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A size dependent evaluation of the cytotoxicity and uptake of nanographene oxide

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† Electronic Supplementary Information Available

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Supplementary information

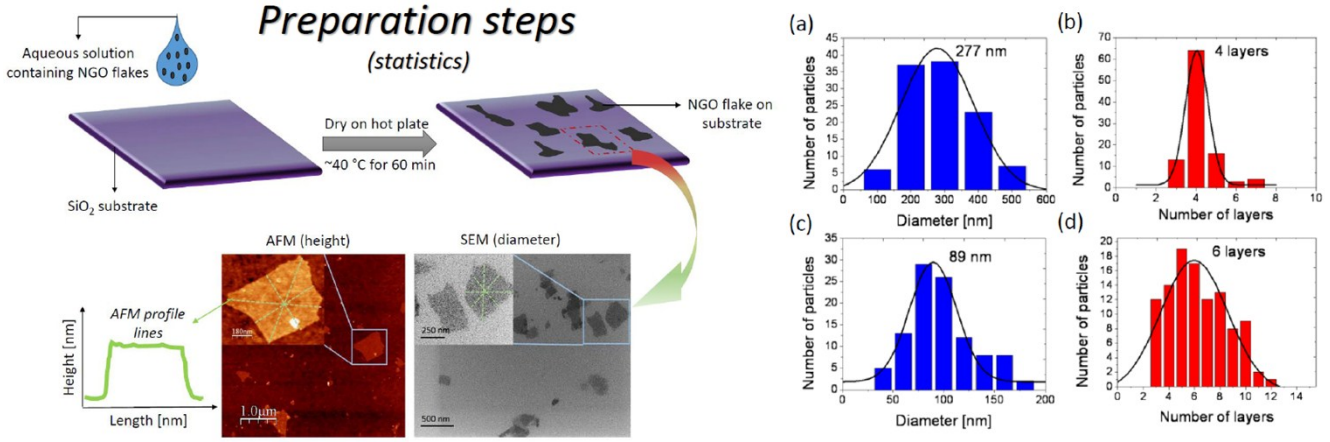

Figure S1. In order to measure the size of the synthesized NGO flakes, a droplet of the aqueous solution containing the NGO flakes was dried on a silicon (Si) substrate. After 60 minutes on a hot plate at 40 °C the substrate was imaged using SEM and AFM. The SEM images were used to obtain the diameter of individual flakes and the AFM images were used to extract the height of the flakes. Both the diameter and height were measured four times for each flake and are then averaged. Panel (a) shows the statistics of the NGO with an average diameter of 277 nm. Panel (b) shows the statistics of height for 277 nm NGO sample and indicates that the NGO has in average 4 layers. Panels (c) shows the statistics of the NGO with 89 nm diameter while in panel (d) shows the statistics of height for 89 nm NGO sample and indicates that the NGO has in average 6 layers.

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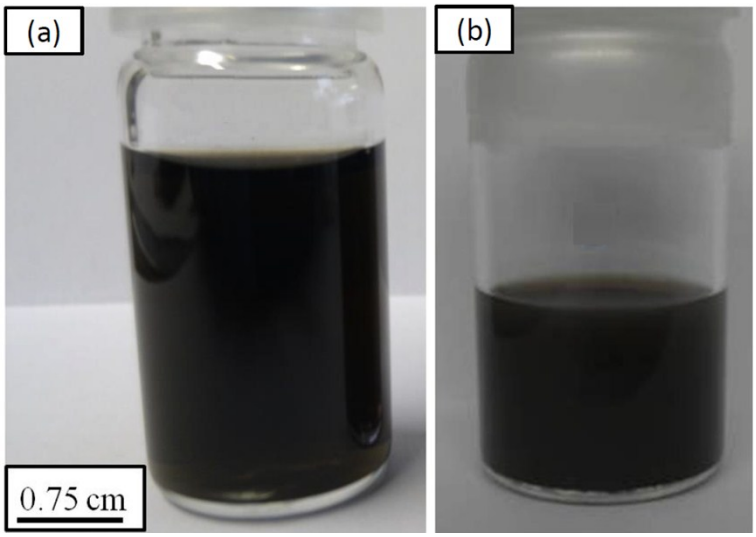

Figure S2. Nanographene oxide samples in aqueous solution. The sample cannot be dried because the layers agglomerates and form graphene oxide buckypaper and cannot be easily dispersed. Panels (a) and (b) correspond to the samples containing nanographene oxide nanoparticles with 277 and 89 nm, respectively.

