



# Determination of serotonin using gold nanorod-terminated carbonaceous electrode by differential pulse voltammetry

İlknur Üstündağ<sup>1</sup> · Samet Şahin<sup>2</sup>

Received: 4 May 2023 / Accepted: 29 July 2023 / Published online: 14 August 2023  
© Institute of Chemistry, Slovak Academy of Sciences 2023

## Abstract

Waste coal tar pitch is a carcinogenic substance but it can be turned into an environmentally safe and high-value electrode material when it is pyrolyzed at high temperatures. In this study, carbonized coal tar pitch (CTP)-based electrode material was first prepared using an eco-friendly green chemistry approach. Gold nanorods (AuNRDs) were then attached to CTP with 4-aminothiophenol junctions by self-assembled monolayers. AuNRDs-terminated carbonaceous material was characterized with microscopy and spectroscopy techniques. The nanocomposite material was modified on a glassy carbon electrode by drop coating under an infrared lamp. An analytical method for the determination of 5-HT was developed on the new carbonaceous electrode using differential pulse voltammetry. The prepared sensor exhibited a limit of detection of 0.03  $\mu\text{M}$  ( $S/N = 3$ ) with a linear range from 0.5 to 10  $\mu\text{M}$  and shows good reproducibility and stability. Finally, real sample tests have been successfully applied to measure 5-HT in human serum to demonstrate the feasibility of the proposed sensing platform.

**Keywords** Serotonin · Differential pulse voltammetry · Gold nanorods · Carbonaceous materials · Coal tar pitch

## Introduction

Serotonin, also known as 5-hydroxytryptamine or 5-HT, is a crucial neurotransmitter involved in regulating various aspects of the emotional system. It plays a vital role in controlling mood, sleep patterns, vomiting, sexual behavior, and appetite (Ressler and Nemeroff 2000; Perry and Fuller 1992). Insufficient levels of serotonin can lead to serious conditions like depression, bipolar disorder, anxiety, and migraines (Lin et al. 1998; Imeri et al. 1999). Conversely, excessive levels of serotonin can result in a life-threatening condition called serotonin syndrome (Isbister et al. 2004). Therefore, the accurate and reliable detection of serotonin in biological samples is of utmost importance.

Various methods have been employed to determine the levels of serotonin (5-HT) in biological samples. These include spectroscopy (Selvarajan et al. 2018), chromatography (Torfs et al. 2012), chemiluminescence (Barnett

et al. 1998), capillary electrophoresis (Coelho et al. 2014), and fluorometry (Panholzer et al. 1999). However, among these methods, *in vitro* electrochemical detection stands out as a cost-effective solution with a low limit of detection ( $\text{LOD} < 1 \mu\text{M}$ ) (Dăscălescu and Apetrei 2021; Song et al. 2021). It is difficult to accurately detect 5-HT in blood samples due to the electrochemical interaction of easily oxidizable species such as ascorbic acid (AA) and dopamine (DA). Therefore, it is important to use a sensitive electrode configuration to accurately differentiate the signal for a short range of oxidation potential in a complex sample such as human serum. Furthermore, inexpensive and simple electrode configurations are essential for practical applications.

Raw coal tar pitch (r-CTP) is a highly toxic mixture and can include very complex aromatic compounds. On the other hand, its rich carbon content makes it an ideal candidate for the preparation of carbonaceous electrode materials (Üstündağ and Erkal 2017). Carbonized coal tar pitch (CTP) can be used as a non-toxic and highly conductive polycrystalline electrode material for electrochemical applications (Erkal et al. 2016). On the other hand, gold nanoparticles are also widely used as an electrode material because of their excellent electrocatalytic activity, electrical conductivity, and high surface area (Rodriguez et al. 2014; Hebié et al. 2013). Especially, shape-controlled gold nanoparticles

✉ Samet Şahin  
samet.sahin@bilecik.edu.tr

<sup>1</sup> Faculty of Arts and Science, Department of Physics,  
Dumlupınar University, 43100 Kutahya, Turkey

<sup>2</sup> Faculty of Engineering, Department of Bioengineering,  
Bilecik Şeyh Edebali University, 11100 Bilecik, Turkey

(AuNPs) supported on high-area carbonaceous materials on the electrode surface were reported as electrochemically active catalysts in the literature (Erikson et al. 2014). The electrochemical sensing of 5-HT has been studied in the literature using different carbon-based materials and nanoparticles such as multi-walled carbon nanotubes (Babaei et al. 2013), graphene oxide (Han et al. 2013), reduced graphene oxide (Rajarithinam et al. 2023; Xue et al. 2014), and AuNPs (Mahato et al. 2019) with the limit of detection values down to the nM scale (Song et al. 2021).

Moreover, most of these studies incorporate metals and alloys in their electrodes such as RuO<sub>2</sub>/CeO<sub>2</sub>-Au nanofibers (Samie and Arvand 2019) and AuAg nanoalloys (Thanh et al. 2017) alongside the carbon nanomaterials. Such systems offer high sensitivity and low limits of detection but also suffer from the use of expensive carbon materials, durability issues, and complicated synthesis processes for practical applications. Therefore, herein, it is aimed to develop a novel electrode configuration using an easy-to-synthesize, low-cost, and non-toxic electrocatalyst for the sensitive detection of 5-HT from biological samples.

This study presents the development of a novel and highly sensitive sensor for the precise determination of serotonin (5-HT) levels. The sensor was constructed by synthesizing gold nanoparticles and subsequently modifying them onto a carbonaceous thin film electrode surface (CTP) using 4-aminothiophenol (4-ATP) as a linker. The resulting nanocomposite was then drop-coated onto a glassy carbon electrode. The nanocomposite and the modified electrode surfaces underwent comprehensive characterization to assess their structural and electrochemical properties. An electroanalytical method was subsequently devised based on the modified electrode for the sensitive detection of 5-HT in human

serum. The objective was to establish a robust and accurate analytical approach for quantifying 5-HT levels in biological samples.

