

## ELECTRONIC SUPPLEMENTARY INFORMATION

### **A novel MIP-ECL sensor based on RGO-CeO<sub>2</sub>NPs / Ru(bpy)<sub>3</sub><sup>2+</sup>- Chitosan for ultratrace determination of trimipramine**

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**Materials and reagents**

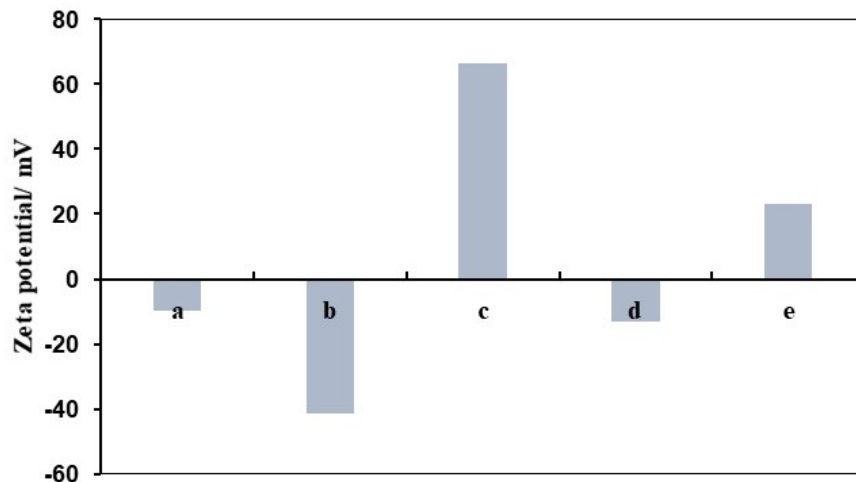
Graphite powder (99.995%), chitosan, sulfuric acid (95%), potassium nitrate (99%), potassium permanganate (99%), NH<sub>4</sub>OH (aqueous solution), N<sub>2</sub>H<sub>4</sub>, chloroform, methanol, ethanol, and urea [CO(NH<sub>2</sub>)<sub>2</sub>] were bought from Merck Chemical Co. (Germany).

Tris(2,2'-bipyridyl) ruthenium(II) (Ru(bpy)<sub>3</sub><sup>2+</sup>) chloride hexahydrate, 2,2'-azobisisobutyronitrile (AIBN) and Ce(NO<sub>3</sub>)<sub>3</sub>·6H<sub>2</sub>O were supplied from Sigma–Aldrich Co. (USA). Vinyl benzene (VB) and methacrylic acid (MAA) were supplied from Sigma–Aldrich (Munich, Germany) and purified via passing through a short column filled with neutral alumina followed by distilling under reduced pressure. Ethylene glycol dimethacrylate (EDMA) was purchased from Fluka (Buchs, Switzerland). The as-supplied EDMA was distilled under reduced pressure in the presence of a hydroquinone inhibitor, followed by storage at 4 °C until use. For ECL determination, the phosphate buffer (pH=7.4) was provided by mixing the aqueous solutions of 0.1M K<sub>2</sub>HPO<sub>4</sub>, 0.1M NaH<sub>2</sub>PO<sub>4</sub>, and 0.1 M KCl. All of the chemicals and reagents consumed were of analytical grade and directly used with no further purification. Trimipramine (TRI) powder was bought from Shenzhen Xiangyun Chemical Co. Ltd. (Shenzhen, China).

