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Differential Pulse Anodic Stripping Voltammetric Determination of Lead with Heparin Modified Electrode

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A novel differential pulse anodic stripping voltammetry for the determination of trace amounts of lead, using a biomacromolecule heparin drop-coated modified glassy carbon electrode, has been described. Pb^{2+} was deposited on the surface of a heparin-modified electrode at −1.0 V (vs. SCE) via forming Pb^{2+}-heparin and subsequent reduction at the electrode. In the following step, Pb-heparin was oxidized, and voltammograms were recorded by scanning the potential in a positive direction. Conditions were optimized with respect to the pH of the medium, the mass of drop-coated heparin, accumulation potential and accumulation time. The peak current was proportional to the Pb^{2+} concentration in the range of 2.0×10^{-9} to 7.0×10^{-7} mol/L. The detection limit was 3.0×10^{-10} mol/L. The relative standard deviation was 4.83% for 1.0×10^{-8} mol / L Pb^{2+} (n=10). The developed method has been applied to the determination of Pb^{2+} in water samples with satisfactory results.

Keywords heparin, chemically modified electrode, lead, differential pulse anodic stripping

Introduction

Overall exposure to lead is of public health concern because of several hazardous effects that may occur to human beings. Lead poisoning may provoke irritability, anorexia, malaise and headache. Intoxication progress may lead to attacks of abdominal pain until coma and death.[1] The determination of trace lead in variety of environmental samples is of great importance since lead is recognized as a cumulative poison to animals and humans. There is a constant demand for improved analytical methods for the sensitive and selective determination of Pb^{2+} in environmental, biological and medical fields. Among the various methods used for the lead determination, electrothermal atomic absorption spectrometry is perhaps the most accepted technique.[2,3] On the other hand, anodic stripping voltammetry (ASV) is one of the most sensitive methods for the determination of trace amounts of numerous ions because of its remarkably low detection limits.[4] Other advantageous features of ASV include the relatively low-cost instrumentation and the capability for simultaneous multi-element determination. In addition, the possibility for portable and compact instruments for stripping analysis and their low power needs make them attractive for on-site monitoring of trace metals.[5]

Mercury-based electrodes, especially mercury-film electrodes, have been widely used in anodic stripping voltammetry for lead determination.[4,6,7] The formation of an amalgam enables the analyte to be accumulated in the mercury film, thus providing the stripping with high sensitivity and reproducibility. However, because of the toxicity of mercury, it is important to develop mercury-free electrodes, especially disposable electrodes, for the voltammetric stripping determination of lead. Chemically modified electrodes (CMEs) have been recognized as one desirable alternative. They can be fabricated easily and conveniently, and have some unique advantages in enhancing selectivity and sensitivity due to the chemical nature and microstructure of the modified electrode surface. Therefore, CMEs have been widely used in analytical chemistry.[8-10]

In recent years, various chemically modified electrodes have been used to determine lead. Many substances such as chitosan,[11] moss,[12] quercetin,[13] *N-p*-Chlorophenylcinnamohydroxamic acid,[14] 1-(2-pyridylazo)-2-naphthol,[15] dibenzo-18-crown-6 and cryptand,[16] lichen,[17] and 2,2'-dipyridyl-2,4-dioxybenzoic acid[18] have been used to fabricate the CMEs for the determination of lead. However, most of these modifiers were used to produce chemically modified carbon paste electrodes (MCPEs). Some of the MCPEs require additional steps for their preparation[12,17] or require a medium exchange,[14] while the 1-(2-pyridylazo)-2-naphthol and Nafion MCPEs,[19] and the moss MCPEs require longer accumulation time.[12] The accumulation times for the two MCPEs are 8 and 15 min, respectively. The sensitivities for some MCPEs are lower. For example, the detection limits of the lichen, dibenzo-18-

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crown-6 and cryptand modified electrodes are 2.0×10^{-5}, 1.0×10^{-6} and 5.0×10^{-6} mol / L, respectively.[16,17] Furthermore, some of these methods have not been applied to the determination of lead in real samples.[17,19] Recently, the determination of lead using a drop-coated modified electrode has been reported. Usually, the preparation of drop-coated modified electrode was easy and the sensitivity of the lead determination was high.[15,20] To our knowledge, there is no report about the determination of Pb^{2+} using heparin as drop coating on the glassy carbon electrode.