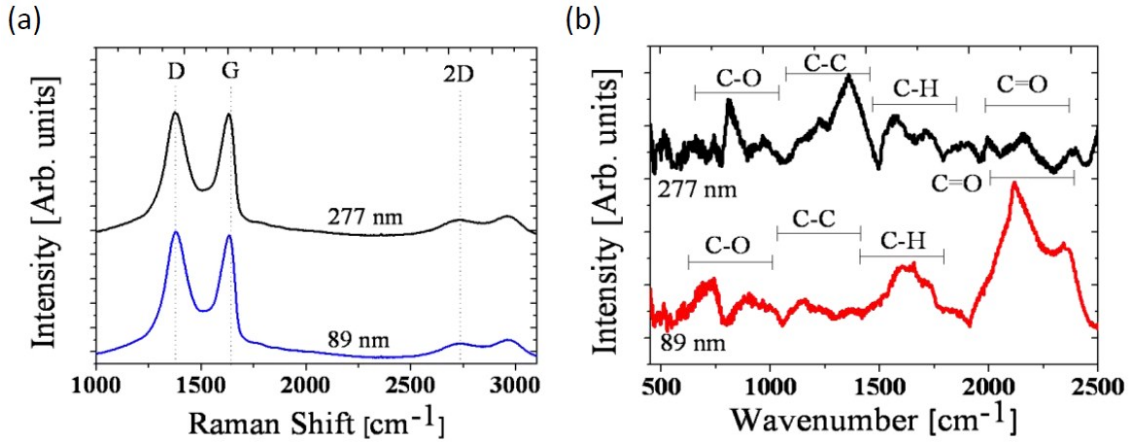

Figure S3. Panel (a) shows the Raman spectra of both NGO sizes. The spectra verify the of D and G modes. The 2D mode can also be identified. Panel (b) shows the IR spectra of both NGO samples and confirms the presence of different functional groups, indicating that the oxidation of the initial graphite material was accomplished.

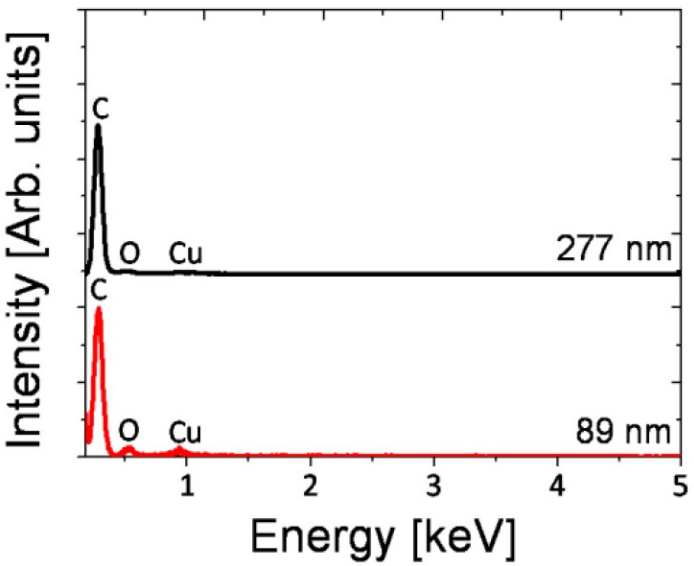

Figure S4. EDX measurements of the two NGO samples conducted with TEM. Both NGO samples contain only carbon (C) and oxygen (O), suggesting no contamination. The copper (Cu) signal is from the copper TEM grid.