## Experimental

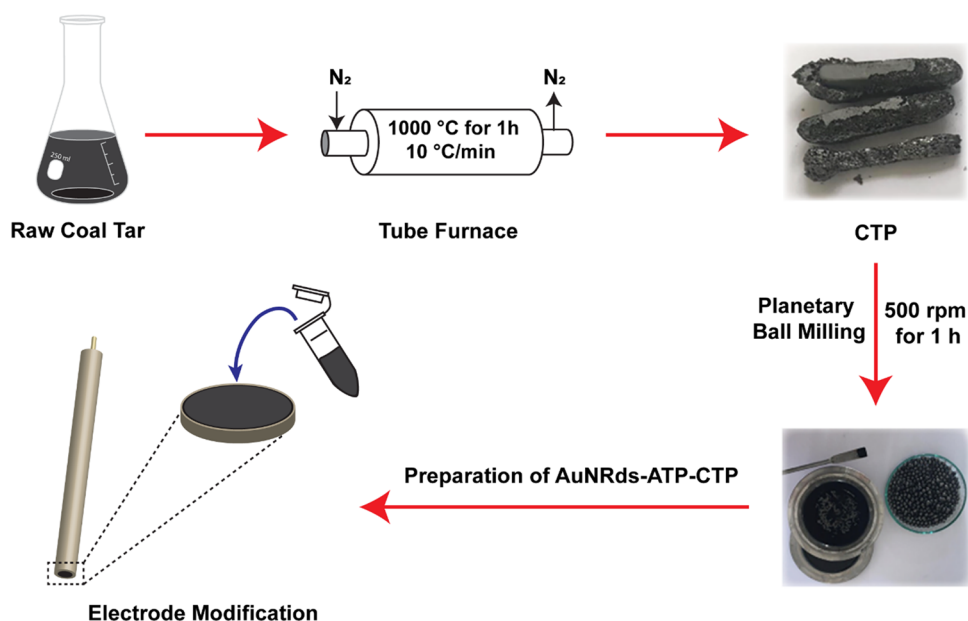
### Materials

All chemicals were obtained from Fluka, Riedel, Merck, and Sigma-Aldrich. CTP was obtained from Orcan Group (Turkey). All electrochemical experiments were performed using an Ivium CompactStat analyzer (Ivium Technologies, Netherlands). In electrochemical experiments, the modified carbonaceous glassy carbon (GC, BASi MF-2012 model,  $\varnothing = 3$  mm, geometric surface area = 7.1 mm<sup>2</sup>) electrode was used as the working electrode, and Ag/AgCl<sub>sat.</sub> and Pt wire were used as a reference and auxiliary electrodes, respectively. The ultrapure water (UPW) used in the experiments has a conductivity of 18.3 M $\Omega$  cm obtained using a Human Power 1<sup>+</sup> purification system.

### Preparation of AuNRds-ATP-CTP/GC electrodes

The pyrolysis process of r-CTP involved heating it at 1000 °C with a heating rate of 10 °C/min under a nitrogen atmosphere for a duration of 1 h. Following pyrolysis, the resulting material was ground using a planetary ball mill (Retsch PM-100) at 500 rpm for 1 h before proceeding with electrode modifications. The schematic representation of the CTP synthesis and electrode modification stages can be found in Fig. 1. To synthesize AuNRds, the seed-mediated method described in the literature was employed (Gole and

**Fig. 1** Schematic diagram of the CTP synthesis and electrode modification stages



Murphy 2004). The synthesized AuNRds were characterized using transmission electron microscopy (TEM, JEOL JEM-2100, Japan) to obtain images and were stored in absolute ethanol at a temperature of +4 °C until further use. The CTP material was synthesized from raw tar using a tube furnace (Protherm, USA) under a nitrogen atmosphere.

Carboxylate-terminated groups were introduced to the CTP material by refluxing it in 4 M HNO<sub>3</sub> for 6 h, following a method described in the literature (Profumo et al. 2006). The CTP material with carboxyl groups was then subjected to treatment with 0.1 M EDC in ethanol for 6 h, and the resulting colloidal solution was centrifuged at 10,000 rpm for 10 min. The activated CTP material was subsequently washed with UPW and ethanol, respectively.

Next, the activated CTP material was combined with a 1-mM solution of 4-ATP in ethanol and stirred on a magnetic stirrer for 6 h. During this process, covalent attachment of 4-ATP to the carbonaceous amide took place, resulting in a modified material referred to as ATP-CTP. The ATP-CTP solution was then centrifuged at 10,000 rpm for 10 min, and the solid material obtained was washed successively with water and ethanol, followed by centrifugation under the same conditions. The final precipitate obtained from the washing steps was added to a solution containing AuNRds, resulting in the formation of a composite electrode material denoted as AuNRds-ATP-CTP.

The preparation steps of the AuNRds-ATP-CTP electrode were characterized with TEM, scanning electron microscopy (SEM, Nova NanoSEM-650, Belgium), and X-ray photoelectron spectroscopy (XPS, PHI 5000 VersaProbe, ULVAC-PHI. Inc., Japan/USA). For the preparation of the modified carbonaceous electrodes, 20 mg of AuNRds-ATP-CTP was diluted with 1 mL of acetonitrile (MeCN) and brought into suspension. Different volumes (i.e., 5, 10, 15, 20, and 25 mL) of the suspension were dropped onto the bare GC electrodes and dried under the IR lamp (75 W) for 10 min.

### Electrochemical characterization and analytical investigations of the modified electrodes

The optimization of GC electrodes modified with various concentrations of carbonized material was carried out using cyclic voltammetry (CV) with a scan rate of 200 mV/s within the voltage range of -0.2 to 0.4 V versus Ag/AgCl<sub>sat.</sub> reference electrode. Subsequently, an electroanalytical method was developed for the accurate determination of serotonin (5-HT) in the concentration range of 0.5–10 μM (specifically, 0.5, 1.0, 1.5, 2.5, 4.0, 5.0, 6.0, 7.5, and 10 μM). This method utilized differential pulse voltammetry (DPV) with specific parameters: a pulse width of 0.06 s, amplitude of 0.05 V, sample period of 0.02 s, and pulse period of 0.20 s. Following the establishment of the calibration curve

