## THE GRAPHENE OXIDE SPECIES INDUCE A DIFFERENT BIOLOGICAL RESPONSE IN SN4741 PARKINSON CELL LINE.

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<u>Text: Introduction:</u> Graphene Oxide (GO)has recently emerged as a reliable material to create scaffolds for the neural tissue because of its biocompatibility, electroconductive and physicochemical properties. Graphene is a 2-dimensional material consisting of rings of carbon atoms with an excellent electrical conductivity originating in the sp2 hybridized carbons network. Nevertheless, there is not a consensus which kind of graphene oxide is most useful of benefit. <u>Aim:</u> In this study we evaluate the capacity of GO and its derivatives, partially reduced graphene oxide (PRGO) and fully reduced graphene oxide (FRGO), to support differentiation and maturation of a dopaminergic cell line-SN4741. Cell viability and cytotoxicity doseresponse assays showed good survival. However, the phenotype and genotype of the cells were differentially regulated by the diverse forms of graphene.

Materials: GO, PRGO, FRGO material were prepared by Abalonyx AS (Oslo, Norway) from natural graphite powder following Hummers and Offeman 1958. SN4741 cells were exposure until 2 weeks. The analysis of the morphological changes of SN4741 cells growing in normal conditions as described by Son et al., 1999. LIVE/DEAD® Viability/Cytotoxicity Assay Kit (Invitrogen) was used for biocompatibility assay. Immunostaining assay: Primary antibodies were used: rabbit anti-tyrosine hydroxylase (TH); mouse anti-Tuj-1 (beta-III tubulin 1); RNA extraction and RT-PCR to cDNA obtained was carried out using RNase Kit (Quiagen). The following primer sequences were used: Pitx3, Lmx1a and Lmx1b and Tuj1.

Results: Our results (normalized expression) in comparison with different species shown a significant increase in the expression levels of Tuj1 and transcription factors specific for DA neurons such as Pitx3, Lmx1a, and Lmx1b in PRGO species.

Conclusion: Our study shows a different biological behavior related to phenotype and genotype in GO species in which PRGO species seem to be the best choice for Parkinson disease studies



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