Electrochimica Acta 54 (2009) 6837–6843

Contents lists available at ScienceDirect

Electrochimica Acta

journal homepage: www.elsevier.com/locate/electacta

# Simultaneous determination of paracetamol and ascorbic acid using tetraoctylammonium bromide capped gold nanoparticles immobilized on 1,6-hexanedithiol modified Au electrode

Santhosh S. Nair, S. Abraham John*, Takamasa Sagara**

*Department of Chemistry, Gandhigram Rural University, Gandhigram 624302, Dindigul, India*

ARTICLE INFO

*Article history:*
Received 12 May 2009
Received in revised form 22 June 2009
Accepted 30 June 2009
Available online 4 July 2009

*Keywords:*
Gold nanoparticles modified electrode
ATR-FT-IR spectroscopy
Amperometry
Paracetamol
Ascorbic acid

ABSTRACT

Tetraoctylammonium bromide stabilized gold nanoparticles (TOAB-AuNPs) attached to 1,6-hexanedithiol (HDT) modified Au electrode was used for the simultaneous determination of paracetamol (PA) and ascorbic acid (AA) at physiological pH. The attachment of TOAB-AuNPs on HDT modified Au surface was confirmed by attenuated total reflectance (ATR)-FT-IR spectroscopy and atomic force microscope (AFM). The ATR-FT-IR spectrum of TOAB-AuNPs attached to the HDT monolayer showed a characteristic stretching modes corresponding to $-CH_2$ and $-CH_3$ of TOAB, confirming the immobilization of AuNPs with surface-protecting TOAB ions on the surface of the AuNPs after being attached to HDT modified Au electrode. AFM image showed that the immobilized AuNPs were spherical in shape and densely packed to a film of ca. 7 nm thickness. Interestingly, TOAB-AuNPs modified electrode shifted the oxidation potential of PA towards less positive potential by 70 mV and enhanced its oxidation current twice when compared to bare Au electrode. In addition, the AuNPs modified electrode separated the oxidation potentials of AA and PA by 210 mV, whereas bare Au electrode failed to resolve them. The amperometry current of PA was increased linearly from $1.50 \times 10^{-7}$ to $1.34 \times 10^{-5}$ M with a correlation coefficient of 0.9981 and the lowest detection limit was found to be 2.6 nM ($S/N = 3$). The present method was successfully used to determine the concentration of PA in human blood plasma and commercial drugs.



## 1. Introduction

Paracetamol (*N*-acetyl-p-aminophenol, 4-acetamidophenol, acetaminophen or tylenol) is an analgesic and antipyretic drug used worldwide mainly for the reduction of fever and also as a painkiller for the relief of mild to moderate pain associated with headache, backache, arthritis and postoperative pain [1–5]. It has no anti-inflammatory action which made it an obvious choice for home medication for more than three decades [4]. At normal therapeutic doses, paracetamol (PA) is rapidly and completely metabolized by undergoing glucuronidation and sulfation to inactive metabolites which are eliminated in the urine [4]. However, an overdose of PA can lead to the accumulation of toxic metabolites, which may cause acute hepatotoxicity and nephrotoxicity [6–9]. It can also cause liver disorders, skin rashes and inflammation of the pancreas [4]. Thus, it is essential to establish a simple inexpensive method to determine the concentration of PA with high selectivity and sensitivity. Many methods have been described in the literature for the determination of PA including titrimetry [10], spectrophotometry [11], liquid chromatography [12], FT-IR Raman spectrometry [13,14], flow injection analysis with different techniques of detection [10,15] and voltammetry [16]. However, both spectrophotometric and titrimetric methods require special reagents and a tedious extraction procedure prior to the determination whereas liquid chromatography method is time consuming. In contrast, voltammetric method of determination involves less time consumption, no tedious procedures and further it is highly selective and sensitive [16]. Therefore, in recent years much interest has been directed to the determination of PA by voltammetric methods using various modified electrodes [5,17–24].

It is well known that ascorbic acid (AA) is one of the major interferents which will always encounter in the determination of PA in biological fluids because its oxidation potential is rather close to the oxidation potential of PA at bare electrodes in physiological pH. Therefore, determination of PA in the presence of AA is very important from the clinical point of view. Carbon based electrodes have been extensively used for the determination of PA [1,5,17–22]. In addition, two reports have been published recently

* Corresponding author. Fax: +91 451 245 3031.
** Corresponding author at: Department of Applied Chemistry, Faculty of Engineering, Nagasaki University, Bunkyo 1-14, Nagasaki 852-8521, Japan.
Tel.: +81 95 819 2676; fax: +81 95 819 2676.
*E-mail addresses:* abrajohn@yahoo.co.in (S.A. John), sagara@nagasaki-u.ac.jp (T. Sagara).

0013-4686/$ – see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.electacta.2009.06.077

for the determination of PA using gold nanoparticles (AuNPs) modified electrodes [23,24]. However, to date, only two reports have been published for the determination of PA in the presence of AA using boron doped diamond electrode [22] and carbon coated nickel magnetic particles modified electrode [1]. But in both the papers, the determination of PA in the presence of AA was carried out at pH 1.96 [22] and pH 3.0 [1]. For clinical applications, it is highly desirable that the electrochemical determination should be effective and sensitive at physiological pH.

