Favorable biological responses of neural cells and tissue interacting with graphene oxide microfibers

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Supplementary figures

(at.)

.4

6.2

45

6.0

C/N (at.)

95.0

-

10.7

15.2

	-									
Sample	%C	% O	%N	Csp ²	Csp ³	C-OH	0-C-0	0=C-0	Trans. π	C/O (a
Pristine GO	68.4	28.5	0.7	47	-	43	-	9	1	2.4
rGO Fiber 220 °C 15 min	75.2	17.2	0.5	57	11	13	11	-	8	4.4

22

17

20

19

22

12

12

11

59

32

45

Table S1. XPS characterization of GO-based samples.

72.0

78.2

13.4 0.2

6.7

5.1

15.8

13.1

rGO Fiber 220 °C 2 h (A) 82.4

A+PLL

A+N-cadherin

	rGO Fiber
-	1/
	silicone

Figure S1. Photograph of three individual rGO microfibers glued to a glass coverslip through their edges by using medical grade silicon and ready for cell culture.



Figure S2. XPS C1s spectra of pristine GO sheets and rGO microfibers prepared at 220 °C for

15 min. BE: Binding energy (eV).



Figure S3. Sequence of images illustrating controlled deformation of a representative rGO

microfiber prepared at 220 °C for 2h on a stereotaxic device.



Figure S4. Representative immunofluorescence images of ENPC cultures on an uncoated rGO microfiber and correspondent peripheral glass areas at 14 days. Neurons were identified by map2 staining (red), glial cells by vimentin (green) and cell nuclei with DAPI (blue). Scale bar: 150 μm.



Figure S5. Effect of the biological coatings (PLL and N-cadherin) on the physic-chemical properties of rGO microfibers prepared at 220 °C for 2 h. (A,B) Roughness profiles by AFM. In the plots, Y-axes represent height (nm) and X-axes represent distance (µm). (C,D) C1s spectra by XPS. BE: Binding energy (eV).



Figure S6. Characterization of the homogeneity of the biological coating of the microfibers with poly-L-lysine (PLL, middle) and N-cadherin (N-Cad, bottom). An image of an uncoated microfiber has been included for comparison (top).



Figure S7. Representative CLSM micrographs of ENPC cultured on a PLL-coated microfiber and its corresponding peripheral glass area after specific staining of neuronal axons (tau, red) and non-neuron cells including glial cells (GFAP, green). A detail of the microfiber is included in the inset. Cells were cultured for 14 days. Scale bars: 200 μ m (panoramic) and 150 μ m (inset).



Figure S8. Representative CLSM images of ENPC cultures on glass coverslips coated with PLL, adsorbed N-cadherin (ADS) and covalently bond N-cadherin (COV) for 21 days. Cells were identified by the presence of map2 (neurons, green) and vimentin (non-neuron cells, red). Cell nuclei were stained in blue (DAPI). Scale bars: 150 µm.



Figure S9. Control immunofluorescence studies of rGO microfibers for CLSM.



Figure S10. Meningeal fibroblasts cultured on rGO microfibers with different biological coatings for 21 days by SEM (top, scale bars: 50 μ m) and CLSM (middle, scale bar: 150 μ m). Peripheral glass coverslip areas are also included as a control substrate (bottom, scale bar: 150 μ m). ADS: adsorbed N-cadherin; COV: covalently bond N-cadherin. Cells in CLSM images were identified by the presence of vimentin (red) and cell nuclei stained in blue (DAPI).



Figure S11. Panoramic views of C5-C7 spinal cord sections at the injury site 10 days after implantation of the rGO microfiber-based scaffold. Representative immunofluorescence images captured by CLSM are shown for the different markers as indicated in the legend. Orientation is indicated by the set of arrows: *C*: caudal, *D*: dorsal, *Ro*: rostral and *V*: ventral. Scale bars: 1 mm.