# Catalytic Oxidation of Uric Acid at the Polyglycine Chemically Modified Electrode and its Trace Determination

**Ai-min Yu, Hai-li Zhang and Hong-yuan Chen***

*Department of Chemistry, Nanjing University, Nanjing 210093, China*

**Cyclic voltammetry was undertaken to investigate the electrochemical behavior of uric acid at a polyglycine modified electrode. The modified electrode shows catalytic ability for the oxidation of uric acid, reducing the overpotential by 250 mV in pH 7.0 phosphate buffer solution. The enhanced voltammetric response can be used to determine uric acid. The linear range is between $5.0 \times 10^{-8}$ and $4.5 \times 10^{-6}$ M with a detection limit as low as $5.0 \times 10^{-9}$ M. The relative standard deviation is 1.4% (8 runs) at a concentration of 50 μM uric acid. The catalytic effect of the modified electrode resulted in the voltammetric resolution of the overlapping of uric acid and ascorbic acid. This allows the simultaneous detection of uric acid and ascorbic acid in the same sample.**

***Keywords:*** *Electrocatalytic oxidation; uric acid; polyglycine chemically modified electrode; amperometric detection*

Uric acid (2,6,8-trihydroxypurine, UA) is a primary end-product of purine metabolism. Extreme, abnormal levels of uric acid in body fluids will lead to some diseases.[1] It is for this reason, that simple and rapid detection methods are required. There are some electrochemical methods for the determination of uric acid based on the oxidation of uric acid at glassy carbon electrodes,[2] and carbon paste electrodes.[3] These methods, however, inevitably suffer from interference from ascorbic acid (AA) which can be oxidized at a potential close to that of uric acid. Some new techniques, such as the adsorption/medium exchange approach,[4] screen-printed electrodes,[5] electrochemically pretreated carbon paste electrodes[6] and various enzyme techniques[7,8] were later developed with respect to the improvement of selectivity, but these methods were either too expensive or the detection limits needed to be improved. Recently, polymer films with high stability and selectivity have been widely used as coatings to detect biological substances.[9-11]

In this paper, we describe the improved voltammetric behavior of uric acid and its trace determination at a polyglycine chemically modified electrode. The resolution of uric acid and ascorbic acid in a mixture is also reported.

## Experimental

### *Reagents and Chemicals*

DL-glycine (>99.0%) was purchased from The Shanghai Biochemical Institute, China. Uric acid (UA) was purchased from Sigma (St. Louis, MO, USA). Ascorbic acid (AA; >99.7%) was from Nanjing Chemicals, China. All other reagents were of analytical-reagent grade. Phosphate buffer solutions (PBS; 0.1 M, at various pH values) were prepared by mixing four stock solutions of 0.1 M $H_3PO_4$, $NaH_2PO_4$, $Na_2HPO_4$ and $Na_3PO_4$. All solutions were prepared with doubly-distilled water.

### *Apparatus*

The electrochemical experiments were carried out with a Model E506 Polarecord and an E612 VA-scanner (Metrohm, Herisau, Switzerland). In some cases a Model 270 electrochemical analyzer and a Model 636 Ring-Disk Electrode (RDE) System (EG&G, Princeton Applied Research, Princeton, NJ, USA) were used. A conventional three-electrode system was used throughout. The working electrode was a bare or a polyglycine coated glassy carbon electrode (GCE, d = 0.3 cm; RDE, d = 0 .6 cm), the auxiliary electrode was a Pt wire and an SCE was used as a reference electrode.

### *Preparation of Polyglycine Coated Glassy Carbon Electrode*

Before the modification procedure, the GCE was polished on wet fine emery paper and 0.05 μm alumina slurries, thoroughly rinsed with water and sonicated in distilled water. It was then placed in 0.01 M glycine solution (pH 7.0 PBS) which was previously deaerated with high purity nitrogen for 10 min. The electrode was treated with cyclic scanning between −0.5 and 1.8 V at a scan rate of 100 mV $s^{-1}$, four times. The electrode was ready for use after the final washing with water.

## Results and Discussion

### *Electrochemical Behavior of UA at the Polyglycine Modified Electrode*

Fig. 1 shows the cyclic voltammograms (CVs) of 0.5 mM UA at a bare GCE [curve (a)] and the polyglycine modified electrode [curve (b)] in pH 5.0 (A), 7.0 (B) and 9.9 (C) phosphate buffer solution. In all solutions, at the bare GCE, UA gives a broad and irreversible oxidation peak and the oxidation peak does not change with the pH of the solution. At the modified electrode, the oxidation peak became well-defined. The potential shifted to the negative direction and the peak current greatly increased compared with the CVs at the unmodified electrode, probably because the modified electrode accelerated the rate of electron transfer of UA.

