



# Simultaneous determination of ascorbic acid, dopamine and uric acid based on tryptophan functionalized graphene



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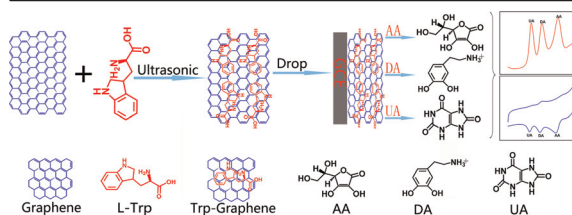
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## HIGHLIGHTS

- Trp-GR was synthesized by utilizing a facile ultrasonic method.
- The material as prepared had well dispersivity in water and better conductivity than pure GR.
- Trp-GR/GCE showed excellent potential for the determination of AA, DA and UA.
- The proposed method was applied for the analysis of AA, DA and UA in real samples.

## GRAPHICAL ABSTRACT



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## ABSTRACT

A new type of tryptophan-functionalized graphene nanocomposite (Trp-GR) was synthesized by utilizing a facile ultrasonic method via  $\pi$ - $\pi$  conjugate action between graphene (GR) and tryptophan (Trp) molecule. The material as prepared had well dispersivity in water and better conductivity than pure GR. The surface morphology of Trp-GR was characterized by scanning electron microscopy (SEM), transmission electron microscopy (TEM) and Raman spectroscopy. The electrochemical behaviors of ascorbic acid (AA), dopamine (DA), and uric acid (UA) were investigated by cyclic voltammetry (CV) on the surface of Trp-GR. The separation of the oxidation peak potentials for AA–DA, DA–UA and UA–AA was about 182 mV, 125 mV and 307 mV, which allowed simultaneously determining AA, DA, and UA. Differential pulse voltammetry (DPV) was used for the determination of AA, DA, and UA in their mixture. Under optimum conditions, the linear response ranges for the determination of AA, DA, and UA were 0.2–12.9 mM, 0.5–110  $\mu$ M, and 10–1000  $\mu$ M, with the detection limits ( $S/N=3$ ) of 10.09  $\mu$ M, 0.29  $\mu$ M and 1.24  $\mu$ M, respectively. Furthermore, the modified electrode was investigated for real sample analysis.

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## 1. Introduction

Ascorbic acid (AA) is an antioxidant that plays significant roles in proper functioning of human metabolism and central nervous system. It can prevent or treat common cold, mental illness and all types of cancer [1]. Dopamine (DA) is one of the important neurotransmitters for message transfer in the central nervous

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system. Abnormal levels of DA are the symptoms of some neurodegenerative diseases like Alzheimer's and Parkinson's [2]. In addition, uric acid (UA) is the primary end product of purine metabolism and long-standing elevated uric acid levels will cause gout and kidney problems [3]. As important molecules for physiological processes in human metabolism, AA, DA and UA usually coexist together in living systems. Since AA, DA and UA usually foul the detection electrodes or their oxidation potentials are severely overlapped, their electrochemical determination is still a challenge [4]. To overcome these difficulties, many types of materials have been employed to modify electrodes such as metal nanoparticles [5], conducting polymers [6], and carbon-based materials [7] for the electrochemical detection of AA, DA and UA and showed excellent results.

Graphene (GR) is a two-dimensional nanomaterial consisting of a single layer of  $sp^2$  network of carbon atoms. It is an ideal material for electrochemistry because of its very large 2D electrical conductivity, large surface area and low cost [8]. However, graphene is hydrophobic and tends to agglomerate irreversibly or even restack to form graphite through van der Waals interactions and strong  $\pi$ - $\pi$  stacking, which limits its further application [9]. In recent years, great efforts have been made to improve the dispersity of graphene, such as poly (diallyldimethylammonium chloride) (PDDA) functionalized graphene [10], porphyrin-functionalized graphene [2], stacked graphene platelet nanofibers (SGNF)/ionic liquid (IL)/chitosan (CS) modified electrode [11].

Tryptophan (Trp), an essential amino acid for the biosynthesis of proteins, which is important in nitrogen balance and the maintenance of muscle mass and body weight in humans, has an indole conjugate structure and can theoretically stabilize the aqueous dispersion of graphene through  $\pi$ - $\pi$  interactions [12,13]. Herein we synthesized tryptophan-functionalized graphene nanocomposite (Trp-GR) through appropriate ultrasonic treatment, which avoided the use of reducing agents and high temperature. The Trp-GR could be dispersed well in water and stabilized for a few months without precipitation. Furthermore, the Trp-GR was applied to the electrochemical determination of AA, DA, and UA. The main idea was illustrated in Scheme 1.

