Enhanced targeted MRI contrast using silica coated magnetite nanoparticles

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Introduction

MRI is a high spatial resolution non-invasive technique but it has low specificity for targeting explicit pathologies. In an effort to improve specificity, MRI contrast agents with strong paramagnetic properties such as Gadolinium have been developed. However recent reports of suspected deaths associated with gadolinium administration have severely limited their use [1-2].

Iron oxide contrast agents have been used for MRI signal enhancement however particle size issues have limited their imaging to liver, spleen and the gastro-intestinal lumen. To achieve a more targeted delivery an iron oxide based MRI contrast agent must be biocompatible, have high chemical stability, be easily functionalised and retain a high net magnetisation value.

In this study, we used silica coated iron oxide to generate highly magnetic nanoparticles small enough to by-pass the reticulo-endothelial system and localise in a specific tumour. We aimed to demonstrate that targeted MRI diagnosis of cancer is possible by the development of highly magnetic iron oxide nanoparticles labelled to a specific breast tumour antibody. We outline the characterisation, high T2 relaxivities and subsequent *in-vitro* and targeted *in-vivo* MRI imaging.

Experimental

Phantom assessment - The silica coated magnetite nanoparticles were fabricated using protocols outlined by Park et al [3] and Fang et al [4]. These iron oxide silica particles were labelled with herceptin using a cysteamine linker. The particles were characterised by high resolution transmission electron microscopy (HRTEM), x-ray diffraction (XRD) and superconducting quantum interference device (SQUID). Phantom studies were conducted on a 3.0 Tesla Siemens Trio MRI scanner using an echo sequence with multiple TE times (50-300ms) and a TR of 2000ms.

In-vitro assessment - Cell uptake studies were conducted using herceptin labeled nanoparticles on human breast cancer cell lines SKBR-3, BT474 and MCF-7. Cell lines were assessed for HER2-neu surface expression by flow cytometry and dose response and time course studies were performed to assess uptake of herceptin-labeled nanoparticles. These cells were then imaged in a 3.0 Tesla Siemens Trio MRI scanner with the above parameters.

In-vivo assessment - BALB/c nude mice were inoculated with SKBR-3 human tumour cells subcutaneously (n=9). Acute toxicity studies were performed by injecting the maximum iron oxide silica herceptin dose intravenously in nude mice. Tumours were grown to a maximum size of 0.8cm in diameter and injected with iron oxide silica herceptin nanoparticle to assess their targeting capabilities.

Results and discussion

HRTEM and XRD showed that iron oxide nanoparticles coated in silica were synthesized with a diameter of 40nm. SQUID and T2 measurement data reveal highly magnetic particles with **80emu/g and a T2 of 235.5 mmol^{-1}l s^{-1}.** Compared to available iron oxides such as AMI-25 (Feridex) and AMI-227 (Sinerem) our particles have higher values. Phantom studies of different iron oxide concentrations showed approximately 95% signal enhancement compared to the control (Figure 1). Confocal and flow cytometry dose response and time course (Figure 2) studies verified that the nanoparticles localized inside the cells. The results showed different levels of iron oxide uptake relative to HER2-neu expression of each cell line indicating the specific binding of the iron oxide silica herceptin nanoparticles (Figure 2). MRI of the 3 cell lines with localized nanoparticles showed the same signal enhancement as the phantom studies.

In-vivo toxicity studies in BALB/c nude mice showed no acute toxicity. Iron oxide silica nanoparticles injected into tumour bearing BALB/c nude mice showed significant MR signal enhancement at the tumour site.

Conclusion

This study using a highly efficient biocompatible iron oxide nanoparticle with well defined magnetic properties was able to specifically target and image a cancer, it also demonstrates the potential for targeted iron oxide silica nanoparticles in the MRI of specific pathologies.

References

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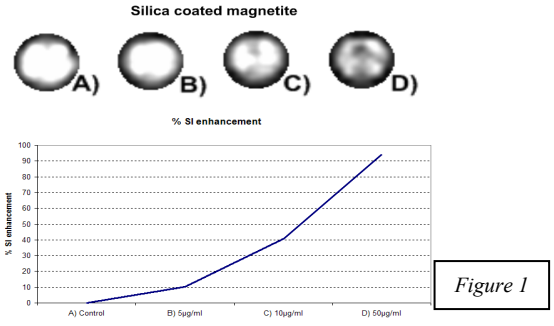

Figure 1

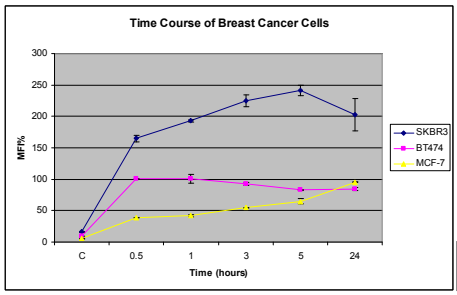

Figure 2