The goal of the present paper is to determine PA in the presence of AA using tetraoctylammonium bromide (TOAB) capped AuNPs (TOAB-AuNPs) modified electrode in 0.20 M phosphate buffer solution (pH 7.20). Recently, Au electrodes modified with AuNPs have been extensively used for the fabrication of various electrochemical biosensors due to their excellent biocompatibility and electrochemical properties [25–27]. It has been shown that the AuNPs modified electrodes exhibit several advantageous features over bare Au electrode [25]. While scanning the literature, citrate-capped AuNPs (C-AuNPs) have been used most extensively for the modification of electrodes for the construction of electrochemical biosensors [25]. It has been shown that densely packed AuNPs could not be achieved on electrodes modified with different linker molecules using C-AuNPs mainly due to interparticle repulsion between the nanoparticles [28,29]. On the other hand, densely packed AuNPs can be prepared by using AuNPs capped with weakly bound ligands such as TOAB [30]. Therefore, we have used TOAB capped AuNPs for the modification of the electrode surface in the present study in light of our recent work [31]. The self-assembled monolayer (SAM) of 1,6-hexanedithiol (HDT) is used as a linker to attach the TOAB-AuNPs. AFM image shows that the AuNPs are densely packed on the surface of the HDT modified Au electrode. Interestingly, TOAB-AuNPs modified electrode resolves the oxidation peaks of AA and PA with a potential difference of 210 mV in 0.20 M phosphate buffer solution (pH 7.20) while bare Au electrode fails to resolve them. We have achieved the lowest detection limit of 2.6 nM PA ($S/N = 3$) at TOAB-AuNPs modified electrode.

## 2. Experimental

### 2.1. Chemicals

1,6-Hexanedithiol (Aldrich) and $HAuCl_4 \cdot 3H_2O$ (Sigma) were used as received. Tetraoctylammonium bromide (TOAB), ascorbic acid (AA) and paracetamol (PA) were purchased from Merck (India). Phosphate buffer solution (pH 7.20) was prepared by using $Na_2HPO_4$ and $NaH_2PO_4$. All other chemicals used in this investigation were of analytical grade and were used without further purification. The Au working electrode was polished with alumina powder (0.50 μm) and sonicated in double distilled water for 10 min. The polished Au electrode was then electrochemically cleaned by cycling the potential between −0.20 and 1.50 V in 0.05 M $H_2SO_4$ at a scan rate of 1.0 V $s^{-1}$ for 5 min or CV characteristics for a cleaned electrode is obtained.

### 2.2. Preparation of tetraoctylammonium bromide capped AuNPs (TOAB-AuNPs)

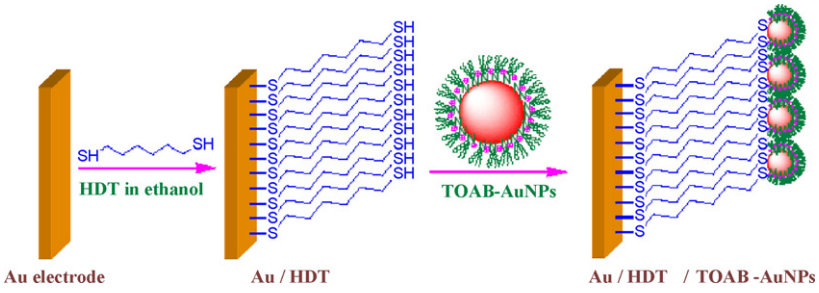
The TOAB-AuNPs were prepared in toluene by the reported two-phase method [32]. An aqueous solution of $HAuCl_4 \cdot 3H_2O$ (500 mg in 40 mL) was mixed with a solution of TOAB in toluene (3.06 g in 100 mL). The two-phase mixture was vigorously stirred until tetrachloroaurate was completely transferred into the organic phase to give a deep orange solution. Then a fresh aqueous solution of sodium borohydride (525 mg in 30 mL) was added slowly while stirring. The deep orange color of the organic phase immediately turned to ruby red. After further stirring (12 h), the organic layer was extracted and washed with Millipore water (3 times) and then dried over anhydrous sodium sulfate. The solution was diluted to 250 mL with toluene before use. The size of colloidal AuNPs was investigated by transmission electron microscopy (TEM) and was found to be ~5–7 nm (Supporting Information, Fig. S1).

### 2.3. Modification of electrode surface

The immobilization of TOAB-AuNPs on polycrystalline Au electrode modified with HDT monolayer was schematically shown in Scheme 1. The monolayer of 1,6-hexanedithiol (HDT) was prepared on Au electrode (Au/HDT) by immersing a clean polycrystalline Au electrode into 5 mM ethanolic solution for 3 h under nitrogen atmosphere. The HDT modified electrode was rinsed with ethanol, then immersed in toluene for 20 min, and soaked in TOAB-AuNPs in toluene for 20 min. Then the electrode was washed with toluene, followed by ethanol and then water, and used for spectral and electrochemical measurements in the phosphate buffer (PB) solution.

### 2.4. Instrumentation

Electrochemical measurements were performed in a conventional two compartment three electrode cell with a polished polycrystalline Au electrode (area = 0.02 $cm^2$) as working electrode, a Pt wire as counter electrode and NaCl saturated Ag/AgCl as reference electrode. All the electrochemical measurements were carried out with CHI Model 643 (Austin, TX, USA) Electrochemical Workstation. UV–visible spectra were recorded with a U-3000, Hitachi spectrophotometer (Japan). Attenuated total reflectance (ATR) FT-IR measurements were carried out using JASCO FT-IR 460 plus model equipped with a horizontal ZnSe crystal (Pike Technologies, USA). The resolution of the spectra was 4 $cm^{-1}$ and the scan was repeated 100 times. The signals due to water vapor and carbon dioxide were removed by the software provided by JASCO FT-IR 460 plus model. The thin gold film produced by sputtering 99.99% pure gold on a glass plate under vacuum was used as the substrate for




**Scheme 1.** Schematic representation of HDT-SAM formation on Au electrode and subsequent immobilization of TOAB-AuNPs on HDT modified Au electrode.

ATR-FT-IR measurements. Atomic force microscope (AFM) imaging was carried out by using SPI3800N (Seiko Instrumental Co.) with a $Si_3N_4$ probe (cantilever SN-AF01, Seiko Instrumental Co.).