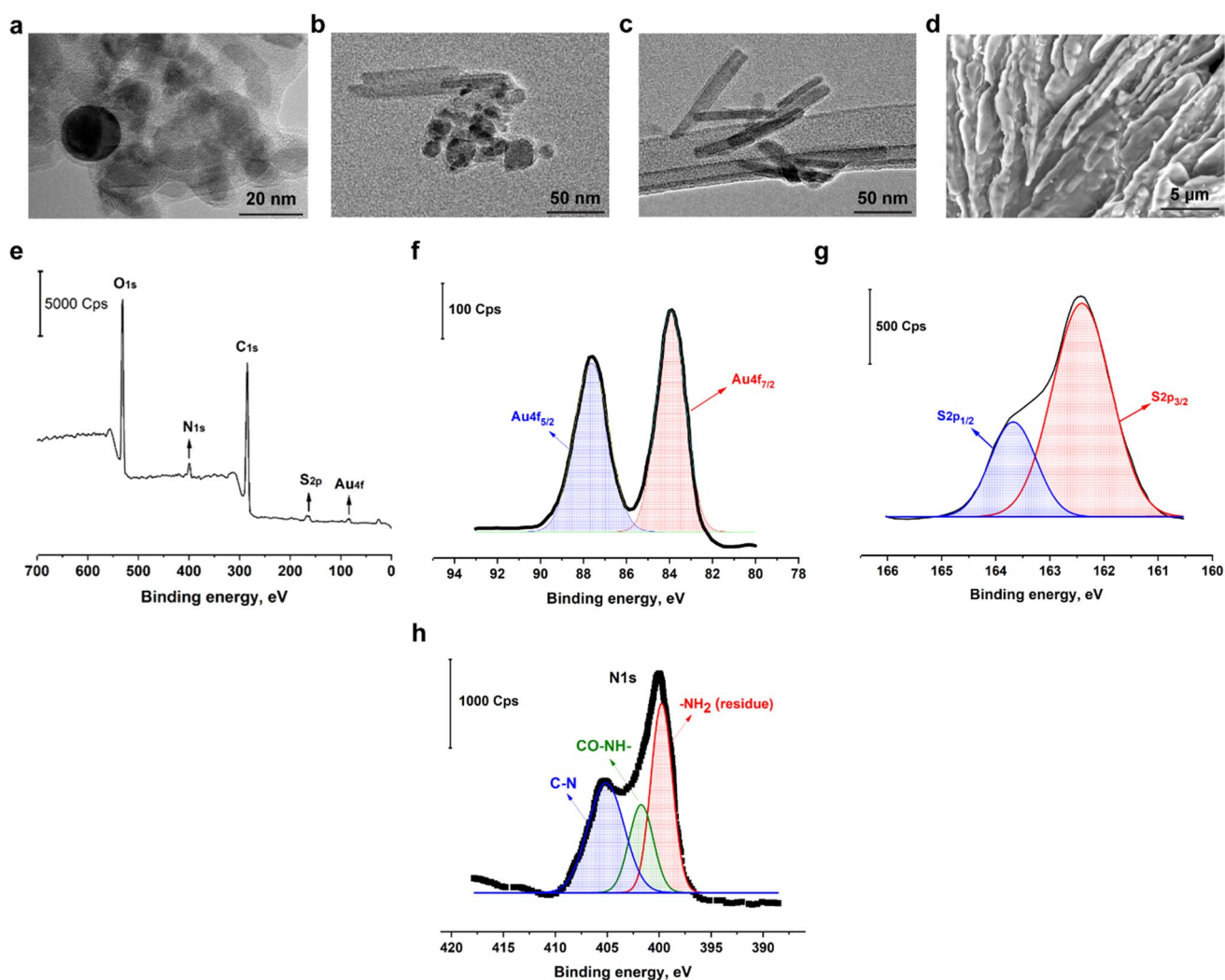
for 5-HT, precision and accuracy tests were conducted, both intraday and interday (over 5 consecutive days), to assess the reliability of the method. Additionally, an interference study was performed to evaluate the influence of potential substances found in blood serum, including Na<sup>+</sup>, K<sup>+</sup>, Mg<sup>2+</sup>, Fe<sup>2+</sup>, ascorbic acid, uric acid, and dopamine. Finally, real blood serum samples with varying concentrations of 5-HT (1.0, 3.0, and 5.0 μM) were analyzed using the developed method to validate its applicability in real sample analysis.

## Results and discussion

The preparation steps of the AuNRds-ATP-CTP nanocomposite were characterized with TEM, SEM, and XPS as shown in Fig. 2. The stages of the synthesis process from nano seeds 10–20 nm Fig. 2a to nanorods 50 nm Fig. 2b can be seen where the length of the final nanorods, is around 100 nm, with a diameter of 20–30 nm (Fig. 2c).

During the synthesis of nanorods, the sizes of the nanorods were determined through image analysis using Gwyddion® software based on TEM and SEM images. A minimum of 20 samples were examined, and the measurements were presented as the average value accompanied by the standard deviation ( $\sigma$ ). Initially, the formation of seed particles with dimensions of  $8 \pm 4$  nm was observed (Fig. 2a). As the crystallization of the nanomaterial progressed in the later stages of the synthesis (Fig. 2b), fully developed nanorods were observed in conjunction with the seed particles, exhibiting dimensions of  $26 \pm 24$  nm. Following the completion of the synthesis, the nanorods had an average length of  $91 \pm 17$  nm and a diameter of  $12 \pm 2$  nm (Fig. 2c).

Scanning electron microscopy (SEM) is commonly employed for the morphological characterization of nanomaterials with multilayer coatings (Saleh et al. 2022). SEM analysis of the AuNRds-ATP-CTP structure (Fig. 2d) revealed the formation of partially agglomerated but well-defined rod-like carbonaceous structures, exhibiting lengths of  $7 \pm 2.3$  μm and diameters of  $1 \pm 0.3$  μm. A notable observation was the close resemblance between the average aspect ratios of the nanorods ( $12:91 = 0.13$ ) and the carbonaceous structures ( $1:7 = 0.14$ ). This indicates that the carbon structures grew on the nanorods in a manner consistent with the underlying material, providing evidence of their presence. While limited literature comparisons may exist for the SEM imaging of this specific structure due to its novelty in synthesis, the morphology bears a resemblance to fiber structures coated with graphene oxide or similar carbonaceous materials (Cui and Zhou 2018; Raagulan et al. 2018). The additional carbon content contributes to the organic appearance of the material and the partially non-uniform widths, which can be attributed to the carbon form. Additionally, the



