Interfacing (reduced) Graphene Oxide to neurons: an electrophysiological study

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The research for new repairing and reconstructing strategies, acting like functional bridges to damaged tissues and bioactive scaffolds for axon regrowth, calls for the development of novel functional materials capable of structurally and functionally interfacing with Central Nervous System at the cellular and molecular level. As the organization of biological sub-cellular structures is in the nanometer scale, researchers identified in nanomaterials the most promising candidates for such applications [1]. Particularly, neuroscientists addressed their interest to Carbon-based nanostructured materials, as their peerless optical, electronic and mechanical properties make them suitable materials for electrodics and neuroprosthetics [2,3]. Graphene has recently found several applications in Neurosciences, in particular as three-dimensional scaffolds for neural stem cells [4] and as transparent neural electrode arrays for electrophysiological recordings [5,6].

In this work we compare for the first time, at the best of our knowledge, the use of Graphene Oxide (GO) and reduced Graphene Oxide (rGO) as substrates for primary cortical neurons culturing.
GO substrates were prepared by spin-coating GO solution on glass coverslips, while rGO substrates were obtained by electrochemical reduction of GO ones. These substrates were then characterized by Scanning Electron Microscopy (SEM), Atomic Force Microscopy (AFM) and Raman Spectroscopy.
We firstly evaluated biocompatibility and cytotoxicity of these substrates, by means of cell viability and Lactate Dehydrogenase (LDH) assays, and then we also quantitatively assessed the contingent effects these substrates exert on neuronal electrophysiological properties, both at single-cell and network-level, by means of patch-clamp recordings.

Our results show that the electrical phenotype of individual neurons, as well as the formation of fully developed and active networks, is not affected by both GO and rGO. Furthermore, we found that neurons grown on GO display modest, yet significant, differences in the spontaneous rate of Action Potential firing, which resulted to be greater when compared to both control glass coverslips and rGO-coated substrates.
These results, being consistent with previous reports employing stem cells and immortalized cell lines, confirm that GO and rGO are suitable candidates for neuronal interfacing [7].

References

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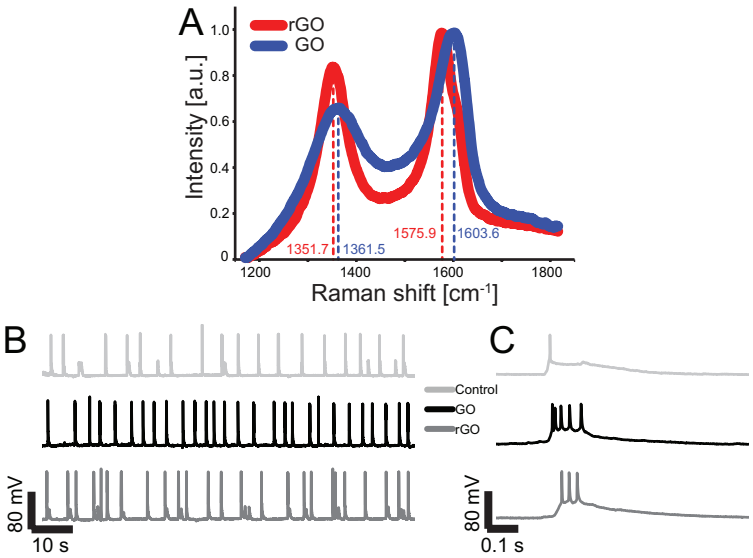

Figure 1: Raman spectra of GO and rGO (A) show the typical D and G peaks. Representative raw intracellular voltage recordings (B-C) display a small, but significant, increase of the rate of spontaneous occurrence of Action Potentials and bursts.