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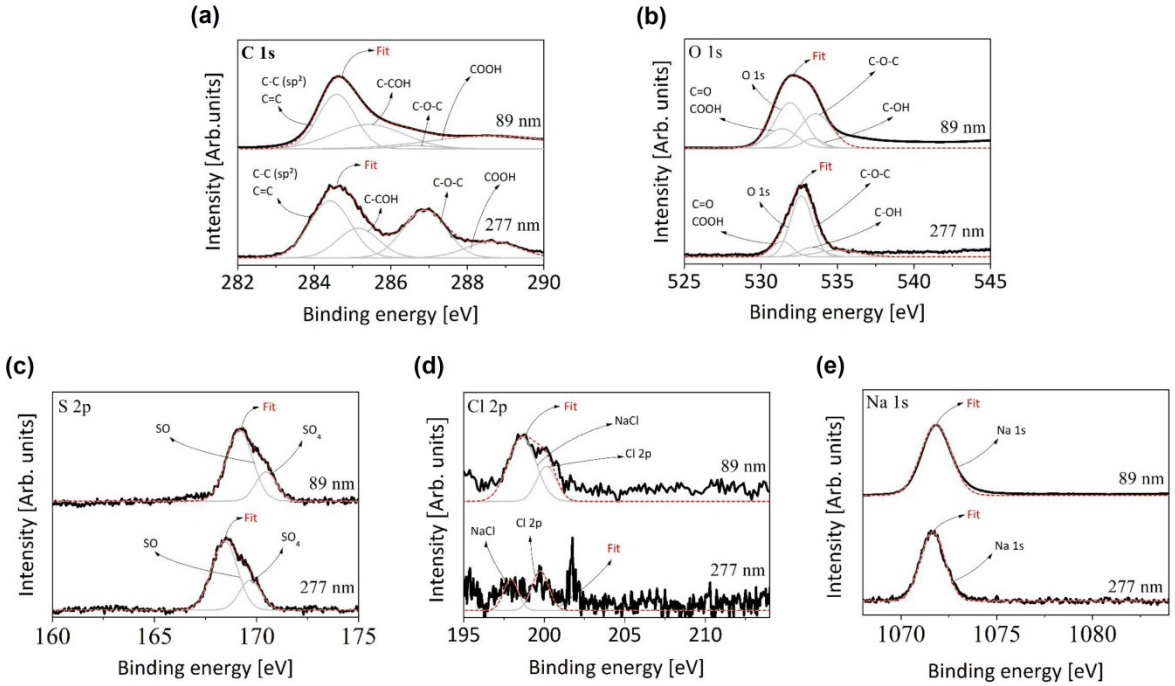

Figure S5. Panels (a) – (e) show the XPS spectra of both NGO samples. Both samples are oxidized and have identical functional groups. Panel (a) shows the C1s XPS spectra of carbon of both samples while in panel (b) the spectra of Oxygen O1s is presented. Panels (c) to (e) reveals traces of sulfur, sodium and chlorine in the NGO samples. The presence of these elements are attributed to the initial synthesis process and cannot be avoided even with intense rinsing.

