

Electronic Supplementary Information

Tailoring of Physical and Mechanical Property of Biocompatible Graphene Oxide/Gelatin Composite Nanolaminates via Altering of Crystal Structure and Morphology

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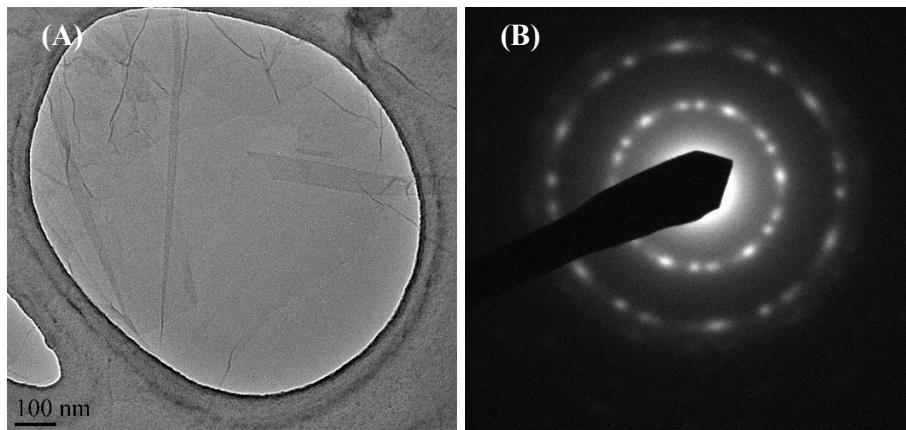
Biocompatibility of Gelatin Graphene Oxide Films

A 15 mm round punch tool was used to cut the GO/GP composite film to make the samples fit into 24-well cell culture plate wells. The resulting circular pieces were cut in half with a scalpel. The samples were ultrasonicated in 99.5% ethanol (Altia Etax Aa, Finland) twice for two minutes and rinsed with a liberal amount of ethanol in between and after. After drying in a laminar flow cabinet, the samples were aseptically packaged into heat-sealed plastic pouches sterilized with ethanol.

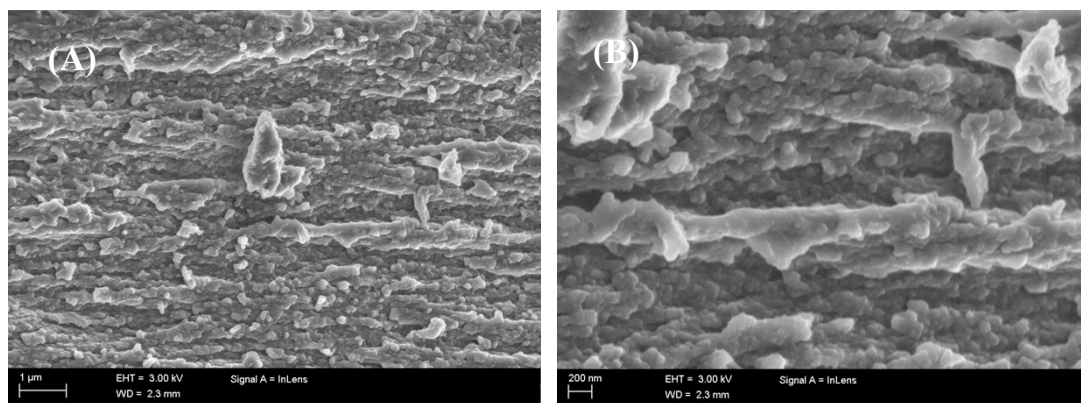
The commercial WI-38 human lung fibroblasts (Culture Collections, Public Health England, UK) were cultured in Dulbecco's Modified Eagle Medium (Gibco DMEM/F-12, USA), supplemented with 10% Fetal Bovine Serum (South American Origin, Biosera, Finland) and 0.5% Penicillin/Streptomycin 100 U/mL (Thermo Fisher Scientific, USA) and incubated at 37^o in 5% CO₂. Tryple Select (Gibco, USA) was used in passaging the cells. 3 parallel samples were used for each sample type. Native cell culture plate surface was used as a positive control. Before plating, the samples and the control wells were prewetted with cell culture medium for 2 h. The medium was subsequently removed. The cells were preplated in 100 µl of cell culture medium at concentration of 407 000 cells/mL on each sample 30 min before adding 400 µl of cell culture medium to each well. The medium was changed every 3 days.

