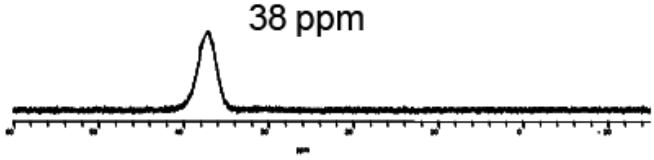
Synthesis of diethylphosphonate ester siloxane-coated tantalum oxide nanoparticles, ***2***

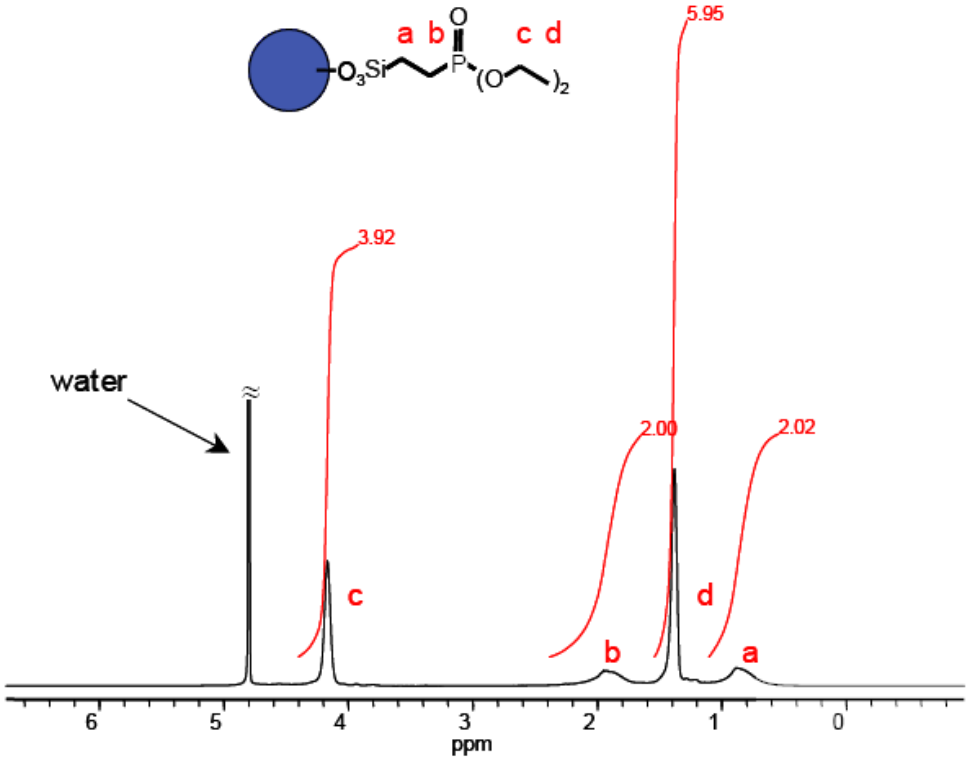
34 mL of *n*-propanol, 0.44 mL isobutyric acid, and 0.5 mL deuterium oxide were combined under nitrogen in the order specified and stirred for 30 minutes at room temperature. Tantalum ethoxide (1.87 g) was added in a drop-wise manner, albeit rapidly, and stirring continued under nitrogen for 16-18 hours to give a water-white clear solution of primary particles. Next, diethylphosphatoethyltriethoxysilane **1** (3 g) was added to the mixture as a 40 mL solution in *n*-propanol and the reaction was refluxed for 2 hours in air. Once cooled to room temperature, NH_4OH was added (0.1 M, 250 μL) and the reaction was allowed to stir overnight. Deionized water (40 mL) was added drop-wise, followed by hydrochloric (1.2 M, 10 mL), and then the reaction was allowed to react/stir for 2 days at 50°C. Upon cooling, the reaction was neutralized with NH_4OH to pH ~ 8-8.5 and filtered through a 100 nm membrane. All volatiles were removed to give the crude impure product. To purify **2** for further characterization and intravenous injection, **2** was solubilized in deionized water (pH 7.5-8), filtered through a 100 nm membrane, dialyzed in water against 3.5 kDa MWCO regenerated cellulose dialysis tubing (with 8-10 bath exchanges over 24 hours), and lyophilized to yield a snow-white flocculent precipitate approximately 30 % wt/wt in tantalum. IR (nujol mull, NaCl plates, cm^{-1}): 1454 (very strong), 1413 (weak), 1376 (strong), 1296 (very strong), 1274 (weak), 1218 (strong), 1170 (medium), 1035 (very strong), 963 (very strong), 782 (medium). NMR (D_2O, ppm): ^{31}P, 37.4 (broad); ^{1}H, 4.16, 1.88, 1.37, 0.85 (all broadened resonances).

