High r1 Relaxivity Sub-5 nm Suprasmall Iron Oxide Nanoparticles (sSIOs) as Intravascular T1-weighted MRI and Cell Tracking Contrast Agents

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INTRODUCTION

Superparamagnetic iron oxide nanoparticles (SPIOs) have been widely used as MRI contrast agents both in clinical applications (e.g. liver, lymph nodes imaging) and preclinical models (e.g. cell tracking and molecular imaging).[1,2] Currently, most SPIOs are fabricated with size larger than 5 nm, therefore exhibit predominant shortening effect on the transverse relaxation time T_2, causing signal void in T_2-weighted MRI. Early studies have shown that r_1 and r_2 relaxivities of SPIOs are size-dependent, while r_1 relaxivity can be preserved in the ultrasmall iron oxide nanoparticles with size below 5 nm.[3,4] Here we report a new class of high r_1 relaxivity, sub-5 nm, suprasmall iron oxide nanoparticles (sSIOs, SIO-4, core size ~3.5 nm) with oligosaccharide coating as intravascular T_1-weighted MRI contrast agents while providing reverse T_2 contrast, a T_1-T_2 contrast switch, when taken up by cells and liver, or between sSIO labeled and lysed cells.

MATERIALS AND METHODS

The hydrophobic iron oxide nanoparticles (IOs) with different sizes were prepared by pyrolysis. After coated with oligosaccharides, the particles (SIO-4, 10, 20) were transferred to aqueous solution. Transmission electron microscopy (TEM) was used to determine the core sizes. SIO solutions at different concentrations were examined on a 3T MRI scanner using T_1, T_2-weighted spin echo sequences, inversion recovery spin echo sequence with different inversion times, multi-echo T2-weighted spin echo sequence. Multihance® (Gd-BOPTA) was used for comparing MRI contrast enhancing effect. r_1 and r_2 relaxivities were calculated by fitting signal changes in multi-IR T_1 and multi-TE T_2 images. To test the T_1-T_2 contrast switch for tracking magnetic nanoparticle labelled cells, human breast carcinoma MDA-MB-231 cells or macrophage (RAW264.7) were incubated with SIOs, and then lysed to release SIOs, Cells in two different conditions were subsequently analyzed by TEM and MRI. To test MRI contrast enhancing effects of SIOs in vivo, SIOs and Gd-BOPTA were intravenously administrated into mice. Fat suppressed T_1-weighted spin echo images were obtained before and after injection of contrast agents to investigate the contrast changes in liver, kidney and iliac artery at the different time points.

RESULTS

TEM images show that the prepared SIO-4 has an averaged core size of 3.5 nm (Fig. 1). They are highly uniform and monodispersed. From MRI phantom studies, SIO-4 exhibits high r1 value (4.5 $mM^{-1}s^{-1}$) at 3T. In particular, SIO-4 has a higher r_1/r_2 ratio (0.23) than those of typical SPIOs with larger sizes. In comparison, Gd-BOPTA, r_1 is 5.8 $mM^{-1}s^{-1}$, and r_1/r_2 is 0.85 while SIO-10 has the highest r_1 value at 6.4 $mM^{-1}s^{-1}$, but much lower r_1/r_2 ratio ~0.09. Furthermore, SIOs exhibit good biocompatibility based on the results of MTT assay. When SIOs were used for cell labelling, high cellular uptake of SIOs was demonstrated by both TEM and MRI. TEM images (Fig. 2) showed SIO-4 and SIO-20 were engulfed and internalized in lysosomes, which led to signal drops (darkening) in both T_1 and T_2-weighted spin echo images. However, significant "bright" contrast enhancement was observed in T_1-weighted MRI when the labelled cells were lysed and SIO-4 were released, suggesting that there is a "contrast switch" between SIO-4 internalized in cells and released from cells. Therefore, it is possible to not only track sSIO labelled cells but also monitor their fate (apoptosis) using MRI. For in vivo MRI experiments, SIO-4 showed excellent T_1 contrast enhancement in the kidney and iliac artery with the effect similar to that observed in Gd-BOPTA enhanced MRI (Fig. 3). However, T_1 contrast enhancement is not obvious when using larger size SIO-10 or 20. The "double" contrast in liver shown by SIO-4 further improves the image quality for visualizing liver tissue and hepatic vasculature. SIO-4 has a much longer blood retention time than small molecule Gd-chelates for prolonged imaging time for organs of interest.

