Advanced microscopy of star-shaped gold nanoparticles and their adsorption-uptake by macrophages.

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Supplementary Electronic Information (SEI).

Figure S1. Electron microscopy imaging of gold seeds. **(A)** Bright Field-STEM imaging (FE-SEM HITACHI S5500 with Duo BF/DF-STEM detector, operated at 30 kV). **(B)** High Resolution-TEM imaging of icosahedral gold seed (JEOL 2010-F, operated at 200 kV).

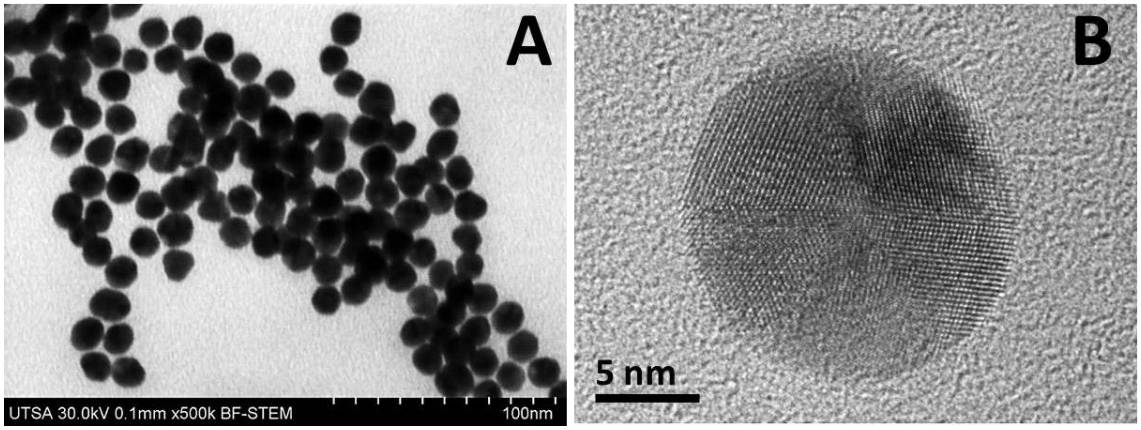

Figure S2. Raman spectroscopy. Surface-enhanced Raman spectroscopy (SERS) of rhodamine 6G (R6G, red line) and GNS-rhodamine 6G (Au-R6G, blue line).

R6G was diluted in 50% ethanol solution at a final concentration of 0.5 mM, GNS stock solution was incubated 12 h at room temperature with equal volume of R6G solution. 20 µl of each sample were mounted onto ultra-flat silicon wafer and dried overnight. Raman spectroscopy (550-1700 cm^{-1}) was performed with HORIBA Jobin Ivon iHR320 imaging spectrometer, operated with a red laser (785 nm, 50 mW).

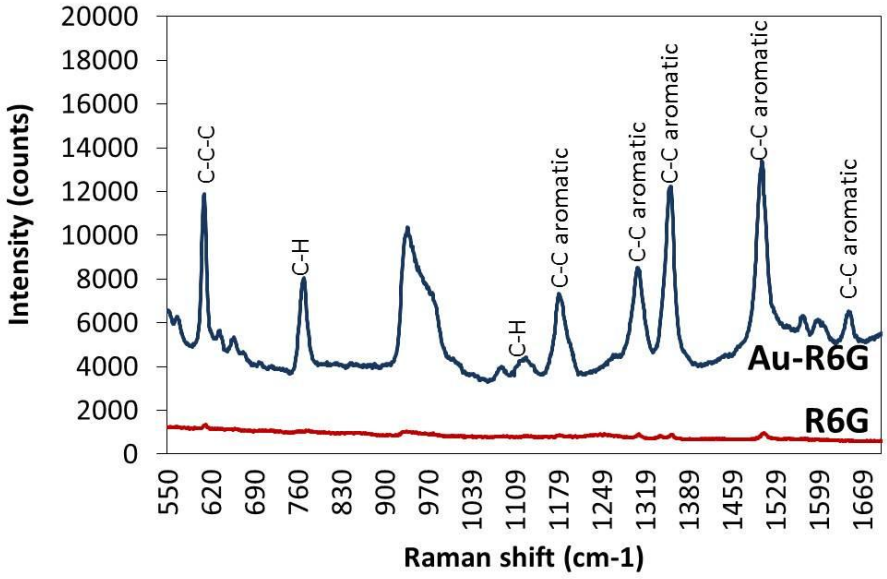

Figure S3. Flow cytometry. **(A)** Histograms obtained by flow cytometry of cells incubated at different concentrations of GNS for 3 or 24 h. **(B)** Cell viability obtained by analysis of histograms statistics, 3 h of incubation (black line), and 24 h of incubation (gray line).