To evaluate cytotoxicity of the films, after 4 and 7 days of cell culture a Live/Dead Viability/Cytotoxicity Kit for mammalian cells (Invitrogen, USA) was used. For staining, the medium was removed from the wells followed by washing with 500 µl of Phosphate Buffer Solution (PBS) (Corning, USA). Each sample was incubated at room temperature for 30 min

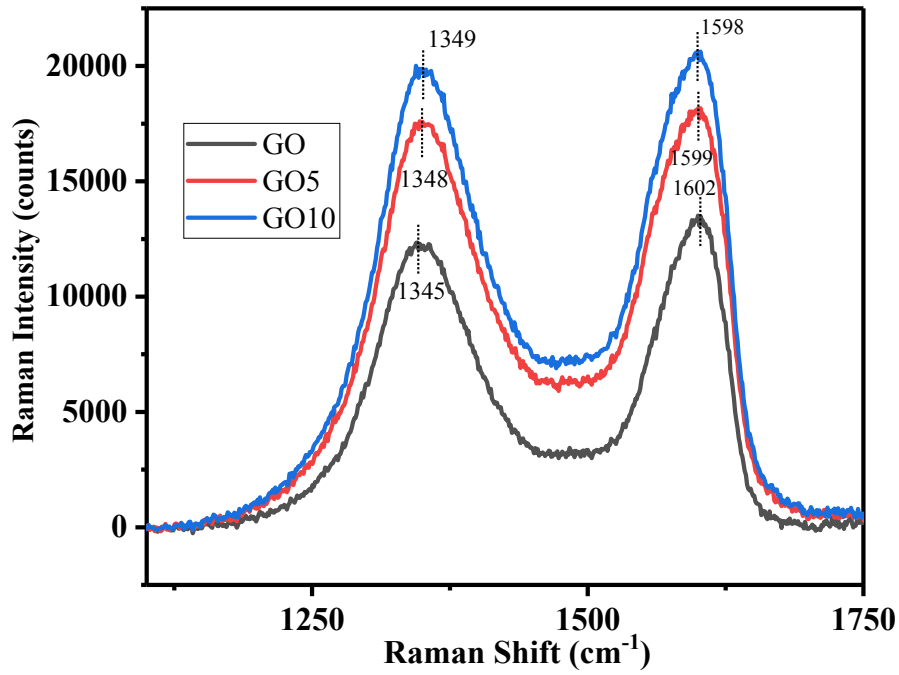
on a rocker with 500 μl of PBS staining solution containing 0.1 μM fluorescent calcein-AM to stain live cells green and 0.5 μM fluorescent ethidium homodimer-1 to stain dead cells red. After incubation, the staining solution was replaced with 500 μl of PBS. Fluorescence microscope (Olympus IX-51, Japan) and a digital camera (Olympus DP30BW, Japan) was used for viability imaging and the images were processed using ImageJ software (National Institute of Health, USA). Merged micrographs showing both live and dead cells were visually inspected for number of dead cells and cell morphology.



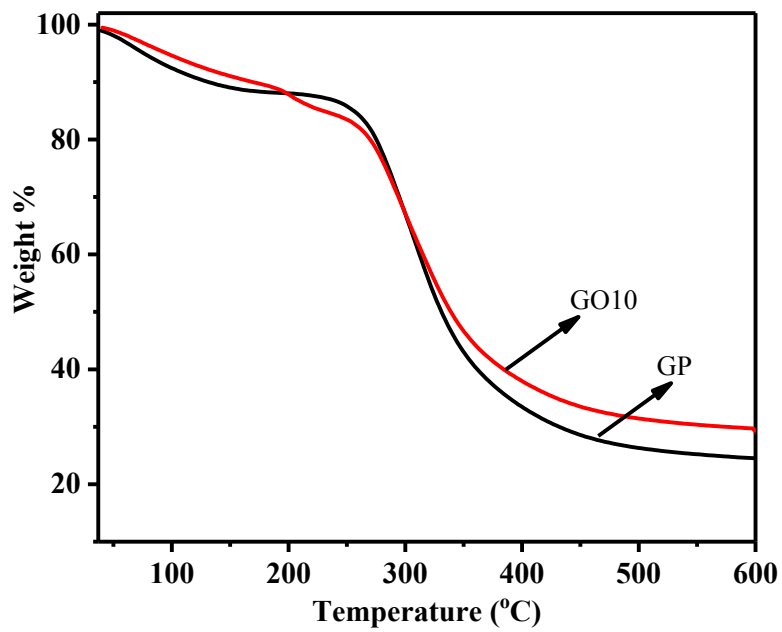
ESI Fig. 1. (A) TEM image and (B) SAED pattern of GO



ESI Fig. 2. (A) FESEM image and (B) enlarge FESEM image of pure GP film



ESI Fig. 3. Raman spectra of GO and GO/GP composite film



ESI Fig. 4. TGA thermogram of GP and GO 10