## 3. Results and discussion

### *3.1. Characterization of Au/HDT and Au/HDT/TOAB-AuNPs by ATR-FT-IR spectroscopy and AFM*

Recently, based on the XPS results, we have shown that HDT formed a monolayer on Au electrode through one of the two –SH groups and the other –SH group is free from binding with the Au surface and these free –SH groups were used for the successful attachment of citrate-AuNPs [27]. We have followed the same experimental conditions used in our previous work [27] for the preparation of HDT monolayer on Au electrode in the present study. In the present study, we have attached TOAB-AuNPs to the free –SH groups of the HDT monolayer and then characterized by ATR-FT-IR spectroscopy. The interaction of TOAB with AuNPs is relatively weaker than the interaction of citrate ions with AuNPs and therefore, deposition of AuNPs on HDT monolayer is facilitated by the release of TOAB molecules from the surface of the AuNPs. Fig. 1A shows the ATR-FT-IR spectra obtained for HDT-SAM on Au coated glass plate and TOAB-AuNPs attached to a HDT-SAM on Au coated glass plate. In contrast to HDT-SAM (curve a), several new bands were observed for TOAB-AuNPs attached to HDT-SAM (curve b) indicating that AuNPs were successfully immobilized on the monolayer of HDT. For comparison, we have also recorded the FT-IR spectrum for solid TOAB in KBr pellet, and it is shown in Fig. 1B. The different bands observed for solid TOAB and TOAB-AuNPs attached to HDT-SAM are given in Table S1 (Supporting Information). It can be seen from curve b in Fig. 1A that TOAB attached with HDT-SAM shows several bands similar to solid TOAB with slight shift (Table S1). The characteristic $CH_2$ vibration bands at 714 (in phase rocking), 1458 ($CH_2$ scissor deformation), 2859 ($CH_2$ symmetric stretch) and $CH_3$ asymmetric stretch at 2925 $cm^{-1}$ were obtained for TOAB-AuNPs attached to HDT-SAM (Fig. 1A, curve b). Similar stretching bands were also observed for solid TOAB with small shift (Fig. 1B), indicating that TOAB molecules remain on the surface of the AuNPs after being attached to HDT monolayer. The $CH_3$ stretching band obtained at 2925 $cm^{-1}$ for TOAB-AuNPs attached to HDT-SAM indicates that all trans-alkyl chains may surround the particle surface and they are highly ordered in the solid phase.

Atomic force microscopy (AFM) was used to examine the size and morphology of the TOAB-AuNPs immobilized on a HDT monolayer. Fig. 2 shows the AFM image obtained for bare, HDT modified and TOAB-AuNPs immobilized on HDT modified Au substrates. When compared to the AFM images of bare Au and HDT coated Au substrates (images A and B), TOAB-AuNPs immobilized on HDT modified Au substrate showed that the AuNPs were densely packed and were mostly spherical in shape. The height of the AuNPs film is ca. 7 nm implying that the size of the AuNPs remains the same after attached with the HDT monolayer. The lateral size of the AuNPs was larger than the height of the AuNPs due to aggregation.

### *3.2. Electrochemical oxidation of paracetamol (PA) and ascorbic acid (AA)*

The main objective of the present paper is to determine PA in the presence of AA at physiological pH using TOAB-AuNPs modified electrode. Before studying the voltammetric response of PA in the presence of AA, we have examined the oxidation of PA. Fig. 3A illustrates the linear sweep voltammograms (LSVs) obtained for 0.50 mM PA in 0.20 M phosphate buffer (PB) solution (pH 7.20) at a scan rate of 50 mV $s^{-1}$. At bare Au electrode, the oxidation of PA occurs at 0.54 V (curve a) whereas Au/HDT electrode shows an ill-defined oxidation wave for PA (curve b). In contrast to bare Au and Au/HDT electrodes, a sharp oxidation peak for PA was observed at 0.47 V with enhanced oxidation peak current at TOAB-AuNPs modified electrode (curve c). The observed 70 mV less positive potential shift for PA at AuNPs modified electrode clearly shows that AuNPs decreased the thermodynamic overpotential of PA. The enhanced oxidation peak current of PA at a AuNPs modified electrode is attributed to the increased effective surface area of the immobilized AuNPs electrode when compared to a bare Au electrode [25,33]. We have determined the effective surface area of the bare Au and Au/HDT/TOAB-AuNPs electrodes by potential step chronoamperometric measurements using 1 mM $K_3[Fe(CN)_6]$ in 0.10 M KCl [34]. At both the electrodes, linear Cottrell plots were obtained, indicating diffusion controlled process, as later confirmed by sweep rate dependence of LSVs. The Cottrell slope for bare Au electrode was 0.59 μA $s^{1/2}$ and that of TOAB-AuNPs modified electrode was 0.94 μA $s^{1/2}$, indicating that the effective surface area of TOAB-AuNPs modified electrode was higher than that at bare Au electrode. Further, the stability of PA oxidation at TOAB-AuNPs modified electrode was studied by recording LSVs at continuous potential scans. Fig. 3B shows the LSVs recorded for the oxidation of PA before and after 10 continuous potential scans. It can be seen from curve b of Fig. 3B that the oxidation potential of PA remains same after 10 continuous potential scans. On the other hand, the oxidation potential of PA was shifted to more positive potential at a bare Au electrode and becomes a shoulder wave after 10 continuous potential scans