NMR characterization

Diethylphosphatoethyltriethoxysilane ligand **1** provides distinctive features in the NMR solution spectrum to securely identify hydrolyzed **1** as a coating on the surface of tantalum oxide nanoparticles **2**. ^{1}H and ^{31}P NMR spectral resonances of the nanoparticle ligand in D_2O are broadened significantly, indicative of grafting onto the nanoparticle surface. The ^{31}P NMR spectrum exhibited a single resonance attributed to the fragment $-CH_2CH_2-P{=}O(OCH_2CH_3)_2$, which is centered around 38 ppm and broadened by nearly 10 ppm. The observed chemical shift and broadening agreed well with a reported value of ~ 35 ppm for the same ligand grafted onto silica particles (*J. Mater. Chem.* **2000**, *10*, 2758). Assignments for the ^{1}H spectrum are as follows: $P{=}O(OCH_2CH_3)_2$ at 4.16 ppm, $Si\text{-}CH_2CH_2\text{-}P$ at 1.88 ppm, $P{=}O(OCH_2CH_3)_2$ at 1.37 ppm, and $Si\text{-}CH_2CH_2\text{-}P$ at 0.85 ppm. Aside from the broadening, ^{1}H resonances are not significantly different or shifted compared to a spectrum of an authentic sample of **1**. The shifts are slightly downfield by ~ 0.03 ppm. Noticeably *absent* from the ^{1}H spectrum of **2**, are the triethoxysilane groups formally attached to silicon in starting material **1**. Of specific note is the lack of process residuals, particularly isobutyric acid, in the spectrum. Treatment of particles with HCl(aq) is thought to be sufficient to remove isobutyric acid (which is presumed to be surface-bound during primary particle formation).



Phosphorous NMR spectrum of **2** in D_2O.



^{1}H NMR spectrum of **2** in D_2O. Labeled resonances correspond to hydrogen atoms as indicated at the top of the figure. Integrated values reflect the proposed structure.

ICP characterization

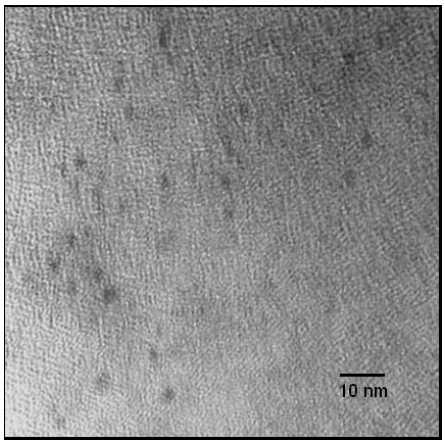
Freeze-dried (lyophilized powder) samples of **2** were analyzed as weight % Ta and Si using inductively coupled plasma atomic emission spectrometry, ICP-AES (Varian Liberty II). The samples were solid, static, and fluffy. Typically, microbalance replicates of 0.003-0.006 g were weighed and placed carefully into 50 mL orange cap tubes. To the samples were added first deionized water washing the walls, and then 1 mL HF, 2 mL HNO_3 and 0.5 mL H_2SO_4. This was diluted to approximately 40 mL and sonicated. Standards of Sc and Zr (ppm standards) were added before measurements. ICP-AES data repeatedly showed weight % Ta as 30 ± 1.

DLS characterization

The hydrodynamic size of the particles was measured by dynamic light scattering (DLS) at 25^{o}C using either a Brookhaven Instruments Corporation Zeta*PALS* or Malvern particle size analyzer. A hydrodynamic size of approximately 6 nm for **2** is based on the Z-average/-effective of intensity-based distribution.