## **Apparatus**

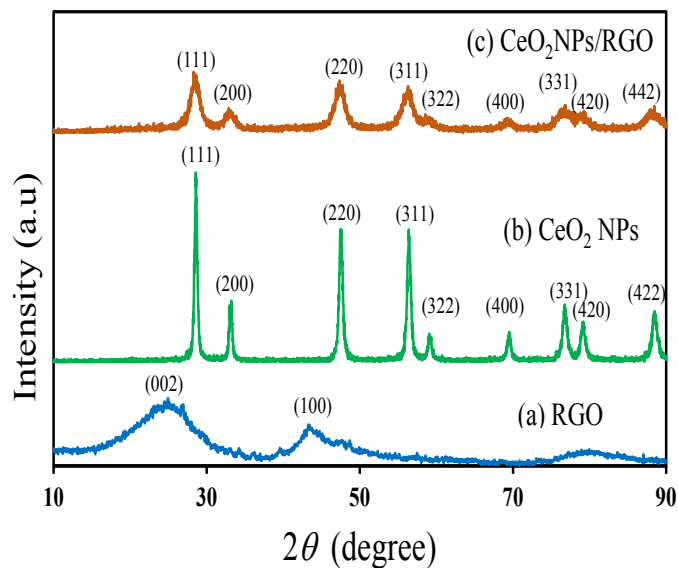
A MPI-B ECL Analyzer Systems (Remax, China) was used for monitoring the ECL emission signals. An AUTOLAB PGSTAT 30 potentiostat/galvanostat (Eco Chemie B.V., Utrecht, Netherlands) apparatus was used for electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV) tests. A modified GCE ( $\phi=3$  mm), a Pt spiral wire, and an Ag/AgCl electrode were applied in the used three-electrode system as the working electrode, the counter electrode, and the reference electrode, respectively. A Philips (USA) PW-1730 X-ray diffractometer (XRD) by CuK $\alpha$  radiation ( $\lambda=1.5405\text{\AA}$ ) was employed for characterizing the crystalline phases of the samples. The morphology and the structure of the nanomaterials were examined by transmission electron microscope (TEM) (Philips EM 2085, USA) and field-emission scanning electron microscopy (FE-SEM) using a Zeiss SIGMA VP with the gold coating (Zeiss, Germany).

## 1. Supplementary Note 1: Characterization of prepared CeO<sub>2</sub>NPs-RGO nanocomposite



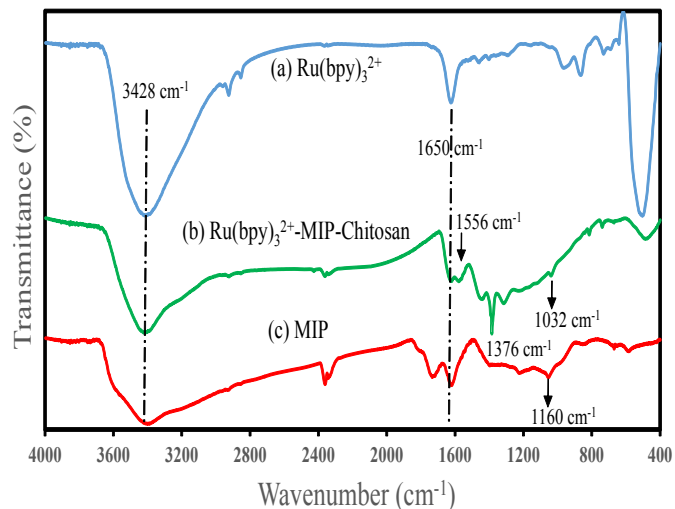
**Fig. S1.** Zeta potential of (a) RGO, (b) CeO<sub>2</sub> NPs/RGO, (c) Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP- CS, (d) MIP, (e) CeO<sub>2</sub>NPs/RGO- Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-CS

In order to confirm the proposed strategy for MIP-ECL sensor, Zeta potential was measured for RGO, CeO<sub>2</sub> NPs/RGO, Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-CS, MIP and CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-CS to explore electric charge on the surfaces. The obtain results showed that: It can be found that Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-CS is positively charged while CeO<sub>2</sub>NPs/RGO and MIP are negatively charged, which can support our strategy for MIP-ECL sensor. Therefore, these results suggested a successful method for fabrication of MIP-ECL sensor.