From Fig. 1, it is also evident that the catalytic effect varied with the pH of the electrolyte solution. The catalytic effect can be evaluated from two values, one is the increment in catalytic current ($\Delta I$), the other is the value of decrease in overpotential ($\Delta E$). Variation of the catalytic effect of the modified electrode with pH is shown in Table 1. It can be seen that, with increasing pH, the enhancement of the peak current decreases but the value of $\Delta E$ increases greatly. It indicates that basic conditions are beneficial for the reaction. A pH of 7.0 was finally chosen for the determination of UA because it had a relatively better catalytic effect when both high catalytic current and low detection potential were considered.

### *Scan Rate Dependence*

The scan rate dependence of the modified electrode in 0.5 mM UA was also studied (see Fig. 2). As the scan rate increased, the

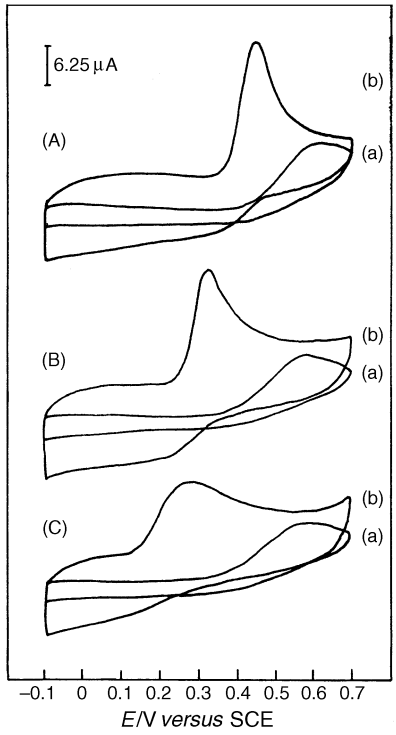


**Fig. 1** Cyclic voltammograms of 0.5 mM UA in (A) pH 5.0 (B) pH 7.0 (C) pH 9.9 PBS at (a) bare GCE and (b) polyglycine modified electrode. Scan rate: 100 mV $s^{-1}$. The bare GCE was polished in the same manner as the modified electrode, but not submitted to the electrochemical treatment.

**Table 1** The pH dependence of the catalytic effect of the polyglycine modified electrode

| | Bare electrode | | Modified electrode | | | |
|---|---|---|---|---|---|---|
| pH | ($E_p$/V) | ($I_{pa}$/μA) | ($E_p$/V) | ($I_{pa}$/μA) | ($\Delta E_p$/V) | ($\Delta I$/μA) |
| 5.0 | 0.59 | 10.6 | 0.44 | 22.5 | 0.15 | 11.9 |
| 7.0 | 0.58 | 10.0 | 0.32 | 18.75 | 0.26 | 8.75 |
| 9.9 | 0.58 | 9.4 | 0.27 | 11.26 | 0.31 | 1.86 |

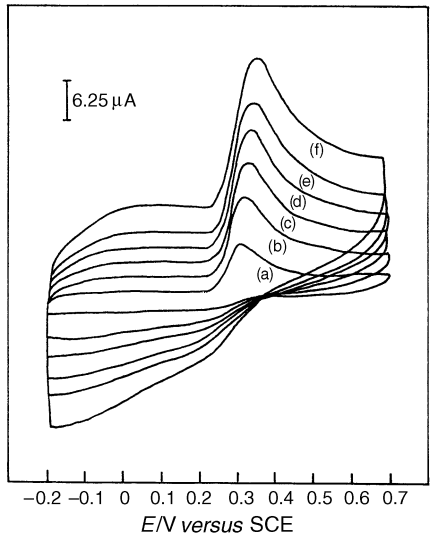


**Fig. 2** Cyclic voltammograms of 0.5 mM UA at the polyglycine modified electrode in PBS of pH 7.0. Scan rate: (a) 20; (b) 60; (c) 100; (d) 150; (e) 200; and (f) 300 mV $s^{-1}$.

anodic peak current increased and the anodic peak potential shifted slightly in the positive direction. For example, at 20 mV $s^{-1}$, the oxidation potential was 0.31 V while at 300 mV $s^{-1}$, the oxidation potential shifted to 0.36 V. The anodic peak current increased linearly with the square root of scan rates in the range from 40 to 200 mV $s^{-1}$. It demonstrates that this electrode reaction is concerned with the diffusion process. Similar behavior of UA has been reported in acidic medium (pH 4.0) with a screen-printed electrode[5] and in basic solution (0.01 M NaOH + 0.10 M $NaClO_4$) with a pretreated carbon paste electrode.[6]

### *Hydrodynamic Amperometry*

Fig. 3 shows the hydrodynamic voltammograms obtained for 0.10 mM UA at the polyglycine modified electrode in PBS of pH 7.0. The current response starts at 0.25 V and increases quickly with increasing potential. It finally reaches a steady response at about 0.60 V. At the unmodified electrode, current increased gradually with increasing potential and there was no plateau in the potential range from 0.1 to 0.8 V. The response to 50 μM UA at the modified electrode was about ten times larger than that at the bare electrode. Thus, the polyglycine chemically modified electrode could be used as an effective amperometric sensor for UA with high sensitivity and low detection potential. In this work, an applied voltage of 0.6 V was chosen for the amperometric detection of UA.