Heparin (Hep) is a polysaccharide belonging to the glycosaminoglycan family. Its structure is shown in Figure 1.[21] It can bind with Pb^{2+}.[22] In the present study, we explored here the possibility of drop coating the chelating agent, heparin, on a glassy carbon electrode for the determination of Pb^{2+} in aqueous samples. This article presents a detailed description of our studies.

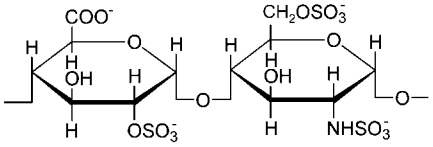

Figure 1 Structure of heparin.

Experimental

Apparatus

A CHI 660A Electrochemical Workstation (CHI Instruments) was used for electrochemical measurements. A conventional three-electrode system was employed with a heparin modified glassy carbon electrode (3.0 mm in diameter) as the working electrode, a platinum wire as the auxiliary electrode, and a saturated calomel electrode (SCE) as the reference electrode. All potentials reported in this paper were referenced to the SCE. Scanning electron micrographs (SEM) were taken with a PHILIPS XL 30 ESEM scanning electron microscope (voltage: 20.0 kV, magnification: 500).

Reagents

A 1.0×10^{-2} mol / L Pb^{2+} aqueous stock solution was prepared by dissolving lead nitrate in water. The working solutions were prepared by diluting the stock solution before being used. Heparin sodium solution (1 mg/mL) was prepared by dissolving 0.1000 g of heparin sodium reagent (160 IU / mg, Shanghai Chemical Reagent Plant) in water and diluting to a 100 mL volume. HAc-NaAc buffer was used to control the pH. All other chemicals used were of analytical-reagent grade. All of the solutions were prepared with deionized water. The water was prepared using MilliQ equipment (Millipore Corp.).

Preparation of heparin modified glassy carbon electrode

Prior to each experiment, the glassy carbon electrode was first polished with 0.05 μm alumina slurry using a polishing cloth to mirror smoothness, cleaned with ultrasonic waves in 1∶1 aqueous solution of nitric acid, and then sonicated in ethanol solution. After being rinsed with water, the glassy carbon electrode was dried under an infrared lamp. The heparin modified glassy carbon electrode (HEPMGCE) was prepared by coating 4 μL of 5.0×10^{-2} mg / mL heparin on the dry electrode with a microsyringe. Then the electrode was placed under an infrared lamp to evaporate the solvent. Before measurement, the modified electrode was rinsed with deionized water.

Determination of Pb^{2+}

A 10 mL volume of the solution containing an appropriate concentration of Pb^{2+} and HAc-NaAc buffer (pH=5.2) was transferred into an electrochemical cell for accumulation of Pb^{2+} at −1.0 V in stirring solution for 4 min. After standing for 30 s, the potential was then scanned from −1.00 to −0.20 V by differential pulse stripping voltammetry with a scan rate of 40 mV/s. The peak height was measured at –0.51 V. After each electrochemical measurement, the electrode was scanned for five times from −1.00 to −0.20 V by linear potential sweep voltammetry in pH=5.2 HAc-NaAc buffer to clean the previous deposits.

Results and discussion

Voltammetric characteristics of Pb^{2+} on the HEPMGCE

After Pb^{2+} was accumulated on the HEPMGCE and the GCE under the same condition, the cyclic voltammograms of the two electrodes are shown in Figure 2. A big anodic peak appeared at −0.50 V and a more poorly defined cathodic peak appeared at about −0.66 V for both electrodes. However, the peak current, especially the anodic peak current was increased obviously

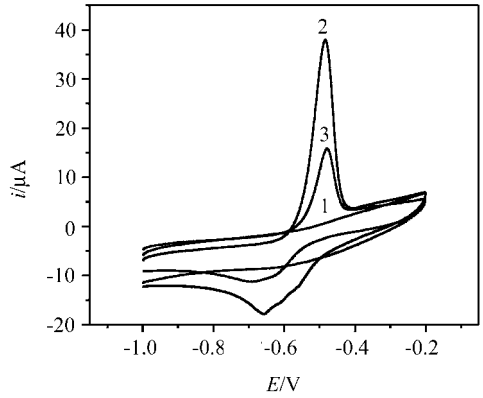

Figure 2 Cyclic voltammograms for the HEPMGCE (1) without Pb^{2+} accumulation and (2) with Pb^{2+} accumulation and (3) unmodified glassy carbon electrode with lead electrodeposition from acetic acid-sodium acetate buffer (pH=5.2). Conditions: Pb^{2+} concentration, 2.0×10^{-6} mol/L; accumulation potential, −1.0 V; accumulation time, 4 min; quiet time, 30 s; scan rate, 400 mV/s.