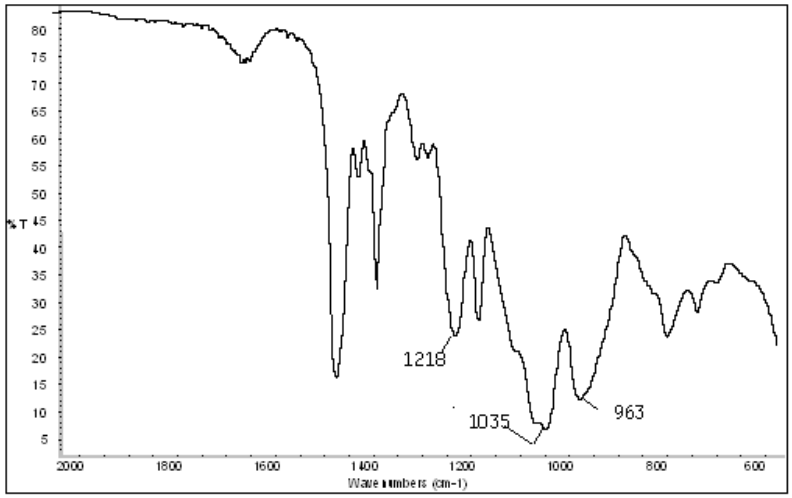
TEM characterization

TEM micrographs were obtained with a JEOL 2010 that operated at 200 kV equipped with a LaB6 thermionic emission filament. **2** was dispersed on a grid as a dilute aqueous solution and < 6 nm amorphous tantalum oxide cores were observed.



IR characterization

The IR spectrum of coated tantalum oxide particles **2** (as a Nujol mull between NaCl plates) is rich in the 800-1450 cm^{-1} region. Characteristic P-OR and P=O stretching bands for phosphonate esters, usually near 900-1050 cm^{-1} and 1200-1250 cm^{-1}(*Chem. Mater*. **2001**, *13*, 4367), are observed at 1035 cm^{-1} and 1218 cm^{-1}, respectively.



IR spectrum of coated tantalum oxide particles **2** as a Nujol mull between NaCl plates.

HPLC-ICP characterization

Approximately 200 mg of each freeze-dried preparation of **2** was dissolved in 1 mL deionized water and allowed to dissolve for at least 30 minutes. This stock solution was then diluted 1:5 with deionized water to a final concentration of ~ 5 mg Ta/mL. Each sample was then filtered through a 0.45 μm PTFE syringe filter, then placed in 2 mL crimp-top sample vials and analyzed by HPLC. Chromatographic analysis was achieved using an Agilent 1100 Series HPLC system equipped with an 1100 Series photo-diode array detector in-line to a Varian Liberty II Radial inductively-coupled plasma atomic emission spectrometer (ICP-AES). Separation was carried out using a Cadenza CD C18 2 x 50 mm reverse-phase column consisting of 3 μm particle media. Tantalum and silicon emission spectra were obtained separately from consecutive chromatographic runs of the each sample tested. The method limit of quantitation (LOQ) by ICP emission was determined to be 1 ppm for tantalum and 2 ppm for silicon. The method parameters were as follows:

Mobile Phase: 1) Solvent A, deionized water containing 0.1% Formic Acid.
2) Solvent B, 100% acetonitrile containing 0.1% Formic Acid.
Flow Rate: 0.4 ml min^{-1}
Photodiode Array Detector Acquisition: 225 nm and 254 nm
ICP-AES Emission: Silicon Emission –251.920 nm and Tantalum Emission – 263.558 nm
Column: Cadenza CD C18 3 μm (2 x 50 mm)
Injection Volume: 20 μL
Gradient Conditions: Presented in **Table SI1**.

Table SI1. Run Conditions for Reverse Phase HPLC Method.

Gradient Conditions	
Time (min)	**Percent Solvent A**
0	90
10	0
12	0
13	90
20	90
5 min Re-Equilibration	90

Viscosity Measurements by Capillary Viscometry

Viscosity measurements were carried out using an Anton Paar AMVn capillary viscometer to determine the dynamic and kinematic viscosity of aqueous suspensions of **2** at various concentrations. Viscosity profiles were obtained from 4-5 serial dilutions of 1M tantalum stock solutions. This measurement is based on the rolling/falling ball principle, which employs the use of a capillary tube ranging between 0.5 mm – 5 mm in internal diameter with a minimum sample volume of between 150 μL and 2.5 mL, depending on the sample viscosity. Measurement parameters for **2** used a capillary tube internal diameter of 1 mm and a sample volume 550 μL. The tilt angle was 70 degrees and an average of 4 measurements were obtained at 37 C. In deionized water, **2** has a viscosity of ~ 25 cps at 0.9 M tantalum (or 163 mgTa/mL).