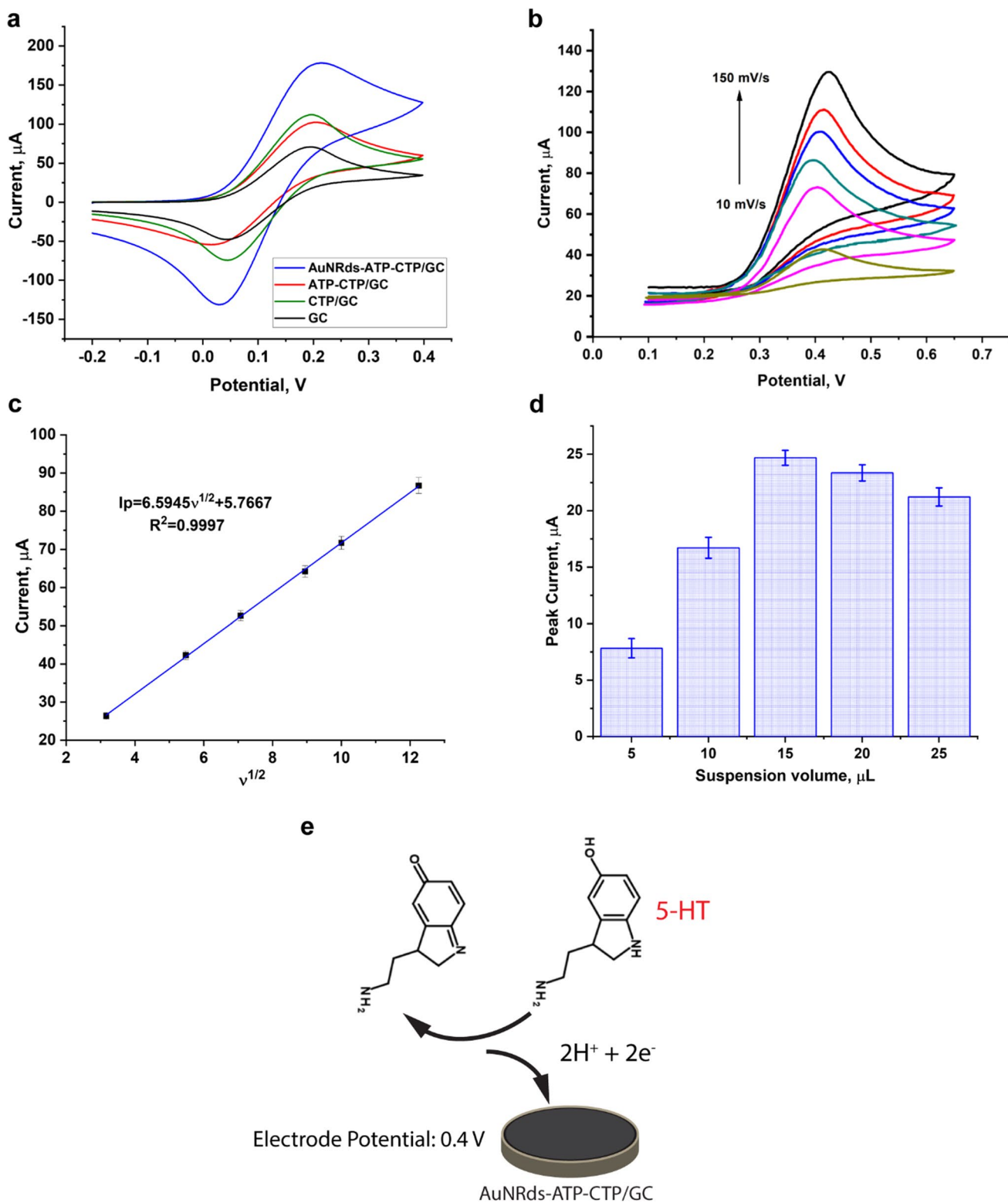
**Fig. 2** TEM images of **a** Au nano seeds, **b** Au nano seed-nanorod hybrid nanoparticles, **c** gold nanorods; SEM image of **d** AuNRs-ATP-CTP, **e** Survey spectrum of AuNRs-ATP-CTP, XPS core spectra of **f** Au4f, **g** S2p, and **h** N1s

energy-dispersive X-ray (EDX) analysis presented in Fig. S1 confirms the presence of Au, S, N, and C elements in the nanocomposite, further supporting the successful synthesis of AuNRs-ATP-CTP.

XPS survey spectrum of AuNRds-ATP-CTP/GC is given in Fig. 2e showing the binding peaks of C1s, N1s, O1s, S2p, and Au4f. XPS core spectra of Au4f, S2p, and N1s of AuNRds-ATP-CTP/GC are also given in Fig. 2. In Fig. 2f, Au4f<sub>5/2</sub> and Au4f<sub>7/2</sub> binding energies at 87.6 eV and 83.9 eV, respectively, indicated that the gold nanorods were bound to the AuNRds-ATP-CTP/GC surface (Kariper et al. 2019). S2p XPS high-resolution narrow region spectra at 163.7 eV and 162.4 eV showed the presence of S2p<sub>1/2</sub> and S2p<sub>3/2</sub> at the surface, respectively (Fig. 2g) (Güzel et al. 2013). Figure 2h shows that CTP oxidized with 4-aminothiophenol (4ATP) is bound by amidation since the N1s peaks at 405.2 eV, 401.8 eV, and 399.8 eV corresponded to –C–N,

–CO–NH, and –NH<sub>2</sub> residues from 4ATP, respectively (Lara et al. 2018).

Figure 3 presents CVs of 1 mM K<sub>3</sub>Fe(CN)<sub>6</sub> on different electrode surfaces: bare GC, CTP–GC, ATP–CTP/GC, and AuNRds–ATP–CTP/GC in 0.1 M KCl electrolyte. The anodic peak currents observed for the redox probe on the respective electrodes were measured as 39.3 μA, 47.6 μA, 59.9 μA, and 91.6 μA. Notably, the AuNRds-ATP-CTP/GC electrode exhibited a higher peak current compared to the other electrodes, suggesting enhanced electron transfer rates for the redox probe on this electrode surface. Furthermore, CVs of 5 μM 5-HT in a pH 7.0 phosphate-buffered saline (PBS) solution at various scan rates are shown in Fig. 3b. These CVs indicate that the anodic oxidation current increases with increasing scan rates. This behavior is further validated by the Randles–Sevcik plot of peak current versus the square root of the scan rate presented in Fig. 3c