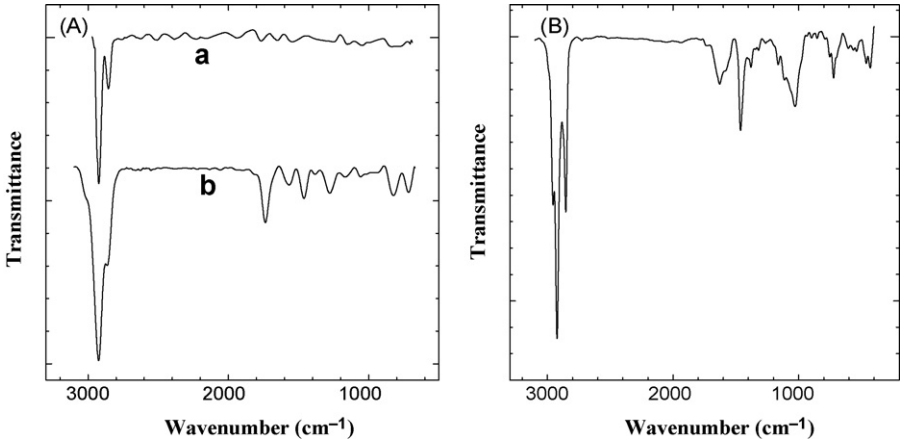



**Fig. 1.** (A) ATR-FT-IR spectra obtained for (a) HDT on Au coated glass plate and (b) HDT/TOAB-AuNPs on Au coated glass plate. (B) FT-IR spectrum of solid TOAB in KBr pellet.

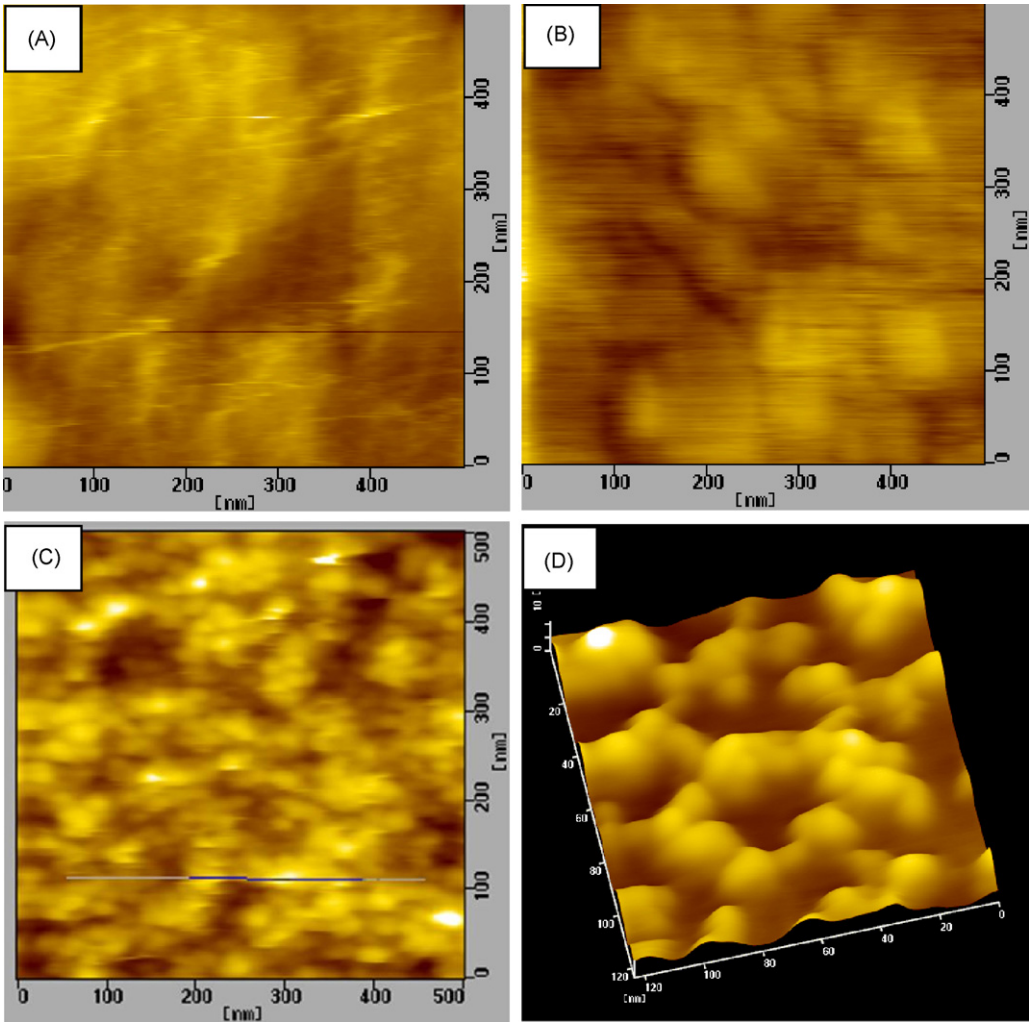



**Fig. 2.** AFM images of (A) bare (B) HDT modified and (C) TOAB-AuNPs immobilized on HDT modified Au plates. (D) Close view of (C).

(Supporting Information, Fig. S2) indicating that stable determination of PA at bare Au electrode is not possible. The oxidation current of PA at TOAB-AuNPs modified electrode increases linearly with the potential scan rate (Supporting Information, Fig. S3). A linear relationship between the PA oxidation current and the square root of scan rate from 0.1 to 1.0 V $s^{-1}$ was obtained with a correlation coefficient of 0.9998. This indicates that the oxidation process of PA TOAB-AuNPs modified electrode is diffusion controlled process. We have also studied the oxidation of AA at AuNPs modified electrode. At bare Au electrode, oxidation of AA occurs at 0.50 V, while at AuNPs modified electrode its oxidation occurs at 0.27 V with 2-fold higher oxidation current (Supporting Information, Fig.