In Vitro (Phantom) CT imaging

For clinical imaging, contrast agents must produce attenuation in the range of a hundred to many hundreds of HU. To achieve that, Ta- and I-based agents must be at a concentration of approximately 20-100 mM of the active element in the tissue of interest. Due to a blood dilution factor in the range of 10:1 to 20:1, these agents must be injected at a concentration of at least 1 M of the active element.

For our *in vitro* studies, samples of each agent were prepared with the same three molar concentrations: 20 mM, 50 mM, and 100 mM. Each sample was placed in an acrylic tube, ¼" ID x 3/8" OD x approx. 2" long. The tube was closed at both ends with ¼" diameter acrylic rod; air was displaced from the acrylic tube as the rod was inserted. The rods were sealed to the tube with wax.

The six sample assemblies (2 materials, 3 concentrations each) were then placed in a custom-designed phantom comprised of a closed acrylic cylinder (10cm OD x 3-5/8" ID x 8" long), with "bulkheads" to position the samples parallel to the phantom. The phantom was filled with water; so all samples were contained within a 10 cm water bath. The phantom was mounted on an alignment fixture that positions the cylinder parallel to the scanner's axis of rotation and centered in the scanner's bore. A sketch of this assembly is shown in **Figure SI1**, and a photograph of the phantom on the alignment fixture is shown in **Figure** SI2.

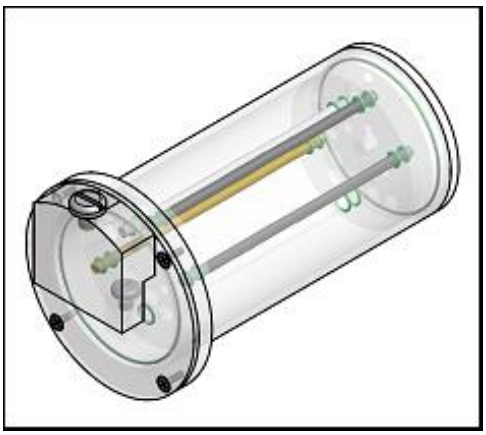


Figure SI1: MCD phantom assembly

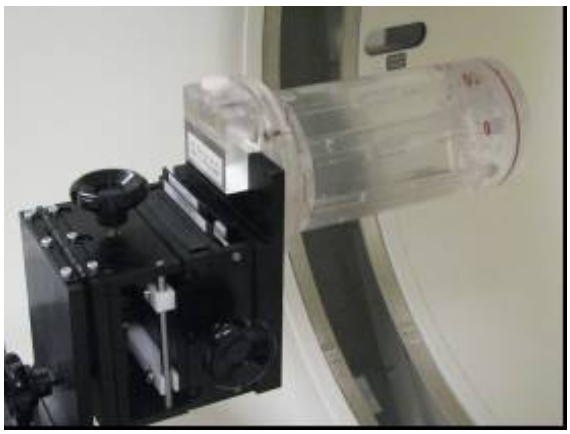


Figure SI2: MCD phantom on alignment fixture

The phantom was scanned at each of the kVps available using the scanner's clinical GUI – 80 kVp, 100 kVp, 120 kVp, and 140 kVp. From reconstructed images at each kVp, the average HU was recorded for each of the six samples. The average HU was determined using the scanner's console GUI, by placing a circular region of interest (ROI) over each ¼" diameter region that contained contrast agent.

Results at each kVp were linear with molar concentration; therefore the relative results *vs*. kVp were the same for each concentration. For simplicity, only 100 mM concentration is shown in **Figure 2A** (see main body text).

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In vivo imaging

Female Dark Agouti rats were anesthetized *via* intraperitoneal injection of ketamine/ diazepam immediately prior to the imaging exam. A 24 gage, 5/8 inch catheter was placed in the tail vein for intravenous injection of 1 mL of contrast agent at a rate of 143 μL/sec using a syringe pump. Rats were imaged on a GE Lightspeed 64 slice CT scanner at 140 kVp, 160 mA with a 0.4 sec gantry rotation and images were performed continuously after injection for 30 seconds. Images were reconstructed using a 25 cm FOV, 0.625 mm slices and "lung" reconstruction filter. Images in **Figure 2B** (see main body text) were obtained immediately before injection and 10 sec following injection to highlight contrast in vena cava and abdominal aorta.