

Supporting Information

Laser-synthesized Ligand-Free Au Nanoparticles for Contrast Agent Applications in Computed Tomography and Magnetic Resonance Imaging

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1. MATERIALS AND METHODS

Description of *in vivo* dual imaging by CT and MR of PC3 CAM tumors:

Briefly, fertilized chicken eggs purchased from Couvoirs Victoriaville (Victoriaville, Quebec, Canada) were incubated for ten days in a Pro-FI egg incubator fitted with an automatic egg turner before being transferred to a Roll-X static incubator for the rest of the incubation time (incubators purchased from Lyon Electric, Chula Vista, San Diego, CA). The eggs were kept at 37° C in a 60% humidity atmosphere for the full incubation period. On Day 10, a hole was gently drilled on the side of the egg and negative pressure was applied to create a new air sac. A window was opened on this new air sac and was covered with transparent adhesive tape to prevent contamination. A freshly-prepared cell suspension (40 µL) of PC3 (4 x 10⁶ cells/egg) cells was applied directly onto the newly-exposed CAM tissue through the window.

On Day 16, the embryos were submitted to low temperature conditions (40 min at 4° C) to minimize motion. 2 µL of Au NPs@PEG-Mn²⁺ in nanopure water (65.1 mM Au/ 1.2 mM Mn²⁺) were then directly injected into the tumor (30G-needle; Hamilton 1710-100 µL syringe; Ultra Micropump III injector; Micro 4 Controller from World Precision Instruments Inc., Sarasota, FL). One (1) implanted egg was MR-scanned before injection of Au NPs@PEG-Mn²⁺ NPs to serve as a control. Subsequently, the same egg and three (3) other implanted eggs were scanned within 10 to 15 minutes and at 18 hours following injection. For the MR scan (1.0 T MRI, M2M, Aspect Imaging, Netanya, Israel), each egg was placed in a 60 mm coil, and a T_1 -weighted echo sequence was performed twice to minimize motion artifacts (TE 18.3 ms, TR 1282.7 ms, dwell time 25 µs, field of view 60 mm, flip angle 90°, slice thickness 0.7 mm, interslice 0.1 mm, matrix 400x320, number of excitations 2, acquisition time 6.5 min). After the MRI scan, the eggs were kept at 37° C in a 60% humidity atmosphere for 1 hour 20 min and incubated at 4° C for 40 minutes before the

CT imaging study (once again to minimize motion). The CT scans were performed at 2 hours and 20 hours following injection. The eggs were scanned in a preclinical CT (eXplore Locus 80, GE Healthcare Technologies, Milwaukee, WI) at a peak voltage of 40 kVp and an x-ray tube current of 450 μ A. The detector exposure time was 90 ms at a detector binning of 4x4 for a voxel resolution of 89 μ m.

At the end of all imaging studies, the embryos were euthanized at 4° C followed by decapitation. CT image reconstruction and Hounsfield Unit (HU) calibration was performed using the Parallax Innovations Reconstruction tool from Parallax Innovations (Ilderton, ON, Canada). MRI and CT 3D images were generated using ImageJ software (version 1.47v, Wayne Rasband, National Institutes of Health, USA).

2. RESULTS

Table S1.	Dynamic light scattering	
	Sample	HD (nm)
Au NPs	17.5±4.9	0.3
Au NPs@PEG	40.9±9.8	0.2
Au NPs@PEG-DMSA-DTPA	34.1±6.2	0.1
Au NPs@PEG-Mn ²⁺	36.5±8.4	0.2

Table S1. Intensity-weighted average hydrodynamic diameter (HD) and polydispersity index (PdI) values of Au NPs, at the end of each step of the functionalization procedure, measured by DLS.

Figure S1:

