

Supporting Information

Methods

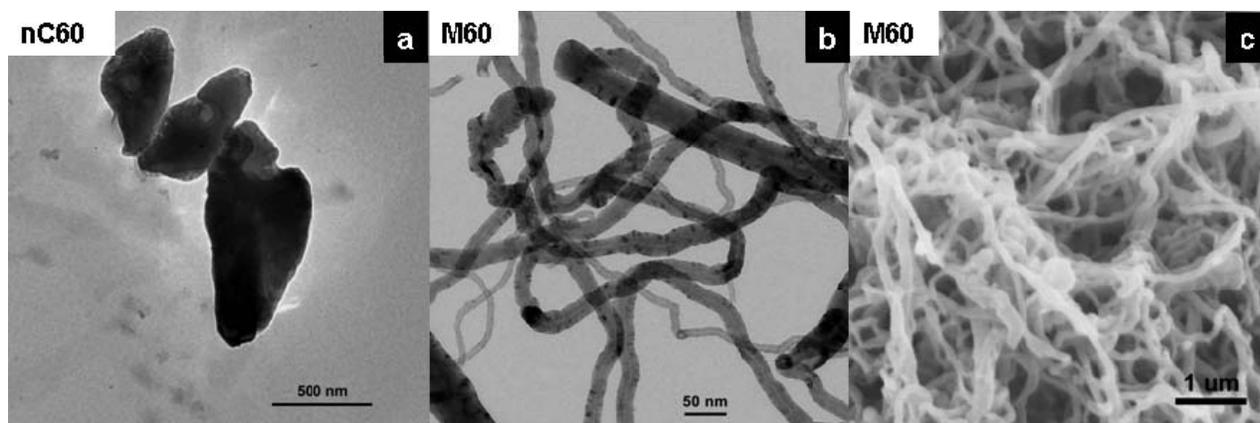
Carbon nanoparticle (CNP) characterization. Solutions of M60 and nC60 in PBS were always freshly prepared. Stock solutions at a concentration of 1 mg/mL were prepared and sonicated prior to use for 1 min at 30 W with and 20 kHz output frequency. Macroscopic aggregates of CNPs readily formed after sonication.

Flow particle image analysis. The size distribution and shape of the nanomaterial agglomerates were investigated by flow particle image analysis of CNP suspensions in plasma using FPIA 3000S (Sysmex, Kobe, Japan, provided by Malvern Instruments, Columbia, MD), as previously published.³ Materials were processed the same way as for platelet experiments. The sonicated suspensions (1 mg/mL) were diluted with platelet-rich plasma to a final CNP concentration of 100 μ g/mL. The Sysmex FPIA-3000S was set up to operate in bright-field mode with the 20x field lenses in high power field (HPF) mode. The size of the particles (the diameter of a circle with the same area as the particle) and circularity (circle circumference/perimeter of the projected particle image) were also evaluated to characterize the shape. Each sample was analyzed in 5 sequential 1-min runs, with at least 4,700 particles in each run. The analysis showed that under our experimental conditions, CNPs formed polydisperse agglomerates whose median size depends on the type of nanomaterial. We also checked for progressive agglomeration in plasma during a period of 30 min. Although nC60 particle size distribution did not show any significant changes, M60 agglomeration progressed slightly while dispersed in plasma.

Zeta potential measurements. The average zeta potential (ζ) of the M60 and nC60 agglomerates (based on at least 10 independent measurements) was determined at 20 °C using a Zetasizer 3000HS instrument (Malvern Instruments, Southborough, MA). This instrument measures the electrophoretic mobility in a quartz capillary using laser Doppler velocimetry. The zeta potential was also measured with a Zeta-Reader instrument (Zeta Potential instruments, Bedminster, NJ), which uses direct imaging (i.e., particle tracking) to measure the electrophoretic mobility.

SI Table 1. Inductively coupled plasma mass spectrometry (ICP-MS) analysis of M60. Elemental analysis of M60 was performed by EAI (Elemental Analysis, Inc). C, H, and N were analyzed by combustion methodology, and Co, Ni and Fe by quadrupole inductively coupled plasma mass spectrometry.