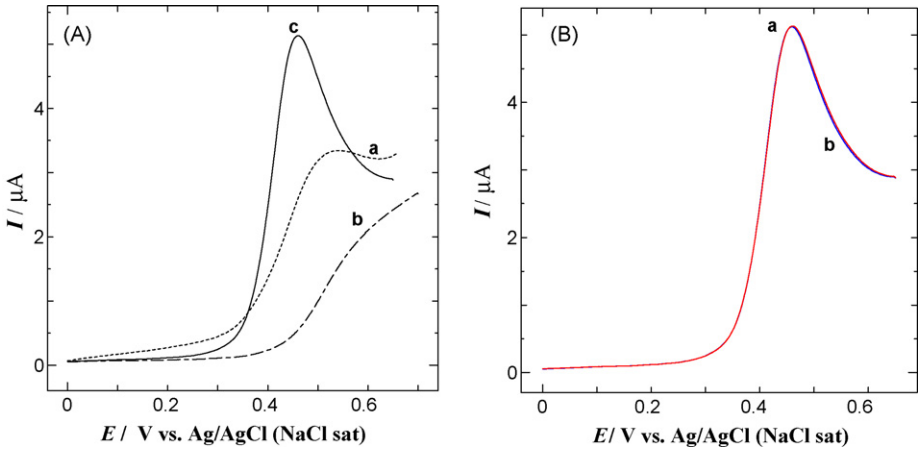



**Fig. 3.** (A) LSVs obtained for 0.50 mM PA at (a) bare Au, (b) Au/HDT and (c) Au/HDT/TOAB-AuNPs electrodes. (B) LSVs obtained for 0.5 mM PA at Au/HDT/TOAB-AuNPs electrode after (a) 1st and (b) 10th scans. Supporting electrolyte = 0.2 M PBS (pH 7.20) and scan rate = 50 mV $s^{-1}$.

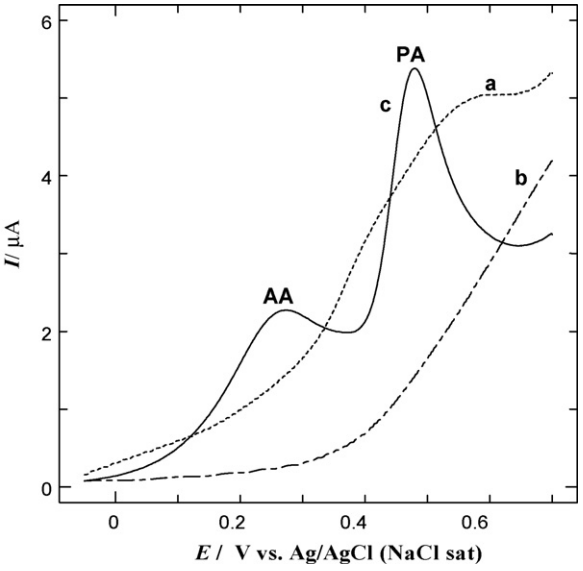



**Fig. 4.** LSVs obtained for mixture of 0.50 mM AA and PA at (a) bare Au, (b) Au/HDT and (c) Au/HDT/TOAB-AuNPs electrodes at a scan rate of 50 mV $s^{-1}$ in 0.2 M PBS (pH 7.20).

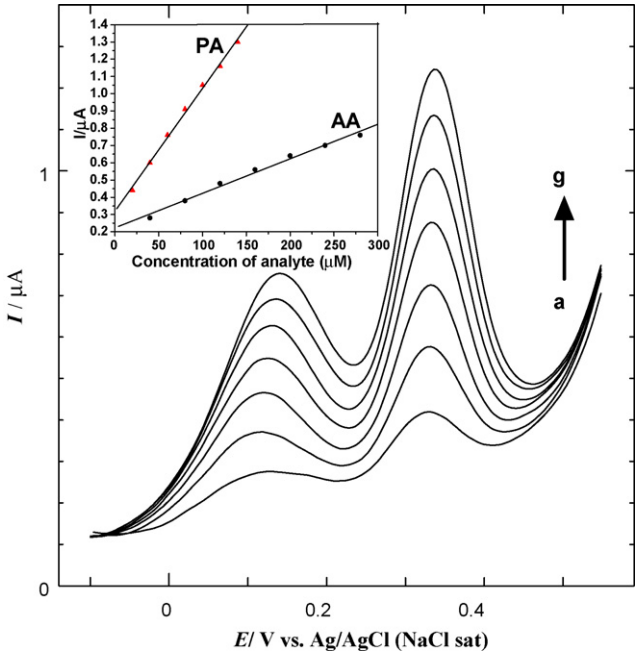



**Fig. 5.** DPVs for the oxidation of AA and PA at Au/HDT/TOAB-AuNPs modified electrode. Each addition increased the concentration of 40 μM AA and 20 μM PA (a–g) in 0.20 M PB solution (pH 7.20). Pulse width = 0.06 s, amplitude = 0.05 V, sample period = 0.02 s and pulse period = 0.2 s.

S4). In contrast to bare Au electrode, 230 mV less positive potential shift with higher oxidation current was observed at AuNPs modified electrode.

### 3.3. Simultaneous determination of AA and PA

Since AA is one of the main interferents in the voltammetric determination of PA, we have also studied the determination of PA in the presence of AA using TOAB-AuNPs modified electrode. The LSVs obtained for a mixture of 0.5 mM each PA and AA in 0.2 M PB solution (pH 7.2) at bare Au, Au/HDT and Au/HDT/TOAB-AuNPs electrodes at a scan rate of 50 mV $s^{-1}$ are shown in Fig. 4. Both bare Au and Au/HDT electrodes fail to separate the oxidation peaks of AA and PA (curves a and b). However, Au/HDT/TOAB-AuNPs electrode successfully resolves the oxidation peaks of AA and PA with a potential difference of 210 mV. The oxidation of AA occurs at 0.27 V while the oxidation of PA occurs at 0.48 V. The oxidation peak potentials of AA and PA were highly stable upon repeated potential cycling indicating that Au/HDT/TOAB-AuNPs electrode does not undergo surface fouling caused by the oxidized products of AA in PB solution. Recently, simultaneous determination of AA and PA was reported at boron doped diamond electrode [22]. Although this electrode separates the oxidation potentials of PA and AA at pH 1.96, but it fails to resolve the oxidation potentials of AA and PA at pH 7.20 [22]. However, in the present study, AuNPs modified electrode successfully separates the voltammetric signals of AA and PA with a peak separation of 210 mV at pH 7.2, which is more than enough to determine the concentrations of both AA and PA simultaneously and also individually in a mixture.

