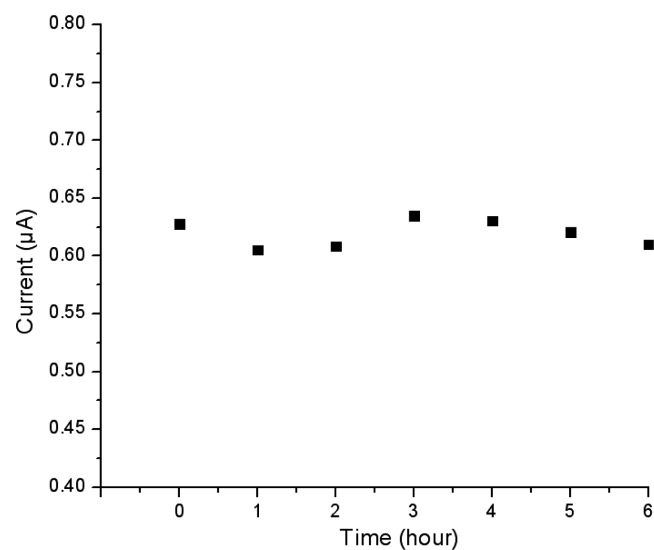

Fig. 6. Continuous measurements over 6 h of Cu^{2+} in the same solution detected by the same Au microelectrode array.

4. Conclusion

Gold MEA plated with mercury film was proposed for simultaneous voltammetric detection of zinc, cadmium, lead and copper ions in water. The deposition of mercury on the electrode surface was validated by the EIS method, and the mercury was prepared successfully and can be continuously used. Zinc, cadmium, lead and copper can be detected using anodic stripping voltammetry, and the linearity between stripping current and concentration of cadmium, lead and copper was good. Because of the analytical baseline excursion and the inaccurate addition, the peak currents of zinc decreased on increasing its concentration. In conclusion, this paper demonstrated that the mercury plated MEA can be applied *in situ*, on line and low-cost detection of heavy metals due to their sensitivity, reproducibility, fast and simultaneous measurement of various elements.

Acknowledgments

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