GRAPHENE OXIDE MODIFIED WITH COLLAGEN I FOR MYOCARDIAL TISSUE REGENERATION

JOANNA JAGIELLO[1]*, MARCIN GWIAZDA[1,2]*, EWA KIJENSKA-GAWRONSKA[2], MAGDALENA BARAN[1], WOJCIECH SWIESZKOWSKI[2], LUDWIKA LIPINSKA[1]

[1] SBL-INSTITUTE OF ELECTRONIC MATERIALS TECHNOLOGY, POLAND
[2] FACULTY OF MATERIAL SCIENCE AND ENGINEERING, WARSAW UNIVERSITY OF TECHNOLOGY, POLAND
*E-MAIL: JOANNA.JAGIELLO@ITME.EDU.PL
ST.M.GWIAZDA@GMAIL.COM

[ENGINEERING OF BIOMATERIALS 153 (2019) 30]

Introduction

Nowadays a lot of scientific effort is focused on the development of new materials for the biomedical use (e.g. for tissue engineering [1] and cancer treatment). One of the most promising material in this field is graphene derivative called graphene oxide (GO). GO is a defected graphene (carbon layer with one atom thickness arranged in hexagonal crystal lattice), where defects are the result of reactive oxygen functional groups bonded to the surface. Great interest of GO is due to its physicochemical properties which allow for modifications of GO structure by different biomolecules attachment. This extend possibilities for interaction with different types of cells and tissues. Moreover, GO without any surface modification was found to be nontoxic and biocompatible towards different cell lines, even human mesenchymal stem cells. [2]

High-molecular weight proteins e.g. collagen are often introduced into biomaterials to improve cell attachment and proliferation. However, bonding such molecules of desired amount with material poses many difficulties. Collagen I fibrils (Col I) is a major compound of Extracellular Matrix (ECM). Furthermore, functionalization of GO with Collagen I contributes in to the mechanism of the cell differentiation from the Mesenchymal Stem Cells (MSC) to the Cardiomyocytes. [3]

The aim of this study was to obtain GO layers modified with Collagen I with the proper distribution of this peptide fibrils on the surface of GO flakes. Here we present several ways to prepare such composite with different GO coverage and morphology of Collagen I.

Materials and Methods

GO was obtained by modified Hummers method. GO flakes with the concentration of 1 mg/ml were deposited on the glass surface (10 μg/cm^2).

In the first experiment Col I dissolved in acetic acid was mixed with N-(3-dimethylaminopropyl)-N' ethylcarbodiimide) (EDC) and N-Hydroxysuccinimide (NHS), put on GO surface and left overnight to react. In the second experiment firstly functionalization of the graphene oxide by the solution consisting of EDC and NHS was conducted. After that Col I dissolved in acetic acid was put on such modified GO surface with EDC and NHS residues and left overnight. In the third experiment collagen I dissolved in acetic acid was put on modified GO surface (without EDC and NHS residues) and after 24 hours EDC and HNS solution was dropped on thin Col I layer (not dried) bonded to GO surface. It was left overnight again to react. Applied concentration of Collagen type I was 1 pg/ml, EDC and NHS were 20 mg/ml and 15 mg/ml, respectively.

Results and Discussion

In case of each type of biofunctionalizations successful bonding between the GO flakes and Col I fibrils was noticed. That was confirmed by FTIR measurements where characteristic amide groups (C=O stretching (Amide I), N–H bending (Amide II) and C–N stretching (Amide III) are present. Moreover, the clusters of the Collagen I were observed with the use of SEM. The presence of Collagen I clusters was noticed for each type of applied modifications.

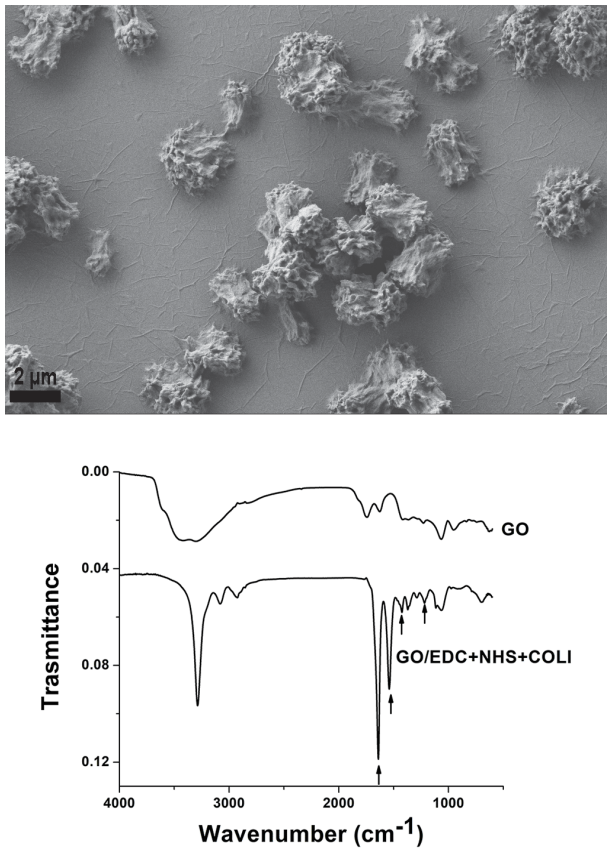

FIG. 1. SEM image and FTIR spectra of the GO modified with Col I mixed with EDC and NHS.

Conclusions

According to the obtained data, the optimal distribution of the Collagen I was received for the third experiment, where suspension of Collagen I was deposited on the modified graphene oxide and crosslinked by the addition of EDC/NHS solution at the last step. That method allowed to receive the homogenous distribution of Collagen I on the surface of graphene oxide flakes. Additionally, the presence of the empty spaces, where Collagen I was not bounded can allow for better interaction between the cells and graphene oxide flakes. Moreover, it has potential application for the additional immobilization of other molecules such as growth factors or drugs (drug delivery system).

Acknowledgments

This work was financed by National Science Centre NCN Poland, "Optimisation of biocompatible scaffolds combining graphene and defined stem cell" UMO-2015/16/W/NZ4/00071

References

[1] Dozois, M. et al., Carbon, 120 (2017) 338-349
[2] Duran, M. et al., Curr Mol Med., 17 (2017) 619-626
[3] Kang, P. L., et al., Journal of Biomedical Materials Research Part A, 101 (2013), 2808-281