Further, the TOAB-AuNPs modified electrode was used for the simultaneous determination of AA and PA. Fig. 5 depicts the differential pulse voltammograms (DPVs) obtained at TOAB-AuNPs modified electrode by simultaneously changing the concentration of each 40 μM AA and 20 μM PA in 0.20 M PB solution (pH 7.20). The current responses due to the oxidation of both AA and PA were increased linearly with a correlation coefficient of 0.9998 and 0.9993, respectively (Fig. 5 inset). These results indicate that TOAB-AuNPs modified electrode can be successfully used for the simultaneous determination of both AA and PA.

Fig. 6 shows differential pulse voltammograms (DPVs) obtained for 400 μM of AA in the presence of 10, 20, 30, 40 and 50 μM of PA in 0.2 M PB solution. It can be seen from Fig. 6 that a well-defined voltammetric signal due to 20 μM PA oxidation appears at 0.41 V even in the presence of 20-fold higher concentration of AA (curve c). The oxidation current of PA increases linearly without affecting the signal of AA. These results indicate that the present modified electrode is more sensitive towards PA even in the presence of high concentration of AA. The peak current corresponding to PA oxidation linearly increased while increasing the PA concentration with a correlation coefficient of 0.9987.

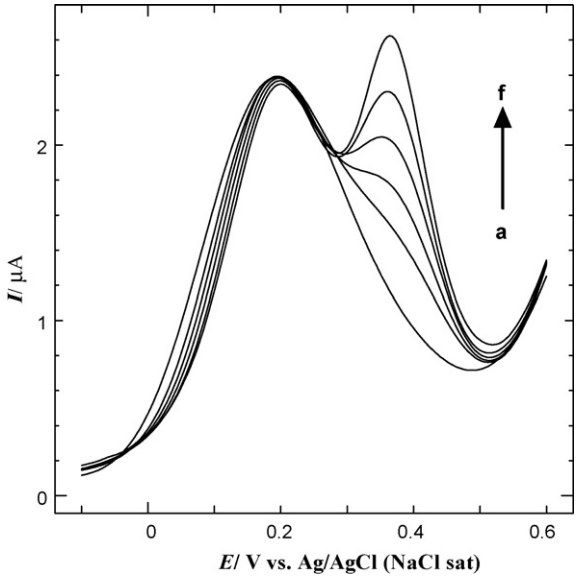



**Fig. 6.** DPVs obtained for PA in the presence of 400 μM of AA at Au/HDT/TOAB-AuNPs modified electrode in 0.20 M PB solution (pH 7.20). Each addition of PA increased by 10 μM: (a) 0, (b) 10, (c) 20, (d) 30, (e) 40 and (f) 50 μM. Pulse width = 0.06 s, amplitude = 0.05 V, sample period = 0.02 s and pulse period = 0.2 s.

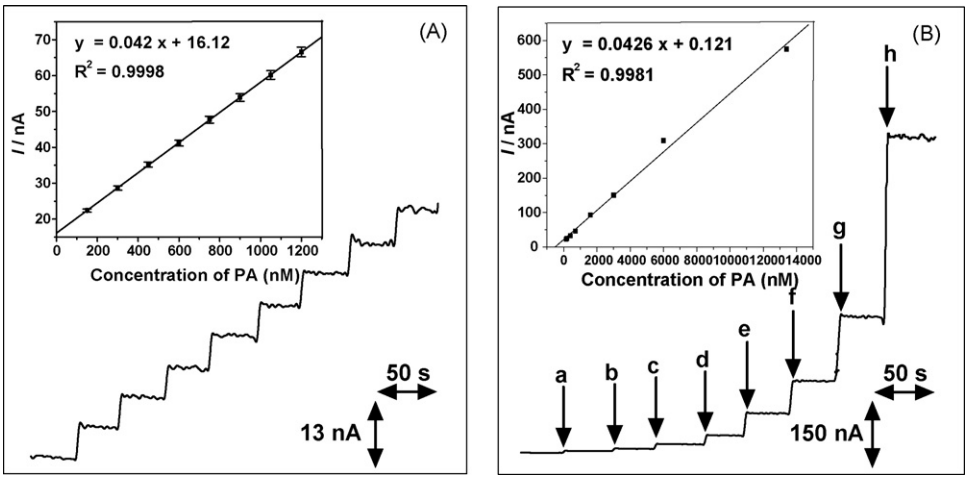



**Fig. 7.** (A) Amperometric current *vs* time curve for the detection of PA at Au/HDT/TOAB-AuNPs electrode. 150 nM of PA was injected into the stirred solution of 0.2 M PB solution (pH 7.20) at regular intervals of 50 s. $E_{app}$ = 0.70 V. Inset shows the corresponding calibration plot. (B) Amperometric *i–t* curve for the determination of PA at Au/HDT/TOAB-AuNPs electrode in the wide range of concentrations: (a) 150, (b) 200, (c) 400, (d) 700, (e) 1600, (f) 3000, (g) 6000 and (h) 13,400 nM in 0.20 M PB solution at an applied potential of 0.70 V.