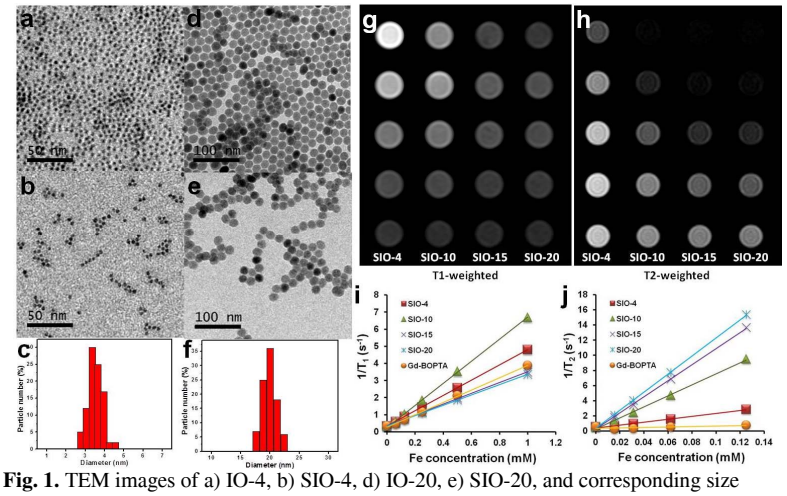

Fig. 1. TEM images of a) IO-4, b) SIO-4, d) IO-20, e) SIO-20, and corresponding size distribution of c) SIO-4, f) SIO-20; g) T_1-weighted, and h) T_2-weighted MR phantom studies of SIOs, and corresponding calculation of relaxivities i) r_1 and j) r_2.

CONCLUSIONS

Sub-5 nm suprasmall iron oxide nanoparticles (sSIOs, SIO-4) were made with high R_1 relaxivity for intravascular T_1-weighted MRI. sSIOs exhibit excellent T_1 contrast enhancement in kidney and iliac artery, providing a potential long half time blood pool MR imaging agents. Furthermore, T_2-to-T_1 contrast switching in cells labeled with sSIOs may enable MRI monitoring of the fate of labeled cells in vivo.

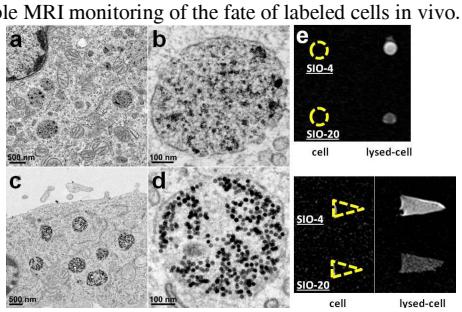

Fig. 2. a, b) TEM images of cells after uptake of SIO-4 and c, d) SIO-20; e) T_1-weighted MR images of cells after uptake of SIOs.

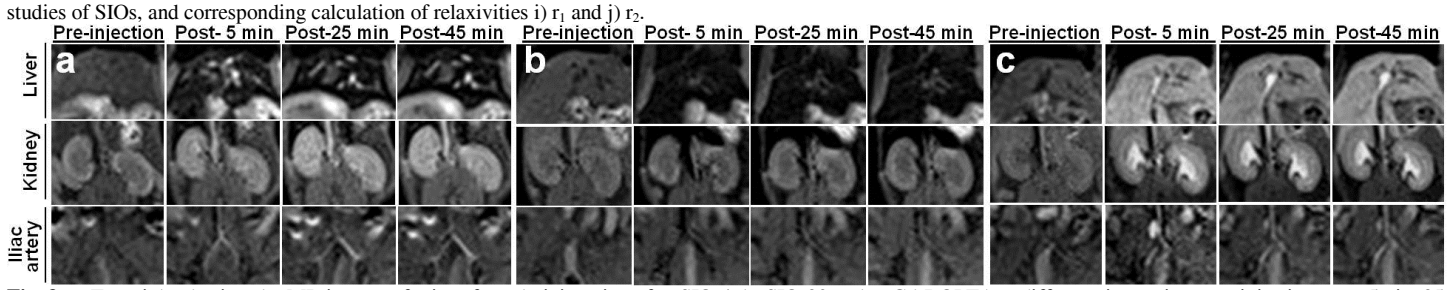

Fig. 3. a) T_1-weighted spin echo MR images of mice after administration of a) SIO-4, b) SIO-20 and c) Gd-BOPTA at different time points, pre-injection, post-5min, 25 min and 45 min. The contrast enhancement by SIO-4 is calculated to be 30% for kidney and 50% for liver.

References:

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