when the HEPMGCE was used. There was no redox peak in the absence of Pb^{2+} at the HEPMGCE in the same condition. These experimental results indicated that the HEPMGCE could improve greatly the sensitivity of the determination of Pb^{2+}. In view of the results obtained, the anodic peak was chosen for lead quantification.

A possible mechanism for the HEPMGCE reaction was proposed as follows:

accumulation:

$$(Pb^{2+})_{solution} + (Heparin)_{surface} \rightarrow (Pb^{2+}\text{-Heparin})_{adsorption} \quad (1)$$

reduction:

$$(Pb^{2+}\text{-Heparin})_{adsorption} + 2e \rightarrow (Pb^{0}\text{-Heparin})_{adsorption} \quad (2)$$

stripping voltammetry:

$$(Pb^{0}\text{-Heparin})_{adsorption} \rightarrow (Pb^{2+})_{solution} + (Heparin)_{surface} + 2e \quad (3)$$

In order to confirm the electrochemical process, the SEM photographs of the GCE and HEPMGCE under different conditions were taken and are shown in Figure 3. The SEM photograph of the bare GCE show that the GCE surface is very smooth. When the GCE was modified with heparin, heparin was distributed with graininess on the modified electrode surface. After Pb^{2+} was deposited on the surface of the HEPMGCE, many new and great grains were displayed on the surface of the HEPMGCE. But, after lead was stripped, the number of the grains was obviously decreased. The results of the SEM indicated deeply that the mechanism proposed above was reasonable.

Differential pulse anodic stripping voltammetry

The differential pulse anodic stripping voltammetry (DPASV) is shown in Figure 4. Systematic studies of the various experimental and instrumental parameters that affected the DPASV response were carried out in order to optimize the experimental conditions.

Effects of electrolytes and solution pH values

Various supporting electrolytes, such as HCl, NH_4Cl, HOAc-NaOAc buffer and Britton-Robinson (B-R) buffer were investigated. It was found that a well-defined and sensitive anodic stripping peak current appeared at -0.51 V when HOAc-NaOAc buffer was used as the electrolyte. The effect of pH of HOAc-NaOAc buffer on the anodic stripping peak current of the HEPMGCE was investigated. The relationship between the anodic stripping peak current and the pH value of HOAc-NaOAc buffer is shown in Figure 5A. It was found that an optimum pH range existed in pH 5.0—5.4, where a maximum stripping peak current could be obtained. Therefore, the HOAc-NaOAc buffer (pH 5.2) was selected as the supporting electrolyte.

Effects of accumulation potential and time

The dependence of the differential pulse anodic stripping peak current on the accumulation potential was examined over the potential range of -0.7 to -1.4 V. Figure 5B shows the effect of accumulation potential on the stripping peak current. The maximum response for lead occurred at the accumulation potentials equal to, or more negative than -1.0 V. Therefore, -1.0 V was chosen as the accumulation potential in our measurement.

Shown in Figure 5C is the plot of the differential specific accumulation period, 240 s, the peak current pulse lead stripping peak current of the HEPMGCE with the accumulation time. At first, the peak current increases with the accumulation time. However, after a tends to level off, illustrating that adsorption equlibrium is achieved. An accumulation time of 240 s was used for further studies.

