Brain tumor mutated DNA detection with 2- and 3-terminal graphene devices

Pedro Alpuim[1,2], Telma Domingues[1,2], Laís C. Lopes[3], Jérôme Borme[1], Paulo R. Bueno[3], Bruno Costa[4]

[1] International Iberian Nanotechnology Laboratory (INL), Av. Mestre José Veiga, Braga, Portugal
[2] Centro de Física das Universidades do Minho e Porto (CF-UM-UP), Universidade do Minho, Braga, Portugal
[3] Instituto de Química, Universidade Estadual Paulista (UNESP), Araraquara, Brazil
[4] Instituto de Ciências da Vida e da Saúde (ICVS), Escola de Medicina, Universidade do Minho, Braga, Portugal
pedro.alpuim.us@inl.intl

Brain tumors are detected by expensive and invasive techniques, effective only when the disease is already in advanced stages. Liquid biopsies have recently emerged as a promising method for early cancer detection and represent a minimally invasive way to capture tumor activity in real time. In the case of malignant glioma, particular biomarkers with clinical relevance were identified, e.g., the TERT promoter mutation at position C228T.[1] Mutations are detected by polymerase chain reaction (PCR) and next-generation sequencing (NGS), either for whole genome sequencing or massive sequencing of the target region. The detection process is long, complicated, and costly. Therefore, a high-sensitivity and high-specificity detection method is needed to detect ultra-low concentrations of circulating tumoral DNA early and fast. In this work, a 25-mer sequence containing the C228T mutation (tDNA) is detected using a complementary strand, immobilized on graphene 25 µm x 75 µm channels. The channels – 20 per chip – are contacted with Au source and drain terminals for electrical transducing of the biorecognition events. A large, coplanar Au electrode surrounds the channel region.[2] The 10 µL droplet containing the mutated DNA diluted in a buffer or human plasma is placed over the 20 functionalized graphene channels. The detection is achieved by operating each transducing device as a 3-terminal liquid-gate transistor (GFET) or a 2-terminal electrochemical impedance electrode (GEE). In the latter case, the chip's source and drain output pads are short-circuited, and the planar gate works as the counter-electrode. In transistor mode, the chips detected the tDNA down to the attomolar range (~ 10 aM) by following the charge neutrality point voltage in the GFET transfer curves. In the GEE mode, the signal was obtained from impedance-derived capacitance spectroscopy by following the minimum of the graphene quantum capacitance as a function of bias potential at a specific low-frequency limit,[3] with a limit-of-detection of 1 aM. These results encourage the pursuit of an integrated, low-cost, ultra-sensitive biosensor to detect circulating tumoral DNA based on single-layer graphene microdevices.

References

[1] Bell, R. J. A., Rube, H. T. et al., Molecular Cancer Research, 14(4) (2016) 315–323
[2] Purwidyantri, A., Azinheiro, S. et al., ACS Sensors 8(2) (2023) 640–654
[3] Lopes, L.C., Santos, A., Bueno, P., Carbon 184 (2021) 821-827

Figures

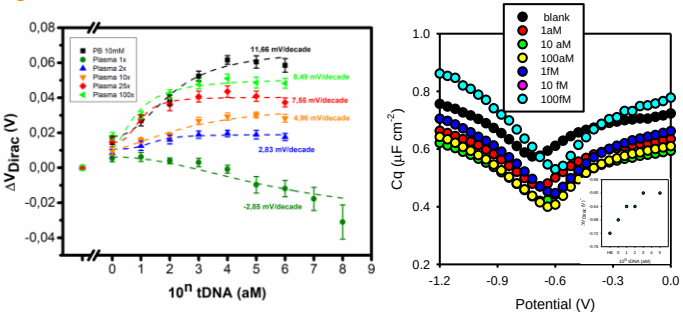

Figure 1: a) Calibration curves for GFET detection of tDNA in PB and plasma. b) Graphene quantum capacitance measurements for different tDNA concentrations in human plasma. The inset shows the Dirac point movement as a function of tDNA concentration.