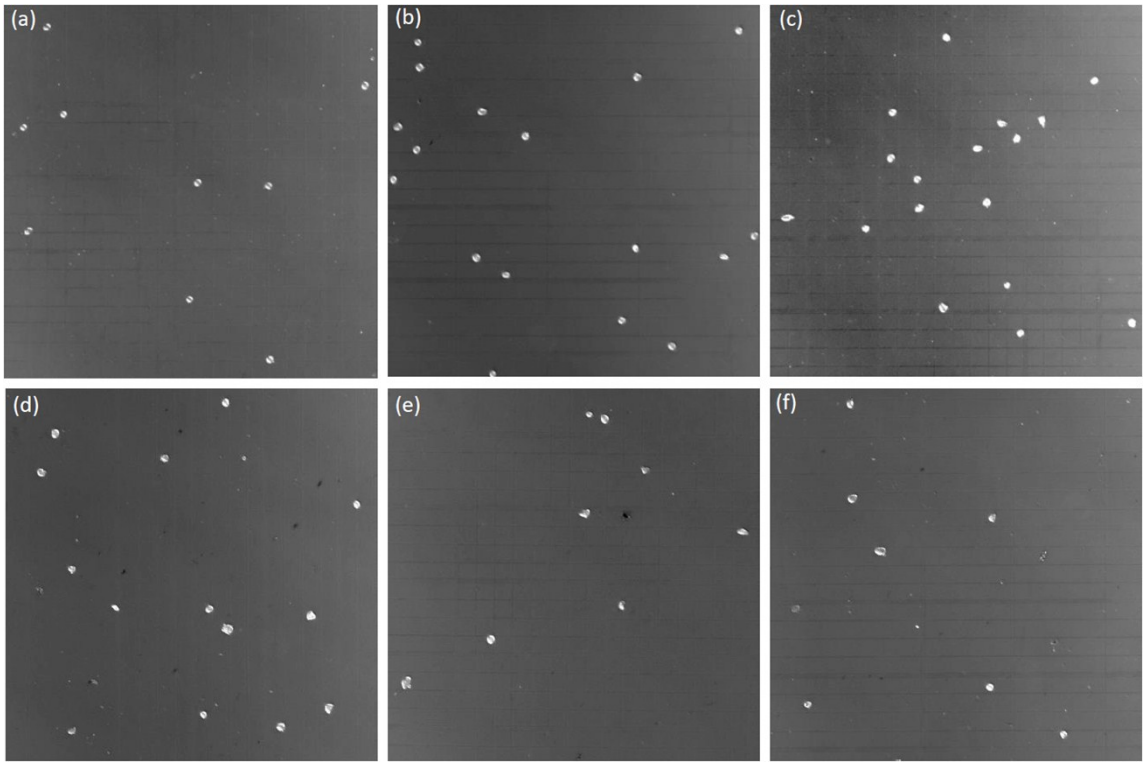

Figure S6. Panels (a) to (f) show representative optical images used to count the number of dead HeLa cells. (a) shows the reference sample of HeLa cells after 12 hours, (b) and (c) show the HeLa cells incubated with the 89 and 277 nm NGO flakes for 12 hours. (d) shows the reference sample of HeLa cells after 48 hours, (e) and (f) show the HeLa cells incubated with the 89 and 277 nm NGO flakes for 48 hours.