### *3.4. Amperometric determination of PA using TOAB-AuNPs modified electrode*

Amperometric *i–t* curve was used to find the lowest detection limit of PA at Au/HDT/TOAB-AuNPs electrode. Fig. 7A shows *i–t* curve for the detection of PA using TOAB-AuNPs modified electrode. 150 nM of PA was injected into the homogeneously stirred solution of 0.20 M PB solution by applying a constant potential of 0.70 V. The AuNPs modified electrode showed the initial current response due to 150 nM PA and addition of further 150 nM PA in each step with a sample interval of 50 s, the current response due to PA increased and a steady state current response was attained within 3 s. Each addition of 150 nM PA is associated with a current response of 6.4 nA. Thus, the sensitivity of 43 nA was obtained for 1 μM of PA. The current response of PA increased linearly from 150 nM to 1.20 μM (inset of Fig. 7A) with a correlation coefficient of 0.9998.

Further, we have also carried out the amperometric determination for PA with wide range of concentrations using Au/HDT/TOAB-AuNPs (Fig. 7B). The modified electrode showed the initial current response due to 150 nM PA and further addition of 200 nM PA into the same solution with a sample interval of 50 s, the current response was increased and a steady state current response was attained within 3 s. Again the current response was increased for further addition of 400, 700, 1600, 3000, 6000 and 13,400 nM PA to the same solution with a sample interval of 50 s. The amperometric current was increased linearly while increasing the concentration of PA from $1.50 \times 10^{-7}$ to $1.34 \times 10^{-5}$ M at TOAB-AuNPs modified electrode with a correlation coefficient of 0.9981 (inset of Fig. 7B) and the detection limit was found to be 2.6 nM ($S/N = 3$).

The determination of PA using the present modified electrode is superior than the reported carbon based [1,5,17–22] and AuNPs modified [23,24] electrodes with respect to sensitivity, selectivity and time consumption for the fabrication of modified electrode. For example, a detection limit of $2.5 \times 10^{-7}$ M PA was reported from the calibration plot ($S/N = 3$) at polyaniline-multiwalled carbon nanotube modified electrode [17]. Similarly, a detection limit of $1.8 \times 10^{-7}$ M was reported at seed mediated AuNPs modified electrode again from the calibration plot ($S/N = 3$) [23]. However, using the present modified electrode, a 100-fold higher detection limit ($2.6 \times 10^{-9}$ M ($S/N = 3$)) was achieved for PA. Further, it has been shown that the current due to PA was affected at seed mediated AuNPs modified electrode in the presence of 5-fold higher concentration of AA [23]. However, as demonstrated in Fig. 6, even in the presence of 20-fold higher concentration of AA, the signal due to PA was not affected at TOAB-AuNPs modified electrode. Furthermore, the modification of the present electrode is simple when compared to the reported AuNPs modified carbon paste electrode [24]. In addition, the time required for the modification (~4 h) was less than the time required for the modification of the reported AuNPs (24 h) electrode [25].

### *3.5. Stability and reproducibility of TOAB-AuNPs modified electrode*

The TOAB-AuNPs modified electrode was very stable while kept in 0.20 M PB solution. Further to examine the stability of the present modified electrode towards the determination of PA, DPV for PA was carried out every 10 min interval time. It was found that the oxidation current of PA remains the same with a relative standard deviation of 1.30% for 20 times repetitive measurements indicating that this electrode has a good durability and reproducibility. No sign of surface fouling was observed during the voltammetric measurements. To further ascertain the electrode sample-to-sample reproducibility of the results, three different TOAB-AuNPs modified electrodes were prepared and their response towards the oxidation of 0.50 mM each AA and PA was tested by 20 repeated measurements. The separation between the voltammetric peaks of AA and PA remains same at all the three electrodes. The peak current obtained in the 20 repeated measurements of three independent electrodes showed a relative standard deviation of 1.50%, confirming that the results are reproducible. The above results show that the present modified electrode was very much stable and reproducible towards the determination of PA in the presence of AA.

### *3.6. Determination of PA in human blood plasma and commercial drugs*

The practical application of the present method was demonstrated by determining the concentration of PA in human blood plasma samples and two commercial drugs (paralab, Laborate and paracetamol, Glaxo Smithkline, India). The standard addition method was used for the determination of PA. The recovery results are given in Tables 1 and 2. The DPVs obtained for human blood plasma in 0.2 M PB solution after the injection of 10 and 20 μM commercial PA showed signals due to PA oxidation (Supporting Information, Fig. S5). The observed clear signal for PA suggested that AuNPs modified electrode could be used for the determination of PA in blood plasma sample. Further, the obtained better recovery

**Table 1**
Determination of paracetamol in human blood plasma samples.

| Human blood serum | Added (μM) | Found (μM) | Recovery (%) | RSD (%) |
|---|---|---|---|---|
| Sample 1 | 10 | 10.00 | 100.00 | 1.7 |
| Sample 2 | 20 | 19.99 | 99.95 | 2.0 |

Three replicate measurements were made on the sample.

**Table 2**
Determination of paracetamol in commercial drugs.

| Tablet | Labeled (500 mg/tablet) | Found | Recovery | RSD (%) |
|---|---|---|---|---|
| Paralab | 500 | 503.24 | 100.65 | 1.7 |
| Paracetamol | 500 | 502.53 | 100.51 | 1.9 |

Three replicate measurements were made on the sample.

in Tables 1 and 2 indicated that TOAB-AuNPs modified electrode can be used for the determination of PA both in blood plasma and commercial drug samples.

