Synthesis of smooth graphene surfaces by CVD for electrochemical biosensors with supported lipid membranes

M. Pittori^{1,2*}, L. Ortolani², D. Gentili³, V. Morandi², R. Rizzoli², and M. G. Santonicola¹

In this work we perform an *ab initio* study on the design of a novel electrochemical biosensor, in which graphene and membrane proteins would serve as transducer and biological recognition elements, respectively. Graphene is used as transducer because of its unique and intriguing properties, namely surface area, electrical conductivity, ultra high electron mobility, wide electrochemical potential window, low charge-transfer resistance, and reduction of overvoltage. All these properties are responsible for the enhancement of the direct electron transfer between the graphene surface and the membrane proteins [1-2]. Membrane proteins are the chosen biosensing element for this study since they represent almost 60% of all human protein drug targets. The main problem is that the contact with electrode surface causes the denaturation of membrane proteins, so they need to be embedded in a system mimicking their native environment (i.e. the cell membrane). Supported lipid bilayers (SLBs) are widely used as artificial cell membranes for biophysical studies and nanobiotechnology applications. They are most often generated starting from a liposome solution in which surfaces are incubated for a certain period of time [3-4]. SLBs form preferentially and are functional on more hydrophilic substrates, whereas graphene surfaces are highly hydrophobic, and so they need to be modified [5].

The first part of this study focused on the synthesis of graphene through catalyzed chemical vapor deposition (C-CVD), on its characterization prior to surface treatments, by micro-Raman spectroscopy, water contact angle (WCA) measurements, and on the investigation of the morphology of graphene using a scanning electron microscopy (SEM). As a result, large domains of mono- to few-layer graphene were synthesized, with a high reproducibility of the process. The second part of the study focused on the surface treatments of graphene to improve its compatibility towards SLBs through a mild oxidation, and WCAs were measured again after surface treatments to evaluate the variations of the wettability of graphene.

The interaction of the modified graphene surfaces with DOPC (1,2-dioleoyl-sn-glicero-3-phosphocholine) lipids and in particular the formation of a SLBs due to liposomes spreading was investigated via electrochemical impedance spectroscopy (EIS) using a custom-designed electrochemical cell. The EIS measurements evidenced that the CVD-graphene surface was a suitable conductive interface in the physiological environment of the electrochemical cell, as it maintained its properties constant over repeated measurements. An increase of resistance at low frequency indicated the formation of an insulating layer after the incubation of the surface with liposome solution, in agreement with the assembly of a lipid membrane onto the conductive graphene surface.

References

- [1] M. Pumera, Chemical Society Reviews, 39 (2010), 4146.
- [2] D.A.C. Brownson et al., The Handbook of Graphene Electrochemistry, Springer, 2014.
- [3] E.T. Castellana et al., Surface Science Reports, 61 (2006), 429.
- [4] G.W. de Groot et al., Nanoscale, 6 (2014), 2228.
- [5] R.P. Richter et al., Langmuir, 22 (2006), 3497.

^{1*} Department of Chemical Materials and Environmental Engineering, Sapienza University of Rome, Rome, Italy ^{2*} Institute for Microelectronics and Microsystems (IMM), Section of Bologna, National Council of Research (CNR), Bologna, Italy

³ Institute for Nanostructured Materials (ISMN), Section of Bologna, National Council of Research (CNR), Bologna, Italy



Figure 1: (a) SEM image of transferred graphene; Raman spectra (c) and corresponding Raman map of the ratio between the 2D and G peak intensities (b).



Figure 2: Bode (left) and Nyquist (right) plots showing the comparison of the CVD graphene surfaces before and after incubation of 4 hours with DOPC liposomes.