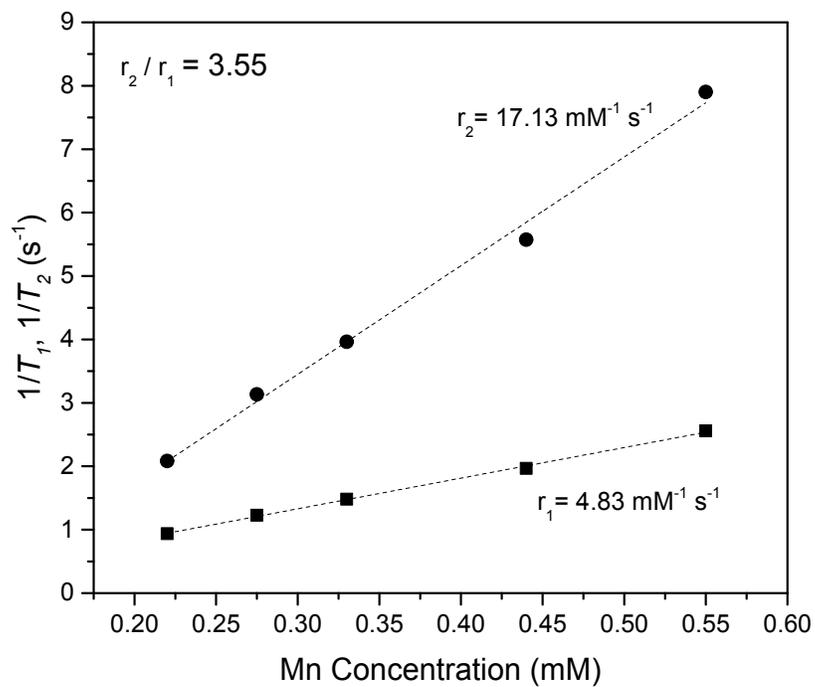


Figure S1. ¹H-NMR relaxometry analysis of Au NPs@PEG-Mn²⁺ performed at 37° C and 1.41 T.

Figure S2:

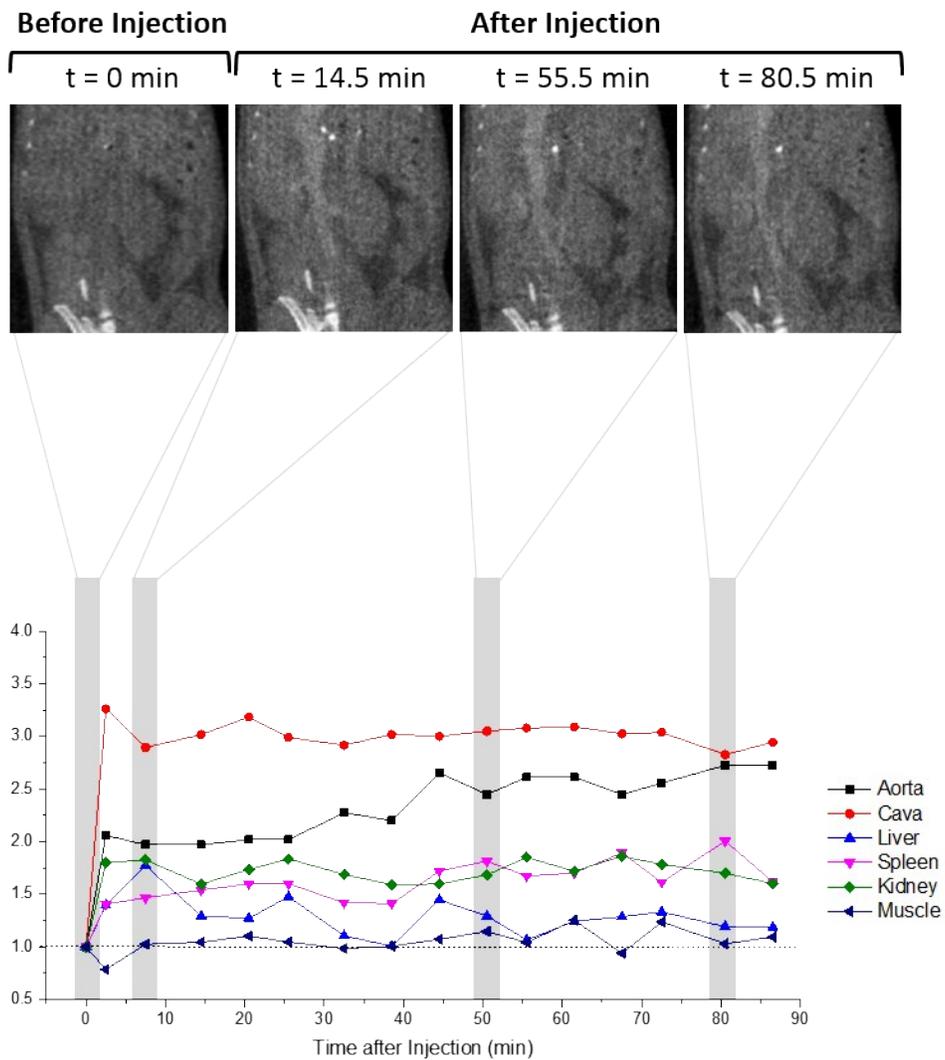


Figure S2. Dynamic contrast enhancement CT study of Aurovist™ using a healthy CD-1 mouse. Images show coronal sections of the abdominal region. The graph displays the contrast enhancement before (0 min), and at different time points after the injection.