Electrochemical detection of mephedrone using a graphene screen-printed electrode: A sensitive and selective new approach to identify synthetic cathinones in forensic analysis

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Fig S1. DPVs of $5.0 \times 10-4$ mol L-1 MEP in 0.1 mol L-1 BR buffer solution with different pH values (pH 2.0 in black, pH 4.0 in red, pH 6.0 in blue, pH 7.0 in magenta, pH 8.0 in olive, pH10.0 in navy and pH 12.0 in purple) on SPE-GP obtained by (A) cathodic and (B) anodic-direction scans. 90 mV amplitude, 10 mV step potential, 50 ms modulation time, 0.5 s time interval.



Fig S2. Plots of (A) E_{ρ} vs. pHs and (B) I_{ρ} vs. pH obtained from data of Fig. S2 for the reduction (blue dots) and oxidation (red dots).

An oxidation reaction mechanism in SPE- Gr^1 and a reduction reaction mechanism in dropping mercury electrode (DME)² have been reported for MEP electrochemical processes, which are demonstrated in simplified form in Scheme S1.



Scheme S1. Proposed redox mechanism for MEP in SPE-Gr and dropping mercury electrode.

As can be seen in Scheme S1, the oxidation of MEP generates the product called normephedrone (I), the demethylated analogue of MEP and one of its main phase metabolites. This product is formed by oxidation of the secondary amine and subsequent hydrolysis of the imine intermediate. The electrochemical reduction of MEP at DME was investigated by Krishnaiah et. al.² The proposed reduction mechanism indicated a breakage of the double bond C=O of the ketone, and the formation of a hydroxyl, whose reduction product is the 2-(methylamino)-1-(4-methylphenyl)propan-1-ol (II), as shown in Scheme S1.



Fig S3. CVs in 0.1 mol L⁻¹ BR buffer solution (red dashed line) and borate buffer solution (black dashed line), both at pH 10.0 before and after addition of 5×10^{-4} mol L⁻¹ MEP (solid lines) on SPE-GP. All potential scans started at 0.0 V in the cathodic direction, see arrow. Scan rate of 50 mV s⁻¹.



Fig S4. (**A**) CVs of 5×10^{-4} mol L⁻¹ MEP in 0.1 mol L⁻¹ BR buffer solution, pH 10.0 on SPE-GP. All potential scans started at -1.1 V in the cathodic direction, see arrow. Scan rate (v) of 10 mV s⁻¹ to 400 mV s⁻¹. Linear regressions of (**B**) cathodic peak current (I_{pc}) vs. v, (**C**) I_{pc} vs. $v^{1/2}$, (**D**) logarithm I_{pc} vs. logarithm v.



Fig S5. (**A**) CVs of 5×10^{-4} mol L⁻¹ MEP in 0.1 mol L⁻¹ BR buffer solution, pH 10.0 on SPE-GP. All potential scans started at +0.4 V in the anodic direction, see arrow. Scan rate (v) of 10 mV s⁻¹ to 800 mV s⁻¹. Linear regressions of (**B**) anodic peak current (I_{pa}) vs. v, (**C**) I_{pa} vs. $v^{1/2}$. (**D**) logarithm I_{pa} vs. logarithm v.

Estimation of real surface area of the SPE-GP

The adsorption of MEP on SPE-GP was characterized by applying the Langmuir isothermal to the data in Fig. 5A, according to Equation (01) with a procedure similar to that described for SPE based on carbon nanotubes³:

$$\theta = \frac{K \times [C]}{1 + K \times [C]} \tag{01}$$

where θ is the fractional occupancy of the adsorption sites, K is the equilibrium constant for adsorption, and [C] is the concentration of MEP. The Langmuir plot is shown in Fig. S6, where a linear regression of $1 / Q (\mu C^{-1}) = -3.6 \times 10^{-3} (\pm 0.8 \times 10^{-3}) + 2.6 (\pm 0.1) 1 / [MEP] (\mu mol^{-1} L)$ was obtained with R² of 0.992, indicating that the adsorption of MEP can be well-described by the Langmuir isotherm. From these equations, it was possible to calculate the maximum charge transfer (Q_{max}) at 280.11 μ C, as well as the equilibrium constant adsorption (K) at 0.0014 μ mol L⁻¹.





The rectangular box model represented in Scheme S2 was used to estimate the area of possible molecular orientations of MEP adsorption on the working electrode surface of the SPE-GP, which is based on carbon graphene. Lateral lengths were estimated by trigonometry for bond lengths, bond angles, and Van der Waals radii of terminal atoms, using ChemSketch software.

Scheme S2 shows possible adsorption orientations for MEP on the graphene surface, which are demonstrated on a flat view (A), edgewise view (B), and endwise view (C).



Scheme S2. Rectangular box model of MEP molecule for both (A) flat view, (B) edgewise view, and (C) endwise view.

From these orientations, it was possible to calculate the areas of the three sides of the box as 6.557×10^{-15} , 4.349×10^{-15} , and 6.017×10^{-15} cm² that correspond to 1.525×10^{14} , 2.2996×10^{14} and 1.662×10^{14} molecules per cm² for flat, edgewise and endwise views, respectively. Considering the electron fundamental charge of 1.602×10^{-19} C, the number of electrons involved in redox process of MEP, which was estimated to be 2, and the Q_{max} found (280.11 µC), the number of molecules adsorbed on the SPE-GP was calculated to be 8.74×10^{14} . Thereby, assuming one monolayer and close packing, the real surface area available for adsorption of the graphene layer of the SPE-GP was estimated to be between 3.80 and 5.70 cm², which values are very close to those reported for the active surface area of other electrodes modified with graphene-oxide⁴. Indeed, the estimated area for this working electrode is larger (at least 19-fold) than its calculated electroactive area (0.2 cm²), corroborating with the high sensitivity presented by SPE-GP for MEP detection.



Fig S7. AdSDPVs in 0.1 mol L⁻¹ BR buffer solution pH 10.0 (black lines) before and after addition of a real seized sample (red lines) without and with standard solution of MEP at 1.0 (blue lines) and 2.0 (magenta lines) μ mol L⁻¹ using anodic (**A**) and cathodic (**B**) scans, on SPE-GP. Experimental conditions are the same as in Fig. 4. Insets are the obtained recovery values.



Fig S8. Typical raw data from AdSDPV measurements (without smoothing and baseline correction) in 0.1 mol L⁻¹ BR buffer solution pH 10.0 (black lines) before and after addition of 5×10^{-4} mol L⁻¹ MEP (red lines) on SPE-GP, using anodic (**A**) and cathodic (**B**) scans. Experimental conditions are the same as in Fig. 4.

REFERENCES

- 1 J. Schram, M. Parrilla, N. Sleegers, F. van Durme, J. van den Berg, A. L. N. van Nuijs and K. de Wael, *Drug Test Anal*, 2021, **13**, 1282–1294. DOI: https://doi.org/10.1002/dta.3018
- 2 V. Krishnaiah, Y. V. R. Reddy, V. H. Reddy, M. T. Reddy and G. M. Rao, *IJSR International Journal of Scientific Research*, 2012, **Volume 1 Issue 4**, 14-17.
- 3 W. T. P. dos Santos and R. G. Compton, *Sens Actuators B Chem*, 2019, **285**, 137–144. DOI: https://doi.org/10.1016/j.snb.2019.01.047
- 4 N. T. A. Thu, H. Van Duc, N. H. Phong, N. D. Cuong, N. T. V. Hoan and D. Q. Khieu, *J Nanomater*, 2018, Article ID 7619419. DOI: https://doi.org/10.1155/2018/7619419.