A typical hydrodynamic amperometry obtained by successively adding 50 μM UA to the continuously stirred buffer solution (pH 7.0) is shown in Fig. 4(A). The response time is

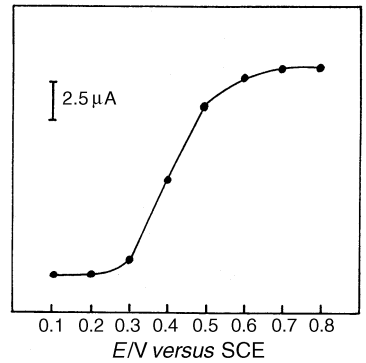


**Fig. 3** Hydrodynamic voltammograms of 0.10 mM UA at the polyglycine modified electrode in PBS of pH 7.0.

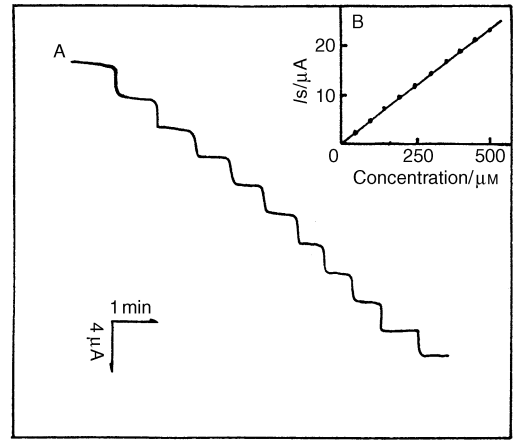


**Fig. 4** (A) Current-time recording at the modified electrode while increasing the UA concentration in $1 \times 10^{-5}$ M steps at 0.60 V in PBS of pH 7.0. (B) The plots of the resulting calibration graph. Rotating speed, 600 rpm.

about 5 s and the RSD for 8 runs at 50 μM is 1.4%. Fig. 4(B) shows the calibration graphs of successive additions of aliquots of a concentrated solution of UA which is shown in Fig. 4(A). The linear range is between $5.0 \times 10^{-8}$ and $4.5 \times 10^{-6}$ M UA. The detection limit is down to $5.0 \times 10^{-9}$ M which was estimated as three times the noise of the determination of a low level of UA. Furthermore, the modified electrode possesses a good stability, even after being in use for a month; 90% of its initial response was still obtained.

### *Separation From Ascorbic Acid*

Ascorbic acid co-presents with UA in many biological samples such as blood and urine. Therefore, the electrochemical behavior of AA was also investigated. At the unmodified electrode, AA was oxidized at the same potential (0.60 V) as UA and the cyclic voltammograms of UA and AA are overlapping [Fig. 5, curve (a)]. However, at the polyglycine modified electrode, the oxidation potential of AA shifted to about 0.30 V and that of UA shifted to 0.45 V [Fig. 5, curve (b)]. Thus, a potential separation of 150 mV between AA and UA was obtained. This separation is large enough for the simultaneous determination of UA and AA in the same sample. It can also be seen that for the same concentration of UA and AA, the modified electrode responds to UA much better than to AA. Since the maximum tolerant concentration of AA for the determination of UA is as large as 40-fold, this method can be applied to various biological samples. With its low cost, high stability and ease of preparation, the polyglycine film appears to open up a new opportunity for the further development of sensors.

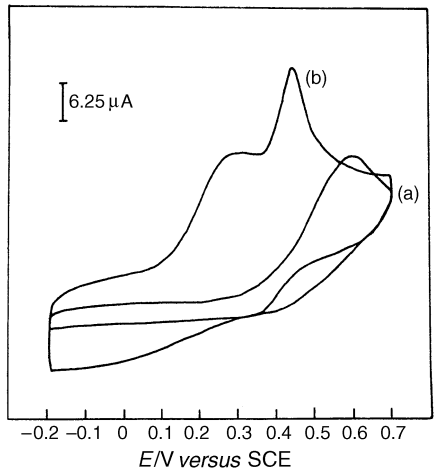


**Fig. 5** Cyclic voltammograms of 0.5 mM UA and 0.5 mM AA at (a) bare GCE and (b) polyglycine modified electrode in PBS of pH 7.0. Scan rate, 100 mV $s^{-1}$.

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