Murine macrophages (J774) were plated at $2x10^5$ in 24-well plates, dosed with GNS (0-350 µl/ml) and incubated for 3 h or 24 h. Cells were washed with sterile 1X PBS, then incubated with 100 µl of propidium iodide (PI) solution at 50 µg/ml for 15 min at room temperature. The samples were then washed in 1X PBS and resuspended in 400 µL 1X PBS by gently scraping. Light scattering analysis by flow cytometry was performed with BD FACSCalibur counting at least 5000 events per sample, and using unstained samples as controls. Acquired data were analyzed with CellQuestPro.

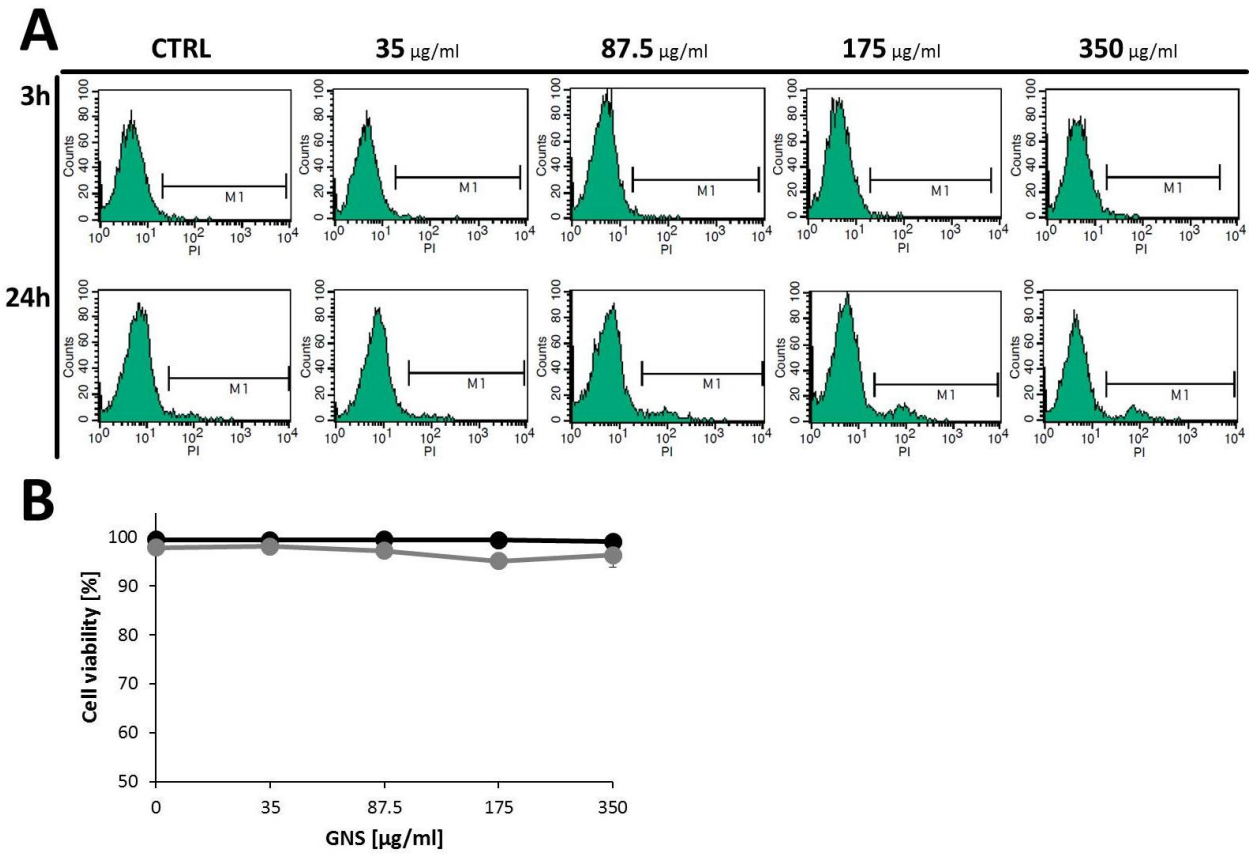

Figure S4. Imaging of GNS after 24 incubation in cell culture media. (A) SE Imaging. **(B)** BF-STEM. **(C)** DF-STEM. **(D)** Size distribution by dynamic light scattering (hydrodynamic diameter of particles centered at 138 nm). Images obtained with FE-SEM HITACHI S-5500, operated at 30 kV.