## 4. Conclusion

The voltammetric results reported in this paper clearly demonstrated that the electrochemical oxidation of PA was facilitated by TOAB-AuNPs modified electrode. The attachment of TOAB-AuNPs on HDT monolayer was confirmed by ATR-FT-IR spectroscopy and AFM. The TOAB-AuNPs modified electrode enhanced the oxidation current of PA twice and shifting its oxidation potential to 70 mV less positive potential in contrast to bare Au electrode. The AuNPs modified electrode also separated the oxidation potentials of AA and PA by 210 mV while bare Au electrode failed to resolve them. Since the voltammetric signals of AA and PA are well separated at the TOAB-AuNPs modified electrode, the measurement of PA in the presence of AA or simultaneous determination of both of them was achieved. The amperometric current was increased linearly while increasing the concentration of PA from $1.50 \times 10^{-7}$ to $1.34 \times 10^{-5}$ at TOAB-AuNPs modified electrode and the detection limit was found to be 2.6 nM ($S/N = 3$). The practical application of the present method was demonstrated by determining the concentration of PA in human blood plasma and commercial drugs.

## Acknowledgements

This work was financially supported in part by Grant-in-Aids from MEXT of Japan for Scientific Research on Priority Area "Strong Photon-Molecule Coupling Fields (No. 470)" and Exploratory Research (No.18655060 for TS) and by Nippon Sheet Glass Foundation for Materials Science and Engineering. SAJ thanks the Japan Society for the Promotion of Science (JSPS) for the financial support under "Invitation Fellowship for Foreign Researchers" Scheme. We thank Dr. Toyota, Nagasaki University for her assistance in AFM measurements and Mr. P. Kalimuthu, GRU for amperometric measurements.

## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.electacta.2009.06.077.

## References

[1] S.-F. Wang, F. Xie, R. Hu, Sens. Acutators B 123 (2007) 495.
[2] M. Boopathi, M.S. Won, Y.B. Shim, Anal. Chim. Acta 512 (2004) 191.
[3] R.M.D. Carvalho, R.S. Freire, S. Rath, L.T. Kubota, J. Pharm. Biomed. Anal. 34 (2004) 871.
[4] M.E. Bosch, A.J.R. Saĭnchez, F.S. Rojas, C. Bosch Ojeda, J. Pharm. Biomed. Anal. 42 (2006) 291.
[5] N. Wangfuengkanagul, O. Chailapakul, J. Pharm. Biomed. Anal. 28 (2002) 841.
[6] M.J.A. Cañada, M.I.P. Reguera, A.R. Medina, M.L. Fernández de Córdova, A.M. Díaz, J. Pharm. Biomed. Anal. 22 (2000) 59.
[7] F.L. Martin, A.E. McLean, Drug Chem. Toxicol. 21 (1998) 477.
[8] N.A. Buckley, I.M. Whyte, D.L. O'Connell, A.H. Dawson, Clin. Toxicol. 37 (1999) 753.
[9] A. Yesilada, H. Erdogan, M. Ertan, Anal. Lett. 24 (1991) 129.
[10] M.K. Srivastava, S. Ahmad, D. Singh, I.C. Shukla, Analyst 110 (1985) 735.
[11] F.A. Mohamed, M.A. Abdallah, S.M. Shammat, Talanta 44 (1997) 61.
[12] S. Ravisankar, M. Vasudevan, M. Gandhimathi, B. Suresh, Talanta 46 (1998) 1577.
[13] M.H. Ramos, T.F. Tyson, D.J. Curran, Anal. Chim. Acta 364 (1998) 107.
[14] N. Al-zoubi, J.E. Koundourellis, S. Malamataris, J. Pharm. Biomed. Anal. 29 (2002) 459.
[15] A. Ruiz-Medina, M.L. Fernandez de cordova, M.J. Ayora-Canada, M.I. Pascual-Reguera, Molina-Diaz, Anal. Chim. Acta 404 (2000) 131.
[16] J. Wang, Analytical Electrochemistry, 2nd ed., Wiley-VCH, New York, 2000.
[17] M. Li, L. Jing, Electrochim. Acta 52 (2007) 3250.
[18] S.F. Fabiana, M.A.B. Christopher, L. Angnes, J. Pharm. Biomed. Anal. 43 (2007) 1622.
[19] R.N. Goyal, S.P. Singh, Electrochim. Acta 51 (2000) 3008.
[20] Y. Tu, Y. Lin, W. Yantasee, Z. Rena, Electroanalysis 17 (2005) 79.
[21] R.T. Kachoosangi, G.G. Wildgoose, R.G. Compton, Anal. Chim. Acta 618 (2008) 54.
[22] C. Radovan, C. Cofan, D. Cinghita, Electroanalysis 20 (2008) 1346.
[23] R.N. Goyal, V.K. Gupta, M. Oyama, N. Bachheti, Electrochem. Commun. 7 (2005) 803.
[24] Z. Xu, Q. Yue, Z. Zhang, D. Xiao, Microchim. Acta 164 (2009) 387.
[25] M.C. Daniel, D. Astruc, Chem. Rev. 104 (2004) 293.
[26] A. Shipway, N.E. Katz, I. Wilner, Chem. Phys. Chem. 1 (2000) 18.
[27] A. Sivanesan, P. Kannan, S.A. John, Electrochim. Acta 52 (2007) 8118.
[28] K.C. Graber, R.G. Freeman, M.B. Hommer, M.J. Natan, Anal. Chem. 409 (1996) 137.
[29] T. Sagara, N. Kato, N. Nakashima, J. Phys. Chem. B 106 (2002) 1205.
[30] A. Yu, Z. Liang, J. Cho, F. Caruso, Nano Lett. 3 (2003) 1203.
[31] S.A. John, T. Sagara, J. Electroanal. Chem., in press.
[32] M. Brust, M. Walker, D. Bethell, D.J. Schiffrin, R. Whyman, J. Chem. Soc., Chem. Commun. (1994) 801.
[33] L. Alfanta, E. Katz, I. Willner, Anal. Chem. 72 (2000) 927.
[34] A.L. Abderlrahman, A.K. Mohammad, T. Okajima, T. Ohsaka, J. Phys. Chem. B 110 (2006) 2798.