**Fig. S1 (A).** XRD patterns of (a) RGO, (b) CeO<sub>2</sub>NPs, and (c) CeO<sub>2</sub>NPs/RGO.

A broad peak and a small peak seen around  $2\theta$  of  $26^\circ$  and  $42^\circ$  can be attributed to (002) and (100) planes of the graphite-like structure, respectively. Moreover, in the diffraction patterns of CeO<sub>2</sub> and CeO<sub>2</sub>-RGO, the peaks appeared at  $2\theta$  of  $28.58^\circ$ ,  $33.17^\circ$ ,  $47.50^\circ$ ,  $56.41^\circ$ ,  $59.14^\circ$ ,  $69.49^\circ$ ,  $76.80^\circ$ ,  $79.21^\circ$ , and  $88.38^\circ$  can be attributed to the planes of (111), (200), (220), (311), (322), (400), (331), (420), and (422) as well as reflections of CeO<sub>2</sub>, respectively.

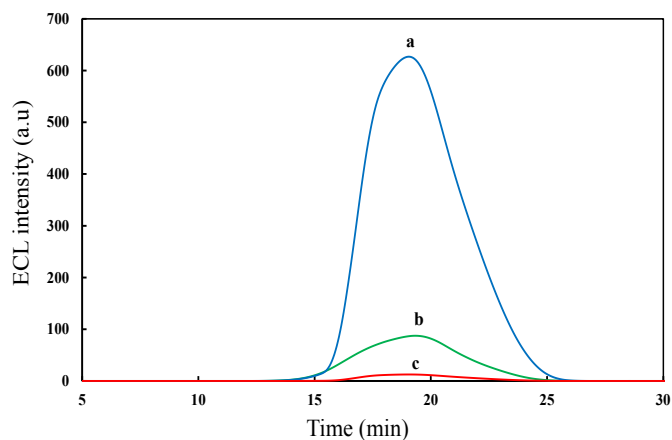


**Fig. S1 (B).** (a) FT-IR spectra (a)  $\text{Ru}(\text{bpy})_3^{2+}$ , (b)  $\text{Ru}(\text{bpy})_3^{2+}$ -MIP-Chitosan, and (c) MIP.

The peak appeared in the wavenumber of  $3428\text{ cm}^{-1}$  may be assigned to the vibrational mode of O-H. The peak at about the wavenumber of  $1650\text{ cm}^{-1}$  may be attributed to C=O stretching mode.

The peak appeared around the wavenumbers of  $1160\text{ cm}^{-1}$  and  $1032\text{ cm}^{-1}$  are attributed to C-O and C-C stretching modes, respectively. The described spectra demonstrate the carboxyl groups presenting in MIP and  $\text{Ru}(\text{bpy})_3^{2+}$ -MIP-Chitosan structures. The peaks appeared at the wavenumbers of  $1556\text{ cm}^{-1}$  and  $1376\text{ cm}^{-1}$  attributed to the superposition of C=C and C=N bonds vibrations confirm the bipyridine ring vibrations.

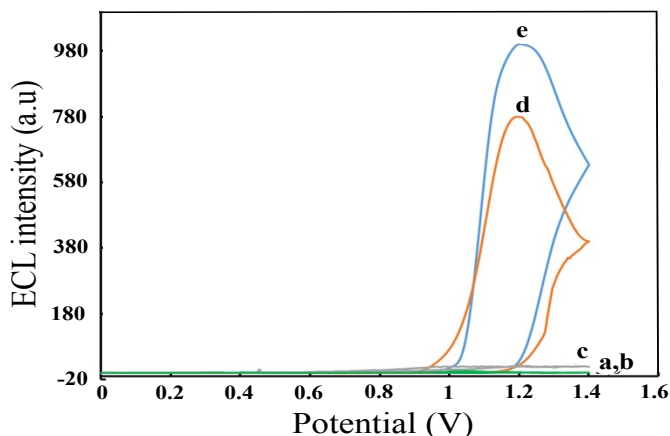
Furthermore, the absorption spectrum of  $\text{Ru}(\text{bpy})_3^{2+}$ -MIP-Chitosan composite has the characteristic peaks of both MIP and  $\text{Ru}(\text{bpy})_3^{2+}$ , which confirms the  $\text{Ru}(\text{bpy})_3^{2+}$  locating on the surface/interface of MIP film.



**Fig. S1 (C).** ECL responses of MIP-ECL sensor (a) after TRI readsorption, (b) before TRI readsorption (10 pM), and (c) NIP-ECL sensor after TRI adsorption (10 pM) in 0.1 M phosphate buffer (pH = 7.4).