## 2. Experimental

### 2.1. Reagents and materials

All chemicals and reagents were of analytical grade and were used without further purification. Graphene (prepared by chemical vapor deposition, CVD) was purchased from XF NANO, INC. (Nanjing, China). L-Tryptophan and ascorbic acid were purchased from Zhongqin Chemical Reagent Co., Ltd. (Shanghai, China). Uric

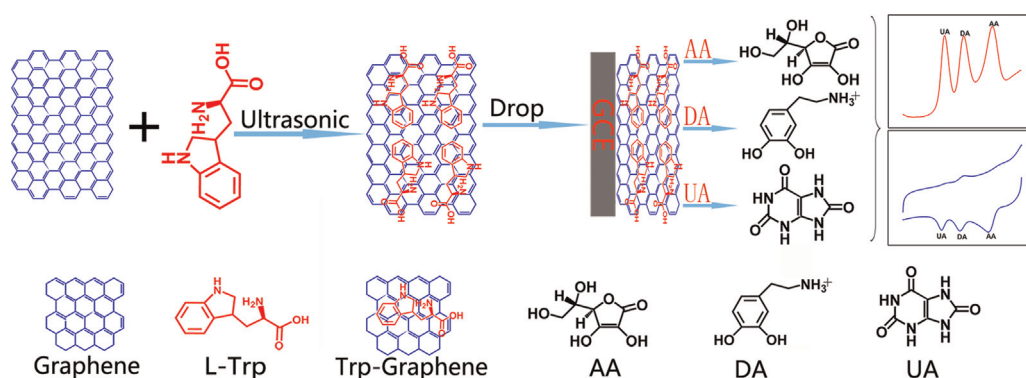
acid was obtained from Alfa Aesar Chemical Co., Ltd. (Tianjin, China). Dopamine was from Aladdin Chemistry Co., Ltd. (Shanghai, China). Formic acid and *N,N*-dimethyl formamide (DMF) were supplied by China Medicine Reagent Co. (Shanghai, China). The 0.1 M standard phosphate buffer solutions with different pH values (4.0, 5.0, 6.0, 7.0, 8.0, 9.0 and 10.0) made from the salts  $K_2HPO_4$  and  $KH_2PO_4$  by adding 1.0 M KOH aqueous solution and 1.0 M phosphoric acid aqueous solution adjusted, were employed as the supporting electrolyte. Double distilled water and nitrogen gas (99.999%) were used for all experiments. All experiments were performed at room temperature.

### 2.2. Instruments and measurements

The scanning electron microscopy (SEM) image was collected on a JSM-6701F (Japan) at 5.0 KV. HRTEM image was obtained by FEI-Tecna G2 TF20 electron microscope (America) operating at HT200 kV. Raman spectra were obtained by inVia Renishaw confocal spectroscopy (Britain) with 633 nm laser excitation. Electrochemical impedance spectra (EIS) measurements were conducted by Princeton Applied Researcher (USA). Impedance measurements were performed with the frequencies swept from  $10^4$  to 0.1 Hz. A Randle's equivalent circuit was used to fit the obtained impedance spectra. Electrochemical experiments and measurements were performed at a CHI660B electrochemical workstation (Shanghai Chenhua Co., China). A conventional three-electrode system was used throughout the measurements. A bare or modified glassy carbon electrode (GCE, CHI104,  $\varphi = 3$  mm, Gaoss Union Co., Ltd., Wuhan, China) was employed as a working electrode, a platinum column as the counter electrode and an Ag/AgCl electrode (saturated KCl) as the reference. Individual or simultaneous determination of AA, DA and UA was carried out using cyclic voltammetry (CV) and differential pulse voltammetry (DPV).

### 2.3. Synthesis of tryptophan-functionalized graphene

The Trp-GR was synthesized by a modified method based on the literature [14]. In a typical experiment, 20.0 mg Trp was dissolved in 1.0 mL formic acid to form a stable solution. Then the solution was mixed with 1.0 mg of graphene. After 2 h ultrasonication, the mixture was allowed to add double distilled water to 10 mL and sonicated for an additional 4 h, resulting in a homogeneous black dispersion solution. The solution was centrifuged at 10,000 rpm for 15 min, and the supernatant was poured away, the precipitate was carefully washed with double distilled water 3–5 times till the extra Trp was removed. At last, the right amount of double distilled water was added in the precipitate to obtain  $0.2 \text{ mg mL}^{-1}$  Trp-GR dispersion solution.



**Scheme 1.** Schematic diagram of the fabrication procedure Trp-GR, Trp-GR/GCE, and the Trp-GR/GCE was used direct determination of AA, DA, and UA.

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