**Fig. 3** **a** CVs (200 mV/s, vs Ag/AgCl<sub>sat.</sub>) of 1 mM K<sub>3</sub>Fe(CN)<sub>6</sub> and K<sub>4</sub>Fe(CN)<sub>6</sub> in KCl on bare GC, CTP/GC, ATP-CTP/GC and AuNRbs-ATP-CTP/GC, **b** CVs of 5  $\mu\text{M}$  5-HT at different scan rates (10–150 mV/s, vs Ag/AgCl<sub>sat.</sub>) for AuNRbs-ATP-CTP/GC electrode, **c**

Randles–Sevcik plot produced from **b**, **d** optimization of the suspension volume of AuNRbs-ATP-CTP/GC electrode, **e** detection mechanism of the 5-HT

(Elgrishi et al. 2018). The linear relationship observed in the plot ( $I_p = 6.5945v^{1/2} + 5.7667$ ,  $R^2 = 0.9997$ ) suggests that the determination of 5-HT could potentially be performed quantitatively under diffusion-controlled conditions. To optimize the suspension volume coated on the AuNRds-ATP-CTP/GC electrode, different volumes (ranging from 5 to 25  $\mu\text{L}$ ) of a suspension containing 20 mg of AuNRds-ATP-CTP in 1 mL of MeCN were tested. Figure 3d displays the peak current responses obtained from DPV experiments in the presence of 10  $\mu\text{M}$  5-HT. The highest peak current response was observed for a suspension volume of 10  $\mu\text{L}$ , thus indicating that the calibration study should be conducted using a suspension volume of 10  $\mu\text{L}$ .

DPV of different concentrations (0.5–10  $\mu\text{M}$ ) of 5-HT standards (in PBS, pH=7) tested on AuNRbs-ATP-CTP/GC electrode are given in Fig. 4. An increasing oxidation current response was obtained related to the increased oxidation of 5-HT on the electrode surface (Fig. 4a). The linear relationship between the peak currents obtained from the DPV results at 0.4 V versus Ag/AgCl<sub>sat.</sub>, and the concentration change of 5-HT is given in Fig. 4b. The high reproducibility of the results is reflected in Fig. 4b showing the low standard deviation (RSD values less than 5%) of signals to different concentrations of the sample. A linear response ( $I_p = -0.1204 + 2.5403[5\text{-HT}]$ ) was obtained with sensitivity and LOD of 2.54  $\mu\text{A}/\mu\text{M}$  and 0.03  $\mu\text{M}$ , respectively. The summary of the analytical performance parameters of AuNRbs-ATP-CTP/GC electrode for the detection of 5-HT is given in Table 1.

Table 2 summarizes the analytical performance comparison of the developed sensor with similar studies in the literature for the detection of 5-HT. It can be seen that the developed sensor performance was satisfactory based on the literature data and especially shows fairly better performance than recently published studies using carbonaceous-based electrodes (Dong and Zhang 2022; Winiarski et al. 2022).

The precision and accuracy values of the developed method for intraday and interday (for five consecutive days) were investigated for 1 and 5  $\mu\text{M}$  5HT and summarized in Table S1 ( $N=5$  samples). Precision (RSD %) was calculated

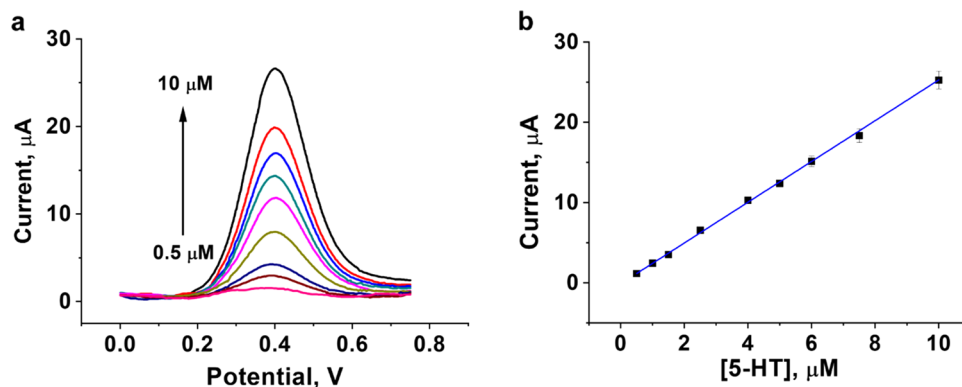
**Table 1** Summary of analytical parameters of the developed method ( $n=5$ )

Analytical parameters	Values
Working dynamic range ( $\mu\text{M}$ )	0.5–10
Regression equation, $I_p$ ( $\mu\text{A}$ ) and [5-HT] ( $\mu\text{M}$ )	$I_p = -0.1204 + 2.5403[5\text{-HT}]$
Standard error of the slope, $\pm$	0.04
Standard error of the intercept, $\pm$	0.02
$R^2$	0.998
Sensitivity ( $\mu\text{A}/\mu\text{M}$ )	2.54
LOD, $\mu\text{M}$ ( $S/N=3$ )	0.03
LOQ, $\mu\text{M}$ ( $S/N=3$ )	0.09

as the coefficient of variation which is the standard deviation divided by the mean measured concentration  $\times 100$  (Garrido et al. 2008). On the other hand, accuracy was calculated as a % difference from the mean value. The accuracy of the developed method for 1 and 5  $\mu\text{M}$  5HT was calculated as 2.08% and  $-0.8\%$  for intraday and  $-3.0\%$  and 2.4% for interday, respectively. The precision of the method for 1 and 5  $\mu\text{M}$  5HT was calculated as 2.94% and 3.63% for intraday and 4.12% and 3.32% for interday, respectively. In general,  $\leq 15\%$  values of RSD % are acceptable for analytical methods in the literature (Karnes and March 1993). The results of precision and accuracy are  $< 5\%$  indicating that the developed sensor provides significantly precise and accurate results when tested in different conditions.