Accordingly, the ECL intensity of the MIP-ECL sensor after TRI readsorption (curve a) was significantly more intense than that before TRI readsorption (curve b) and also more intense than ECL of the NIP-ECL sensor after TRI adsorption (curve c). It seems that TRI can be oxidized on the prepared electrode generating cation radicals of  $\text{TRI}^+$ . The short-lived  $\text{TRI}^+$  cation radicals could lose a proton leading to a strong reductant, i.e.,  $\text{TRI}^\cdot$  intermediate.  $\text{TRI}^\cdot$  has the potential of  $\text{Ru}(\text{bpy})_3^{3+}$  reducing to  $\text{Ru}(\text{bpy})_3^{2+*}$  generating ECL emission.

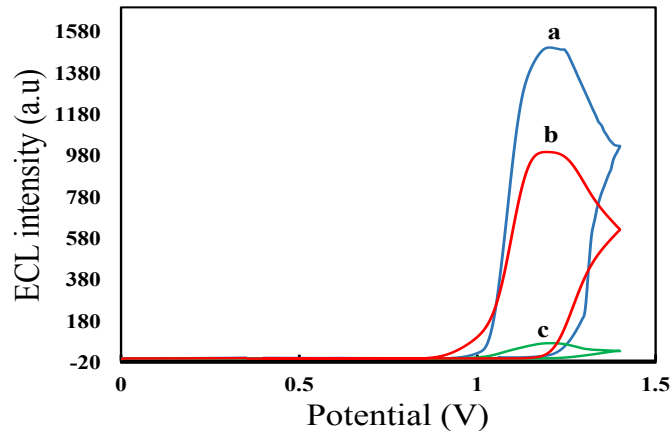
## 2. Supplementary Note 2: Electrochemical and ECL behaviors of MIP-ECL sensor



**Fig. S2 (A).** ECL signals of (a) Bare GCE, (b) CeO<sub>2</sub>NPs/RGO, (c) CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-CS with no TRI, (d) CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-CS in presence of 100 pM TRI, and (e) CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-CS in presence of 100 pM TRI (Scan rate: 100 mVs<sup>-1</sup>, potential range: 0.0-1.4 V).

Bare GCE, as well as the GCE modified with CeO<sub>2</sub>NPs/RGO, had no considerable ECL emission (curves a and b). This may be attributed to no Ru(bpy)<sub>3</sub><sup>2+</sup> existing on these electrodes. However, the electrode modified with CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-Chitosan showed a weak ECL signal assigned to the reaction of electro generated Ru(bpy)<sub>3</sub><sup>2+</sup> in the absence of TRI (curve c). These observations may be attributed to the effect raising by Ru(bpy)<sub>3</sub><sup>2+</sup>. The electrode modified with CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-Chitosan created an intense signal in the presence of TRI (curve d). The signal produced by the electrode modified with CeO<sub>2</sub>NPs/RGO-Ru(bpy)<sub>3</sub><sup>2+</sup>-MIP-Chitosan was still more intense (curve e), which can be attributed to the higher TRI uptake from solution by MIP composite film. This observation demonstrates that the MIP film can provide more binding sites (i.e., cavities) for uptaking TRI.

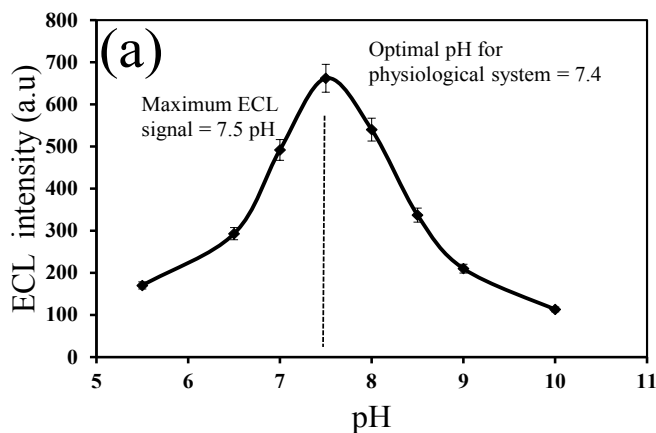




**Fig. S2 (B).** (a) As-prepared imprinted film with a high TRI concentration, (b) Eluted imprinted polymer immersed in the TRI standard solution (50 pM, 15 min), and (c) Eluted imprinted film after TRI removal.