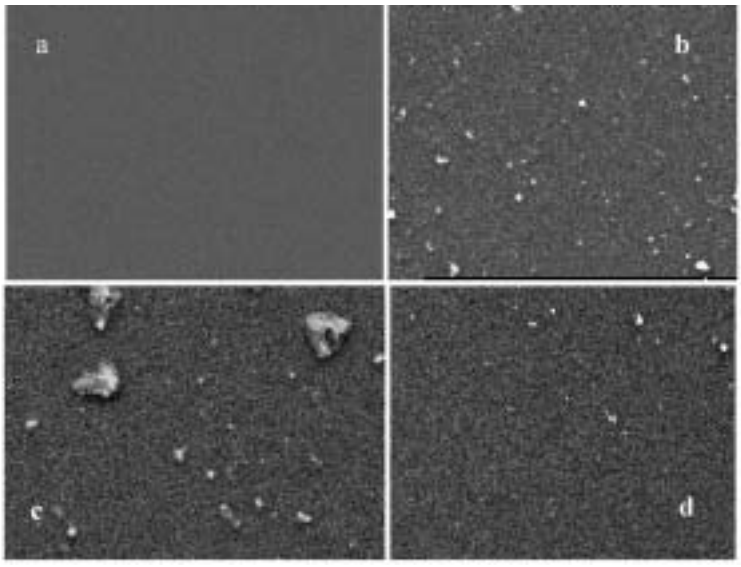

Figure 3 Scanning electron micrographs (×500) of (a) the surface of glassy carbon electrode, (b) the surface of HEPMGCE, (c) electrodeposition lead on the HEPMGCE and (d) the HEPMGCE after lead was stripped

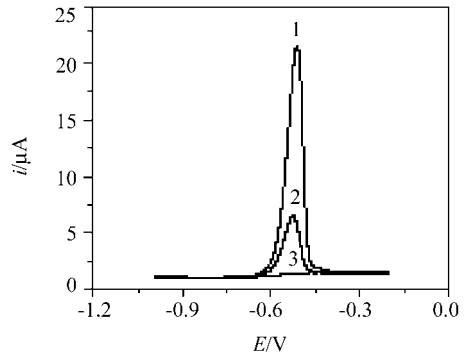

Figure 4 Differential pulse stripping voltammogram at scan rate of 40 mV/s. 1: acetic acid-sodium acetate buffer, containing 5.0×10^{-7} mol / L Pb^{2+}, HEPMGCE; 2: acetic acid-sodium acetate buffer, containing 5.0×10^{-7} mol/L Pb^{2+}, GCE; 3: buffer only, HEPMGCE. Other conditions were the same as those in Figure 2.

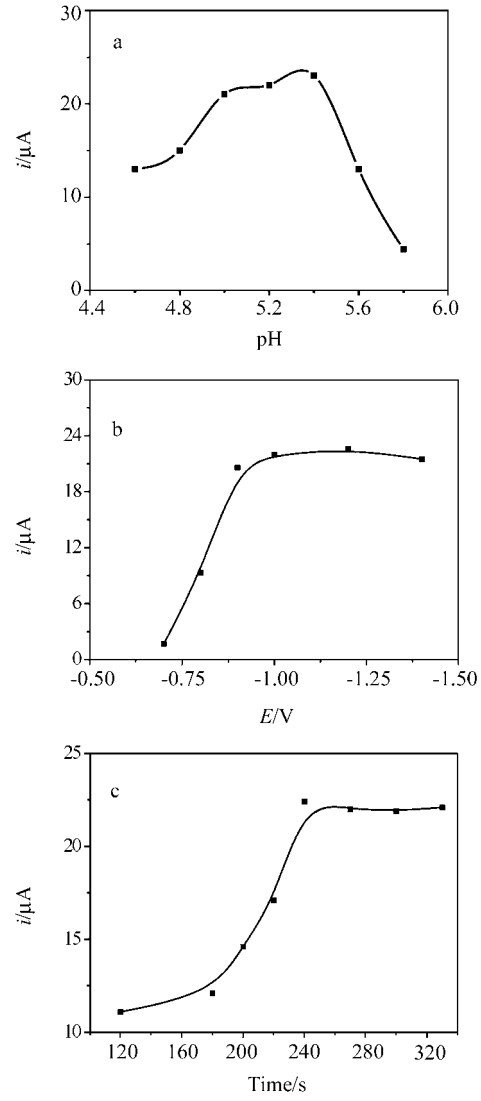

Figure 5 Effects of pH, accumulation potential and accumulation time on the anodic stripping peak current of the HEPMGCE. Pb^{2+} concentration, 5.0×10^{-7} mol / L; quiet time, 30 s; scan rate, 40 mV/s. (a) Accumulation potential, −1.0 V; accumulation time, 4 min. (b) pH=5.2; accumulation time, 4 min. (c) Accumulation potential, −1.0 V; pH=5.2.