The possible interfering substances that might be present in human serum such as ions ( $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{Mg}^{2+}$ ,  $\text{Fe}^{2+}$ ) and molecules (ascorbic acid, uric acid, and dopamine) were also tested for their effect on the peak current responses of 5  $\mu\text{M}$  5-HT in buffer solution (PBS, pH=7). Table S2 shows that the signal change of the peak current of 5-HT was approximately  $< 3\%$  and  $> -4\%$ . Therefore, it can be concluded that no remarkable interference effects were observed with the tested substances. The validity of the developed method was finally confirmed by spike sample experiments with human serum samples. The spike method was selected due to its widely demonstrated

**Fig. 4** **a** DPVs (pulse width of 0.06 s, amplitude of 0.05 V, sample period of 0.02 s and pulse period of 0.20 s, vs Ag/AgCl<sub>sat.</sub>) of ATP-CTP/GC electrode for different 5-HT concentrations (tested in PBS, pH 7) and **b** linear calibration curve at the working dynamic range



**Table 2** Response of different electrochemical sensors for 5-HT in literature

Method	Electrode materials	Range ( $\mu\text{M}$ )	LOD ( $\mu\text{M}$ )	Refs.
CV/EIS	rGO-Ag <sub>2</sub> Se/GCE	0.1–15	0.030	Selvam and Yun (2020)
Amperometry	Poly(3-amino-5-mercapto-1,2,4-triazole) modified GC electrode	0.01–50	0.013	Revin and John (2012)
SWV	AuNP@Ppy/GSPE	0.1–15	33.22	Tertiř et al. (2017)
SWV	MS/CPE	0.49–13.6	0.1	Winiarski et al. (2022)
SWV	AuNPs/poly-AMT/f-CNT/GCE	0.015–5	0.008	Shekher et al. (2023)
DPV	CSE/PdNPs/C dots/SiO <sub>2</sub>	0.5–7	0.036	Ferreira et al. (2023)
DPV	SPCE/ZnONR/PMBDES/AuNP	0.1–25	1.91	Atici et al. (2023)
DPV	e-CS/MWCNTs/GCE	9–1000	10	Tang et al. (2023)
DPV	BVG/HG:	1–350	0.361	Xue et al. (2023)
DPV	Fe <sub>3</sub> O <sub>4</sub> /MWCNT-p(BCG)/GC	0.5–100	0.0	Ran et al. (2017)
DPV	Pt/MWCN T/PPy/AgNPs	37.5–125	0.15	Cesarino et al. (2014)
DPV	DGNW/SPGE	1–500	0.2	Boonkaew et al. (2022)
DPV	CTAB/NiS/CS/GCE	0.01–200	0.033	Dong and Zhang (2022)
DPV	Ag/Cu <sub>2</sub> O/Ppy/GCE	0.01–250	0.0124	Selvarajan et al. (2018)
DPV	AuNRds-ATP-CTP/GC	0.5–10.0	0.0288	This study

rGO reduced graphene oxide, GC or GCE glassy carbon (electrode), AuNP gold nanoparticles, Ppy polypyrrole, GSPE graphite-based screen-printed electrodes, MWCNT multi-walled carbon nanotube, p(BCG) poly(bromocresol green), DGNW diamond graphene nanowalls, CTAB cetyltrimethyl ammonium bromide, MS mill scale, CSE/PdNPs/C dots/SiO<sub>2</sub> PdNPs/carbon dots/SiO<sub>2</sub> nanoparticle on the printed carbon electrode, SPCE/ZnONR/PMB<sub>DES</sub>/AuNP zinc oxide nanorods/polymethylene blue (deep eutectic solvent)/gold nanoparticles on screen-printed carbon electrodes, e-CS/MWCNTs/GCE electrodeposited chitosan and carboxylated multi-walled carbon nanotubes on glassy carbon electrode, AuNPs/poly-AMT/f-CNT/GCE gold nanoparticle/poly-(amino-5-mercapto-1,3,4-thiadiazole)/functionalized carbon nanotube on glassy carbon electrode, BVG/HG B-doped vertical graphene/horizontal graphene

**Table 3** Recovery values of 5-HT in human serum sample ( $N=5$ )

Spiked ( $\mu\text{M}$ )	Found ( $\mu\text{M}$ )	RSD%	Recovery%
1.0	0.98 $\pm$ 0.03	3.06	98.0
3.0	2.87 $\pm$ 0.06	2.09	95.7
5.0	4.93 $\pm$ 0.08	1.62	98.6

performance in the literature for validation experiments to help identify matrix effects and determine the recovery of the analyte (Thakur et al. 2022). The results showed very promising sensor performance with recovery values between 95 and 99% versus spiked 5-HT concentrations (1.0, 3.0, and 5.0  $\mu\text{M}$ ) as given in Table 3. These recovery values of  $\geq 95\%$  suggest that it's within the acceptable range of between 70 and 120% in the literature (Schoenau 2019). It is reported that the standard levels of human serum 5-HT are between 0.28 and 1.24  $\mu\text{M}$  (Chernecky and Berger 2012), therefore, the developed sensor can be used as a sensitive and accurate measurement tool from human serum samples.

## Conclusion

In conclusion, this study successfully developed a highly sensitive and reliable sensor for the determination of 5-HT using a novel carbonized electrode material. The electrode material, prepared from CTP through a green chemistry approach, was modified with AuNRds using 4ATP as a linker. The nanocomposite material was characterized using microscopy and spectroscopy techniques, confirming the successful attachment of AuNRds to the carbonaceous material.

The modified electrode exhibited excellent electrochemical properties, enabling the accurate detection of 5-HT through DPV. The sensor demonstrated a LOD of 0.03  $\mu\text{M}$  with a linear range of 0.5–10  $\mu\text{M}$ . The sensor showed good reproducibility and stability, as evidenced by low standard deviations and high precision values obtained from intra-day and interday experiments. Furthermore, an interference study revealed minimal effects of potential substances present in human serum, highlighting the selectivity of the sensor for 5-HT detection. Real-sample tests conducted on human serum samples validated the applicability of the developed sensor for practical use. The recovery values obtained from spiked samples demonstrated the accuracy of the sensor, with recovery percentages exceeding 95% for different spiked concentrations of 5-HT.

Overall, this study highlights the potential of carbonaceous electrode materials, derived from waste coal tar pitch, in electroanalytical applications. The combination of gold nanorods and carbonaceous materials resulted in a sensitive and cost-effective sensor for the determination of serotonin. The developed sensor offers a promising tool for the accurate detection of serotonin in biological samples, with potential applications in clinical diagnostics and research.

**Supplementary Information** The online version contains supplementary material available at <https://doi.org/10.1007/s11696-023-03016-7>.