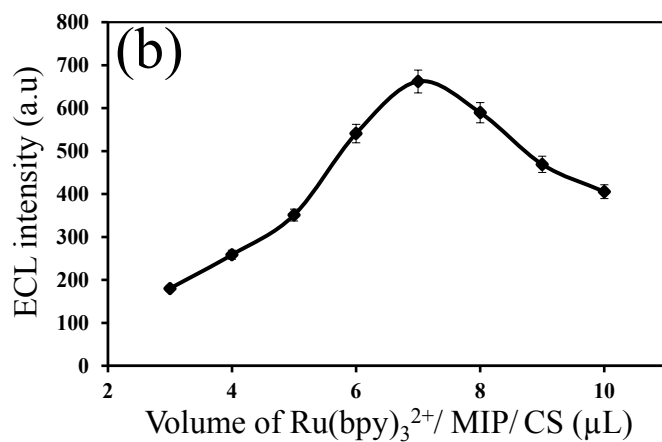
Thanks to the high TRI concentration, the as-prepared imprinted film had the highest ECL signal intensity (a). After rinsing the imprinted film, the ECL intensity lowered demonstrating the efficient TRI template removal from the imprinted film via rinsing (c). Immersing the imprinted polymer in the TRI standard solution (50 pM) for a duration time of 15 min leads to rebind the TRI template molecule with the imprinted film, and consequently, the ECL signal intensity increased (b).

### 3. Supplementary Note 3: Optimization of detection conditions



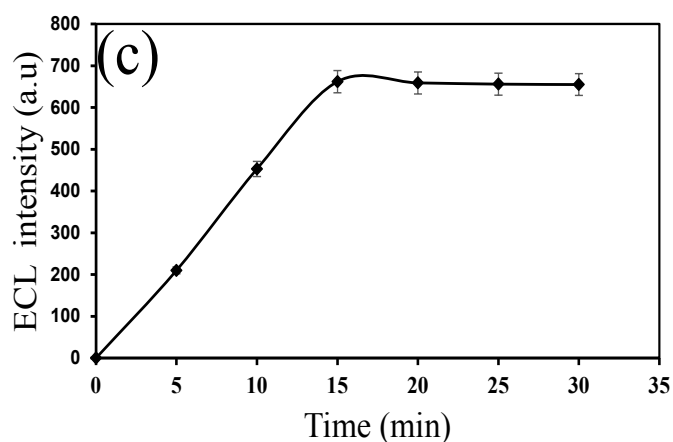
**Fig. S3 (A).** ECL signal intensity of MIP-ECL sensor in 0.1 M phosphate buffer containing 10 pM TRI with different pH values.

The ECL signal intensity increased significantly by increasing pH from 5.5 to 7.5 and subsequently reduced. The maximum intensity of ECL signal was gained at pH = 7.5. The higher pH values leads to the lower ECL signal intensity which was ascribed to  $\text{OH}^-$  ions undergoing a competitive reaction with  $\text{Ru}(\text{bpy})_3^{2+}$ . Considering the potential of the MIP-ECL sensor applying while in vivo monitoring, a buffer of 0.1 M phosphate with pH = 7.4, which is equal to the physiological pH, was employed as the supporting electrolyte for the subsequent MIP-ECL sensor evaluations.



**Fig. S3 (B).** ECL signal intensity of MIP-ECL sensor loaded with different amount of Ru(bpy)<sub>3</sub><sup>2+</sup>/MIP/CS composite in 0.1 M phosphate buffer (pH = 7.4) containing 10 pM TRI.