Effect of the heparin concentration

The anodic stripping peak current of Pb^{2+} at the HEPMGCE was greatly higher than that at the GCE. The influence of the heparin concentration on the peak current was also studied and the results indicated that the peak current reached the maximum when the concentration heparin was 5.0×10^{-2} mg/mL. The dependence of the peak current on the volume of heparin (5.0×10^{-2} mg/mL) added to the GCE surface was examined over the range of 2 to 8 μL. It was clear that the presence of 4 μL of 5.0×10^{-2} mg / mL heparin was sufficient for obtaining the maximum peak current.

Effects of the pulse amplitude and scan rate

The influence of the pulse amplitude was investigated and it is suggested that the differential pulse anodic stripping peak current reached the maximum when the pulse amplitude was 0.075 V. As for the scan rate, the current response rose with the increasing scan rate. But when the scan rate exceeded 40 mV/s, the noise increased. Hence, a scan rate of 40 mV/s was chosen, since it could afford the highest signal-to-noise ratio.

Calibration plot, detection limit and precision

The dependence of the current response (i) on the concentration of Pb^{2+} (c) was linear in the range of 2.0×10^{-9} to 7.0×10^{-7} mol / L with a correlation equation:

$$i(\mu A)=4.09\times10^{7}c(\text{mol/L})+0.260 \quad \text{(correlation coefficient, 0.9990)}$$

The detection limit defined as the concentration at signal-noise ratio of 3 was 3.0×10^{-10} mol / L, and the relative standard deviation for 10 determinations of 1.0×10^{-8} mol / L Pb^{2+} was 4.83%.

Effect of other ions

The interference of various foreign metal ions on the determination of 1.0×10^{-7} mol/L Pb^{2+} was investigated. These species were added to the sample and the tolerable limit of a foreign substance was taken as a relative error less than 5%. The tolerated ratio of the foreign substances to 1.0×10^{-7} mol/L Pb^{2+} was 1000 for Na^{+}, K^{+}, Ca^{2+}, Al^{3+}, Ga^{3+}, Cr^{3+}, Mn^{2+}, Ba^{2+}, Co^{2+}, Ni^{2+}, Mg^{2+}, Zn^{2+}; 180 for Fe^{3+}; 5 for Cd^{2+}. Equal concentration of Cu^{2+} interfered with the determination of Pb^{2+}. Taking into account that the relative high concentration of Cu^{2+} was in most potable waters compared to Pb^{2+}, the interference of Cu^{2+} must be eliminated when the proposed method was applied to the determination of Pb^{2+} in natural and drinking water samples.

Cu^{2+} is known to react easily with iodide to form a stable compound, CuI, and the reaction is shown in Eq. (4):

$$2Cu^{2+}+4I^{-}=2CuI+I_2 \qquad (4)$$

When iodide was added into the solution-containing amount of Cu^{2+}, the iodide can react with Cu^{2+} to form CuI and then prevent Cu^{2+} from forming a complex

with heparin, and finally eliminate the interference of Cu^{2+}. For the determination of 1.0×10^{-7} mol/L Pb^{2+}, 500-fold amounts of Cu^{2+} can be eliminated in the presence of 5.0×10^{-3} mol/L iodide.

Analytical application

The utility of the developed method for the Pb^{2+} determination was tested by the determination of Pb^{2+} in different water samples. The river water samples were filtered with a 0.4 μm polycarbonate membrane filter, and then acidified to pH=2 with nitric acid. In order to destroy the organic substances, it is necessary to pretreat the water samples. The wet digestion and UV irradiation are the two usual methods. In this work the wet digestion method was used and the pretreatment procedure was the same as that described in literature.[23] Aliquots of water which contained 5.0×10^{-3} mol/L iodide were buffered with HOAc-NaOAc buffer to pH=5.2. Three replicate determinations of Pb^{2+} in unspiked and spiked water samples were carried out using the standard addition method. The results obtained by the proposed method are summarized in Table 1. They made evident that the present modified electrode was much effective for the determination of trace levels of Pb^{2+}.