**Acknowledgements** The authors would like to express their gratitude to Prof. M. Oguzhan Caglayan for his valuable scientific support during the preparation of this manuscript.

## References

- Atici T, Kamaç MB, Yilmaz M, Kabaca AY (2023) Zinc oxide nanorod/polymethylene blue (deep eutectic solvent)/gold nanoparticles modified electrode for electrochemical determination of serotonin (5-HT). *Electrochim Acta* 458:142484
- Babaei A, Taheri AR, Aminikhah M (2013) Nanomolar simultaneous determination of levodopa and serotonin at a novel carbon ionic liquid electrode modified with Co(OH)<sub>2</sub> nanoparticles and multi-walled carbon nanotubes. *Electrochim Acta* 90:317–325
- Barnett NW, Hindson BJ, Lewis SW (1998) Determination of 5-hydroxytryptamine (serotonin) and related indoles by flow injection analysis with acidic potassium permanganate chemiluminescence detection. *Anal Chim Acta* 362:131–139
- Boonkaew S, Dettlaff A, Sobaszek M, Bogdanowicz R, Jönsson-Niedziółka M (2022) Electrochemical determination of neurotransmitter serotonin using boron/nitrogen co-doped diamond-graphene nanowall-structured particles. *J Electroanal Chem* 926:116938
- Cesarino I, Galesco HV, Machado SAS (2014) Determination of serotonin on platinum electrode modified with carbon nanotubes/polypyrrole/silver nanoparticles nanohybrid. *Mater Sci Eng C* 40:49–54
- Chernecky, Cynthia C, and Barbara J Berger (2012) Laboratory tests and diagnostic procedures. Elsevier Health Sciences
- Coelho AG, Aguiar FPC, de Jesus DP (2014) A rapid and simple method for determination of 5-hydroxytryptophan in dietary supplements by capillary electrophoresis. *J Braz Chem Soc* 25:783–787
- Cui J, Zhou S (2018) Highly conductive and ultra-durable electronic textiles via covalent immobilization of carbon nanomaterials on cotton fabric. *J Mater Chem C* 6:12273–12282
- Dăscălescu D, Apetrei C (2021) Nanomaterials based electrochemical sensors for serotonin detection: a review. *Chemosensors* 9(1):14
- Dong Y, Zhang L (2022) Coupling surfactants with 3D hollow raspberry-like NiS/carbon microspheres with S vacancies for enhanced sensitivity monitoring of serotonin and L-tryptophan. *Sens Actuators B Chem* 368:132140
- Elgrishi N, Rountree KJ, McCarthy BD, Rountree ES, Eisenhart TT, Dempsey JL (2018) A practical beginner's guide to cyclic voltammetry. *J Chem Educ* 95:197–206
- Erikson H, Sarapu A, Tammeveski K, Solla-Gullón J, Feliu JM (2014) Shape-dependent electrocatalysis: oxygen reduction on carbon-supported gold nanoparticles. *ChemElectroChem* 1:1338–1347
- Erkal A, Aşık İ, Yavuz S, Kariper A, Üstündağ Z (2016) Biosensor application of carbonaceous nanocoil material: preparation, characterization, and determination of dopamine and uric acid in the presence of ascorbic acid. *J Electrochem Soc* 163:H269
- Ferreira JHA, Peres RM, Nakamura M, Toma HE, Canevari TC (2023) PdNPs/carbon dots/silica hybrid nanostructures: the development of an electrochemical sensor for simultaneous determination of dopamine and serotonin in real samples. *J Nanoparticle Res* 25:9
- Garrido EMP, Garrido JM, Esteves M, Santos-Silva A, Marques MPM, Borges F (2008) Voltammetric and DFT studies on viloxazine: analytical application to pharmaceuticals and biological fluids. *Electroanal Int J Devoted Fundam Borges Pract Asp Electroanal* 20:1454–1462
- Gole A, Murphy CJ (2004) Seed-mediated synthesis of gold nanorods: role of the size and nature of the seed. *Chem Mater* 16:3633–3640
- Güzel R, Ekşi H, Üstündağ Z, Solak AO (2013) Synthesis, characterization, and application of silver nanoparticle-thiophenol nanocomposite film on the glassy carbon surface. *Surf Interface Anal* 45:1821–1829
- Han HS, You J-M, Jeong H, Jeon S (2013) Synthesis of graphene oxide grafted poly (lactic acid) with palladium nanoparticles and its application to serotonin sensing. *Appl Surf Sci* 284:438–445
- Hebié S, Boniface Kokoh K, Servat K, Napporn TW (2013) Shape-dependent electrocatalytic activity of free gold nanoparticles toward glucose oxidation. *Gold Bull* 46:311–318
- Imeri L, Mancina M, Bianchi S, Opp MR (1999) 5-Hydroxytryptophan, but not L-tryptophan, alters sleep and brain temperature in rats. *Neuroscience* 95:445–452
- Isbister GK, Bowe SJ, Dawson A, Whyte IM (2004) Relative toxicity of selective serotonin reuptake inhibitors (SSRIs) in overdose. *J Toxicol Clin Toxicol* 42:277–285
- Kariper İA, Çağlayan MO, Üstündağ Z (2019) Heterogeneous Au/Ru hybrid nanoparticle decorated graphene oxide nanosheet catalyst for the catalytic reduction of nitroaromatics. *Res Chem Intermed* 45:801–813
- Karnes HT, March C (1993) Precision, accuracy, and data acceptance criteria in biopharmaceutical analysis. *J Pharm Res* 10:1420–1426
- Lara GG, Andrade GF, Cipreste MF, da Silva WM, Gastelouis PL, Gomes DA, de Miranda MC, de Almeida Macedo WA, Neves MJ, de Sousa EM (2018) Protection of normal cells from irradiation bystander effects by silica-flufenamic acid nanoparticles. *J Mater Sci Mater Med* 29:130
- Lin MT, Tsay HJ, Su WH, Chueh FY (1998) Changes in extracellular serotonin in rat hypothalamus affect thermoregulatory function. *Am J Physiol-Regul Integr Comp Physiol* 274:R1260–R1267
- Mahato K, Purohit B, Bhardwaj K, Jaiswal A, Chandra P (2019) Novel electrochemical biosensor for serotonin detection based on gold nanorattles decorated reduced graphene oxide in biological fluids and in vitro model. *Biosens Bioelectron* 142:111502
- Panholzer TJ, Beyer J, Lichtwald K (1999) Coupled-column liquid chromatographic analysis of catecholamines, serotonin, and metabolites in human urine. *Clin Chem* 45:262–268
- Selvam SP, Yun K (2020) A self-assembled silver chalcogenide electrochemical sensor based on rGO-Ag<sub>2</sub>Se for highly selective detection of serotonin. *Sens Actuators B Chem* 302:127161
- Perry KW, Fuller RW (1992) Effect of fluoxetine on serotonin and dopamine concentration in microdialysis fluid from rat striatum. *Life Sci* 50:1683–1690
- Profumo A, Fagnoni M, Merli D, Quartarone E, Protti S, Dondi D, Albini A (2006) Multiwalled carbon nanotube chemically modified gold electrode for inorganic as speciation and Bi(III) determination. *Anal Chem* 78:4194–4199
- Raagulan K, Braveenth R, Jang HJ, Seon Lee Y, Yang CM, Mi Kim B, Moon JJ, Chai KY (2018) Electromagnetic shielding by MXene-graphene-PVDF composite with hydrophobic, lightweight and flexible graphene coated fabric. *Materials* 11:1803