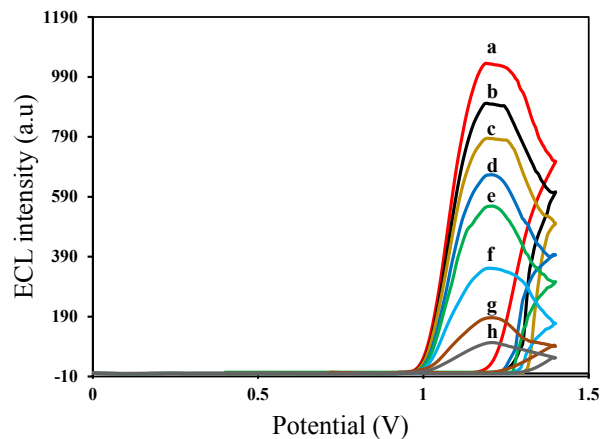
Through modifying the GCE, as the amount of the applied Ru(bpy)<sub>3</sub><sup>2+</sup>/MIP/Chitosan composite increases up to 7 μL, ECL signal intensity increases continuously and decreases after that. This may be explained by the fact that a more amount of the Ru(bpy)<sub>3</sub><sup>2+</sup>/MIP/Chitosan composite leads to more TRI and Ru(bpy)<sub>3</sub><sup>2+</sup> molecules presenting on the surface of GCE, leading to a higher ECL response.



**Fig. S3 (C).** Effect of the incubation time on ECL signal intensity of MIP-ECL sensor in 0.1 M phosphate buffer (pH = 7.4) containing 10 pM TRI (Scan rate: 100 mVs<sup>-1</sup>, potential range: 0.0-1.4 V).

Sharply raising of the ECL signal intensity by increasing the incubation time from 5 min to 15 min. After that, however, no raise of the ECL signal intensity occurs by more incubation time. So, considering that the shorter the sensing time, the more desirable, an incubation time of 15 min was considered as the optimal value.

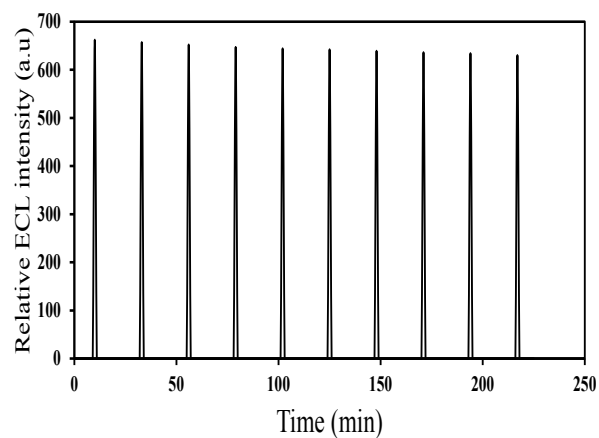
#### **4. Supplementary Note 4: Analytical performance of MIP-ECL sensor**



**Fig. S4.** ECL signals of MIP-ECL sensor with different TRI concentrations labeled as a–h, respectively.  $C(\text{TRI}) = 0.2, 0.5, 1, 5, 10, 25, 50,$  and  $100 \text{ pM}$ , and a potential range from  $0.0$  to  $1.4 \text{ V}$ .

Under the optimal conditions, the MIP-ECL sensor was utilized for detecting different TRI concentrations. The more TRI concentration, the more ECL intensity. There is a direct relationship between the ECL signal intensity and the logarithm of TRI concentration ranging from  $0.2 \text{ pM}$  to  $100 \text{ pM}$ .

## **5. Supplementary Note 5: Selectivity, reproducibility, and stability of MIP-ECL sensor**



**Fig. S5.** Stability of ECL signal in 0.1 M phosphate buffer (pH = 7.4) containing 10 pM TRI during 10 continuous cyclic potential scans of MIP-ECL sensor (Scan rate: 100 mVs<sup>-1</sup>, potential range 0-1.4 V).

10 consecutive cyclic potential scans were performed. The ECL signals emitted by very great intensities with RSD < 2.9%. Additionally, in a long-time test, the sensor ECL signal was evaluated after 10 days. It was examined that the sensor responds with an intensity of 86.4% of its initial value after a duration of 10 days.