Conclusion

A new chemically modified glass carbon electrode has been developed using heparin for the determination of Pb^{2+} at trace levels by differential pulse anodic stripping voltammetry. The proposed method can be used to determine trace amounts of Pb^{2+} in real samples. The detection limit of this method is 3.0×10^{-10} mol/L Pb^{2+}. In conclusion, the proposed method can be a potential candidate for the practical use of Pb^{2+} determination with high sensitivity, selectivity, simplicity and rapidness.

Table 1 Results of the determination of Pb^{2+} in water samples

Sample	Pb^{2+} found[a]/ (10^{-8} mol•L^{-1})	Pb^{2+} added/ (10^{-8} mol•L^{-1})	Pb^{2+} found after addition[a]/ (10^{-8} mol•L^{-1})	Recovery/%	SD
Drinking water	1.17	3.00	4.22	101.7	2.2
		10.00	10.81	96.4	4.1
		20.00	20.37	96.0	4.2
Tap water	8.82	4.00	12.58	94.0	2.0
		16.00	25.53	104.4	3.3
		24.00	33.15	101.4	2.2
River water	54.68	8.00	62.53	98.1	2.2
		20.00	75.22	102.7	2.8
		30.00	85.91	104.1	3.6

[a] Mean of three determinations.

References

1 Li, Z.; Tang, J.; Yu, X.; Liu, Y. *Anal. Lab.* **1999**, *18*, 57.
2 Sweileh, J. A. *Anal. Chim. Acta* **2001**, *448*, 151.
3 Huang, S. J.; Jiang, S. J. *Analyst* **2000**, *125*, 1491.
4 Bond, A. M. *Modern Polarographic Methods in Analytical Chemistry*, Marcel Dekker, Inc., New York, **1980**, p. 439.
5 Dong, Z. Z.; Han, J. S. *Potentiomtic Stripping Analysis*, Nankai University Press, Tianjin, **1992**, p. 3.
6 Felfman, B. J.; Osterloh, J. D.; Hata, B. H.; D'Alessandro, A. *Anal. Chem.* **1994**, *66*, 1983.
7 Zen, J. M.; Huang, S. Y. *Anal. Chim. Acta* **1994**, *296*, 77.
8 Schlereth, D. D. *J. Electroanal. Chem.* **1997**, *425*, 77.
9 Johnson, D. C.; Ryan, M. D.; Wilson, G. S. *Anal. Chem.* **1986**, *58*, 33R.
10 Dong, S. J.; Chen, G. L.; Xie, Y. W. *Chemically Modified Electrodes*, Chinese Science Press, Beijing, **1995**, p. 362 (in Chinese).
11 Xu, J.; Liu, B. *Analyst* **1994**, *119*, 1599.
12 Ramos, J. A.; Bermejo, E.; Zapardiel, A.; Perez, J. A.; Hernandez, L. *Anal. Chim. Acta* **1993**, *273*, 219.
13 Fei, J. J.; Li, J. N.; Yi, F. Y. *Chin. J. Anal. Chem.* **2001**, *29*, 916 (in Chinese).
14 Degfa, T. H.; Chandravanshi, B. S.; Alemu, H. *Electroanalysis* **1999**, *11*, 1305.
15 Honeychurch, K. C.; Hart, J. P.; Cowell, D. C. *Anal. Chim. Acta* **2001**, *431*, 89.
16 Prabhu, S. V.; Baldwin, R. P.; Kryger, L. *Electroanalysis*, **1989** *1*, 13.
17 Connor, M.; Dempsey, E.; Smyth, M. R.; Richardson, D. H. S. *Electroanalysis* **1991**, *3*, 331.
18 Zayats, G. D.; Meryan, V. T.; Revenco, M. D.; Chiugureanu, D. G. *Anal. Lett.* **2002**, *35*, 577.
19 Hu, Z.; Seliskar, C. J.; Heineman, W. R. *Anal. Chim. Acta* **1998**, *369*, 93.
20 Zhang, S.; Huang, W. *Anal. Sci.* **2001**, *17*, 983.
21 Wolfrom, M. L.; Tomomatsu, H.; Szarek, W. A. *J. Org. Chem.* **1966**, *31*, 1173.
22 Li, H. L.; Liu, S. Q.; Jiang, M.; Li, P. B. *Anal. Lab.* **1997**, *16*, 13.
23 Dong, Z. Z.; Han, J. S. *Potentiometric Stripping Analysis*, Nankai University Press, Tianjin, **1992**, p. 193 (in Chinese).

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