- Rajarathinam T, Kang M, Hong S, Chang S-C (2023) Nanocomposite-based electrochemical sensors for neurotransmitters detection in neurodegenerative diseases. *Chemosensors* 11:103
- Ran Gu, Chen X, Xia Y (2017) Electrochemical detection of serotonin based on a poly(bromocresol green) film and Fe<sub>3</sub>O<sub>4</sub> nanoparticles in a chitosan matrix. *RSC Adv* 7:1847–1851
- Ressler KJ, Nemeroff CB (2000) Role of serotonergic and noradrenergic systems in the pathophysiology of depression and anxiety disorders. *Depress Anxiety* 12:2–19
- Revin SB, John SA (2012) Electrochemical sensor for neurotransmitters at physiological pH using a heterocyclic conducting polymer modified electrode. *Analyst* 137:209–215
- Rodriguez P, Plana D, Fermin DJ, Koper MTM (2014) New insights into the catalytic activity of gold nanoparticles for CO oxidation in electrochemical media. *J Catal* 311:182–189
- Saleh TA, Tuzen M, Sari A, Altunay N (2022) Factorial design, physical studies and rapid arsenic adsorption using newly prepared polymer modified perlite adsorbent. *Chem Eng Res* 183:181–191
- Samie HA, Arvand M (2019) RuO<sub>2</sub> nanowires on electrospun CeO<sub>2</sub>-Au nanofibers/functionalized carbon nanotubes/graphite oxide nanocomposite modified screen-printed carbon electrode for simultaneous determination of serotonin, dopamine and ascorbic acid. *J Alloy Compd* 782:824–836
- Schoenau EA (2019) Elements of method design. In: *Current challenges and advancements in residue analytical methods*. ACS Publications
- Selvarajan S, Suganthi A, Rajarajan M (2018) A novel highly selective and sensitive detection of serotonin based on Ag/polypyrrole/Cu<sub>2</sub>O nanocomposite modified glassy carbon electrode. *Ultrason Sonochem* 44:319–330
- Shekher K, Sampath K, Vandini S, Satyanarayana M, Gobi KV (2023) Gold nanoparticle assimilated polymer layer on carbon nanotube matrices for sensitive detection of serotonin in presence of dopamine in-vitro. *Inorg Chim Acta* 549:121399
- Song H, Liu Y, Fang Y, Zhang D (2021) Carbon-based electrochemical sensors for in vivo and in vitro neurotransmitter detection. *Crit Rev Anal Chem* 1–20
- Tang S, Liang A, Liu M, Wang W, Zhang F, Luo A (2023) A glassy carbon electrode modified with a composite consisting of electrodeposited chitosan and carboxylated multi-walled carbon nanotubes for simultaneous voltammetric determination of dopamine, serotonin and melatonin. *Carbon Lett* 1–11
- Tertiş M, Cernat A, Lacatiş D, Florea A, Bogdan D, Suciuc M, Săndulescu R, Cristea C (2017) Highly selective electrochemical detection of serotonin on polypyrrole and gold nanoparticles-based 3D architecture. *Electrochem Commun* 75:43–47
- Thakur D, Dubey NP, Singh R (2022) A review on spike and recovery method in analytical method development and validation. *Crit Rev Anal Chem* 1–19
- Thanh TD, Balamurugan J, Van Hien H, Kim NH, Lee JH (2017) A novel sensitive sensor for serotonin based on high-quality of AuAg nanoalloy encapsulated graphene electrocatalyst. *Biosens Bioelectron* 96:186–193
- Torfs SC, Maes AA, Delesalle CJ, Deprez P, Croubels SM (2012) Comparative analysis of serotonin in equine plasma with liquid chromatography–tandem mass spectrometry and enzyme-linked immunosorbent assay. *J Vet Diagn Invest* 24:1035–1042
- Üstündağ İ, Erkal A (2017) Determination of dopamine in the presence of ascorbic acid on digitonin-doped coal tar pitch carbonaceous electrode. *Sens Mater* 29:85–94
- Winiarski JP, Tavares BF, de Fátima K, Ulbrich CE, de Campos M, Souza AAU, Guelli SMA, Souza U, Jost CL (2022) Development of a multianalyte electrochemical sensor for depression biomarkers based on a waste of the steel industry for a sustainable and one-step electrode modification. *Microchem J* 175:107141
- Xue C, Wang Xi, Zhu W, Han Q, Zhu C, Hong J, Zhou X, Jiang H (2014) Electrochemical serotonin sensing interface based on double-layered membrane of reduced graphene oxide/polyaniline nanocomposites and molecularly imprinted polymers embedded with gold nanoparticles. *Sens Actuators B Chem* 196:57–63
- Xue Y, Li H, Wang X, Xuan X, Li M (2023) Preparation of self-supporting vertically/horizontally grown graphene microelectrodes for neurotransmitter determination. *Anal Chim Acta* 1269:341414

**Publisher's Note** Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Springer Nature or its licensor (e.g. a society or other partner) holds exclusive rights to this article under a publishing agreement with the author(s) or other rightsholder(s); author self-archiving of the accepted manuscript version of this article is solely governed by the terms of such publishing agreement and applicable law.