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Presentation Abstract

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Presentation Title:	Positive biocompatibility of several graphene derivatives with dopaminergic cells at long term culture
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Topic:	++A.03.b. Neural differentiation of pluripotent stem cells
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Abstract:	The emerging carbon nanomaterial graphene (G) and its oxidized derivative graphene oxide (GO) have recently gained considerable attention in biomedical applications such as cancer therapy or biosensors. It has for example been demonstrated that G has an efficient bioconjugation with common biomolecules and activates cell differentiation of neuronal stem cells (Li et al., 2013). This way, G could acts as a physical support or scaffold to promote differentiation and axonal sprouting of dopaminergic (DA) cells derived from neural stem cells. Since GO in its multilayer form and with multiple carboxylate and epoxy groups seems to show interesting biological properties (Yang et al., 2013) the aim of the present work has been to test different graphene derivatives searching for the best scaffold to be used in stem cell differentiation. For this purpose we have tested the cytotoxicity of GO and reduced GO, and specifically its biocompatibility with SN4741, a dopaminergic cell line derived from mouse substance nigra, measuring the effect on long term culture. The cells were cultured in Dulbecco's modified Eagle's medium 10% FCS (Gibco) to about 80% confluence. Cells (1.000) were plated onto 96-well microliter plates with graphene using three chemically different types of GO as powders and films: 1) hydrophilic GO; 2) partially reduced GO (PRGO) which is hydrophobic and 3) fully reduced GO (FRGO), also hydrophobic, each of them in five concentrations: 1 mg/ml; 0.1 mg/ml; 0.05 mg/ml; 0.02 mg/ml and 0.01 mg/ml. Cells were cultured with GO and cell viability was determined after 24 hours, 1 week and 2 weeks using the MTT assay (Roche) and

cytotoxicity was determined by the lactate dehydrogenase (LDH) assay (Roche). Our results show positive biocompatibility between the G-derivatives and SN4741 cells. We conclude that the use of our G-derivative scaffolds can enhance the morphological differentiation towards DA neurons (TH positive) providing microenvironments appropriate for neural differentiation and axon guidance. These findings suggest that biocompatible scaffolds can contribute to the future generation of successful clinical applications of G. Future experiments will examine whether G could offer a platform for neural stem cell and neural regeneration for neurological diseases such as PD. (Refs: Li N., Zhang Q, Gao S. et al., 2013, Nature/Sci Rep. 3:1604. doi: 10.1038/srep01604; Yan K., Li Y., Tan X., et al., 2013, Small., 9(9-10): 1492-1503) This work has been supported by the University of Malaga, Campus de Excelencia Internacional Andalucía Tech, Spain; The Norwegian Research Council (grant nº 215086) and funds from Karolinska Institutet, Stockholm, Sweden.

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