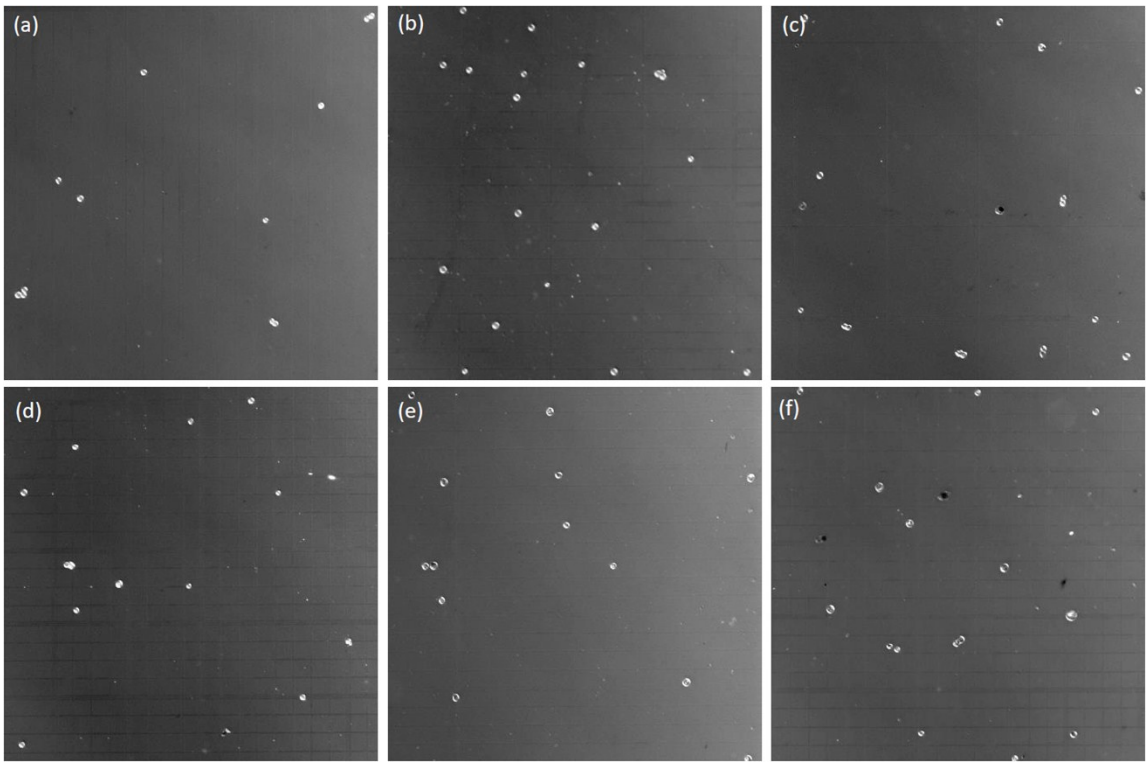

Figure S7. Panels (a) to (f) show representative optical images used to count the number of dead macrophages. (a) shows the reference sample of macrophages after 12 hours, (b) and (c) show the macrophages incubated with the 89 and 277 nm NGO flakes for 12 hours. (d) shows the reference sample of macrophages after 48 hours, (e) and (f) show the macrophages incubated with the 89 and 277 nm NGO flakes for 48 hours.

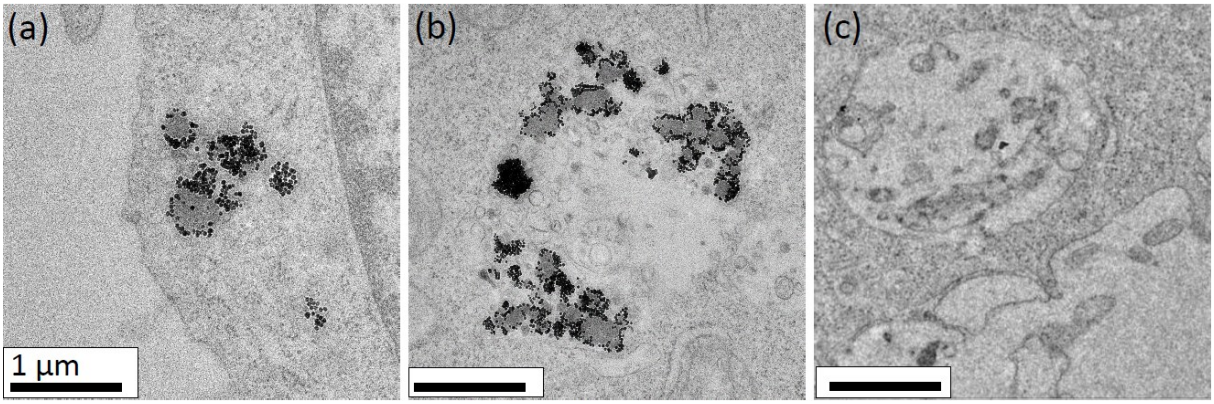

Figure S8. TEM images of iron oxide nanoparticles in a concentration of 10 µg/ml incubated with macrophages for (a) 12 hours, (b) 24 hours and (c) 48 hours. The images show that the macrophages also internalize the particles for all incubation periods. In images (b) and (c) it is possible to see that the nanoparticles are inside vacuoles, however much smaller than the ones observed in the incubation with NGO nanoparticles.