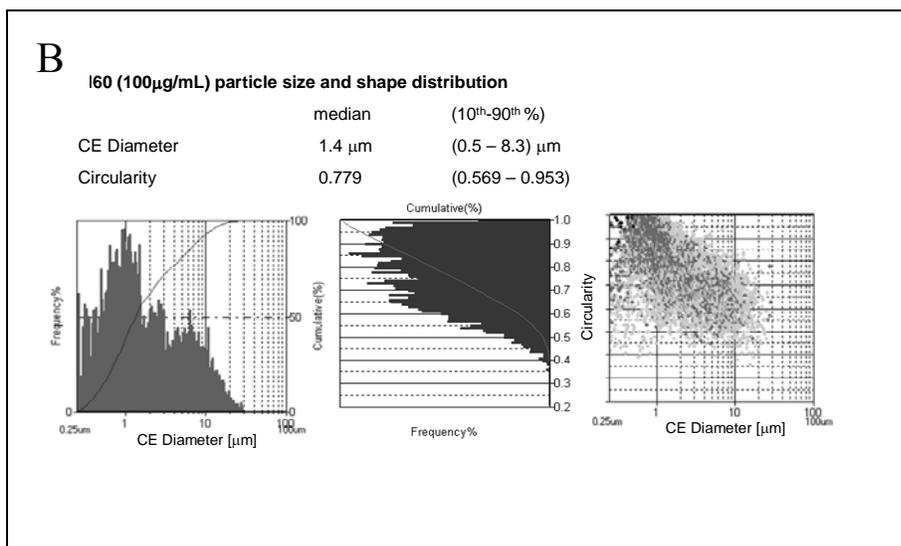
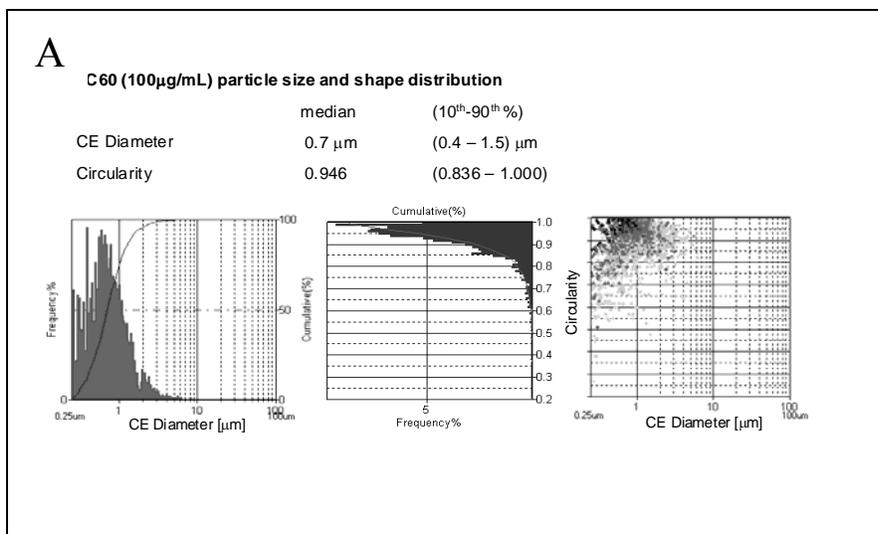
Carbon	95.59 %
Hydrogen	0.22 %
Nitrogen	0.31 %
Cobalt	0.05 %
Nickel	1.13 %
Iron	ND



SI Figure 1. TEM image of pristine nC60 (a), M60 (b) and FESEM of (c) M60 in platelet poor plasma.

SI Table 2. Particle size distribution in suspension of tested nanomaterials in plasma. The size distribution and shape of the nanomaterial agglomerates were investigated by flow particle image analysis of nanomaterial suspensions in blood plasma using FPIA 3000S (Sysmex, Kobe, Japan, provided by Malvern Instruments, Columbia, MD). The particle CE diameter distribution is expressed as median (10th – 90th percentile).

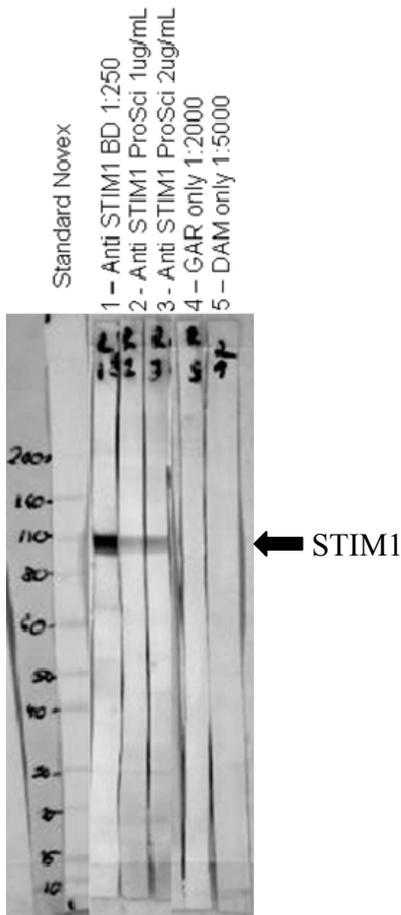
Nanomaterial	Particle CE diameter [μm]	
	Time 0 min	Time 30 min
M60	1.4 (0.5 – 8.3)	1.0 (0.4 – 2.9)
nC60	0.7 (0.4 – 1.5)	0.7 (0.4 – 1.5)



SI Table 3. Particle size and shape distribution in suspension of tested nanomaterials in plasma. The Flow particle image analysis of nC60 (A) and M60 (B) was performed as described in Methods. The analysis was repeated five times with at least 4,700 particles analyzed in each run. Representative histograms of CE diameter and circularity, and dot plots of CE diameter vs. circularity are shown.

SI Table 4. The physical characteristics of nC60 and M60 materials. The zeta potential measurement is described in Methods. Standard uncertainties are reported.

	M60	nC60
Zeta Potential in PBS (mV)	-1.28 ± 0.05	-1.15 ± 0.03
Zeta Potential in Plasma (mV)	-0.72 ± 0.01	-0.56 ± 0.005



SI Figure 2. Western blot analysis of platelet lysate confirms the target specificity of the anti-STIM1 antibody used for immunolabeling. Two different anti-STIM1 antibodies were utilized. Binding of the antibodies was visualized using alkaline phosphatase conjugated goat anti-rabbit (GAR) or donkey anti-mouse (DAM) secondary antibodies and BCIP/NBT substrate. Both STIM1 antibodies recognize a band with the molecular weight corresponding to STIM1. Antibody binding was prevented with a blocking peptide (not shown).