

NEWLY DEVELOPED NANOPARTICLES FOR CELL TRACKING USING MAGNETIC RESONANCE IMAGING

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INTRODUCTION

The utilisation of magnetic resonance imaging (MRI) to noninvasively monitor cells using superparamagnetic iron oxide nanoparticles (SPIONs) as MRI contrast agents has been one of the major research focuses over the past several years. The ability to use MRI to image longitudinally offers an invaluable opportunity to track the migration, persistence and distribution of cell-based therapeutics *in vivo* in humans. Current approaches for cell labelling have focused on using commercial SPIONs with varying success. Therefore, there is a clinical need to develop SPIONs that are non-toxic, with appropriate size and high MRI contrast. We have utilised SPIONs with a silica coating to investigate the labelling ability and contrast enhancement of CD45⁺ and CD34⁺ cells.

EXPERIMENTAL

Silica SPION synthesis and analysis

Silica coated SPIONs were formed using a large scale thermal decomposition method¹ and were coated in silica using a well known modified Stober process². The SPIONs were analysed using high resolution transmission electron microscopy (HRTEM) and x-ray diffraction (XRD).

Cell preparation

Peripheral blood mononuclear cells were collected by apheresis from appropriately consented human donors for subsequent sorting using FLOW cytometry to generate a CD45⁺ fraction and a CD34⁺ fraction representing a conventional mononuclear leukocyte fraction and a CD34⁺ haematopoietic progenitor fraction respectively.

Cytotoxicity studies

Cytotoxicity of the silica coated SPIONs was assessed after incubation of silica coated SPIONs at 5µg, 10µg, 50µg and 100µg using MTT following manufacturers protocol.

MRI studies

Approximately 20,000 cells were seeded and incubated with 5µg, 10µg, 50µg and 100µg of silica coated SPIONs for 6hrs at 37°C. MRI was performed with a clinical 3.0 Tesla Siemens Trio scanner.

RESULTS

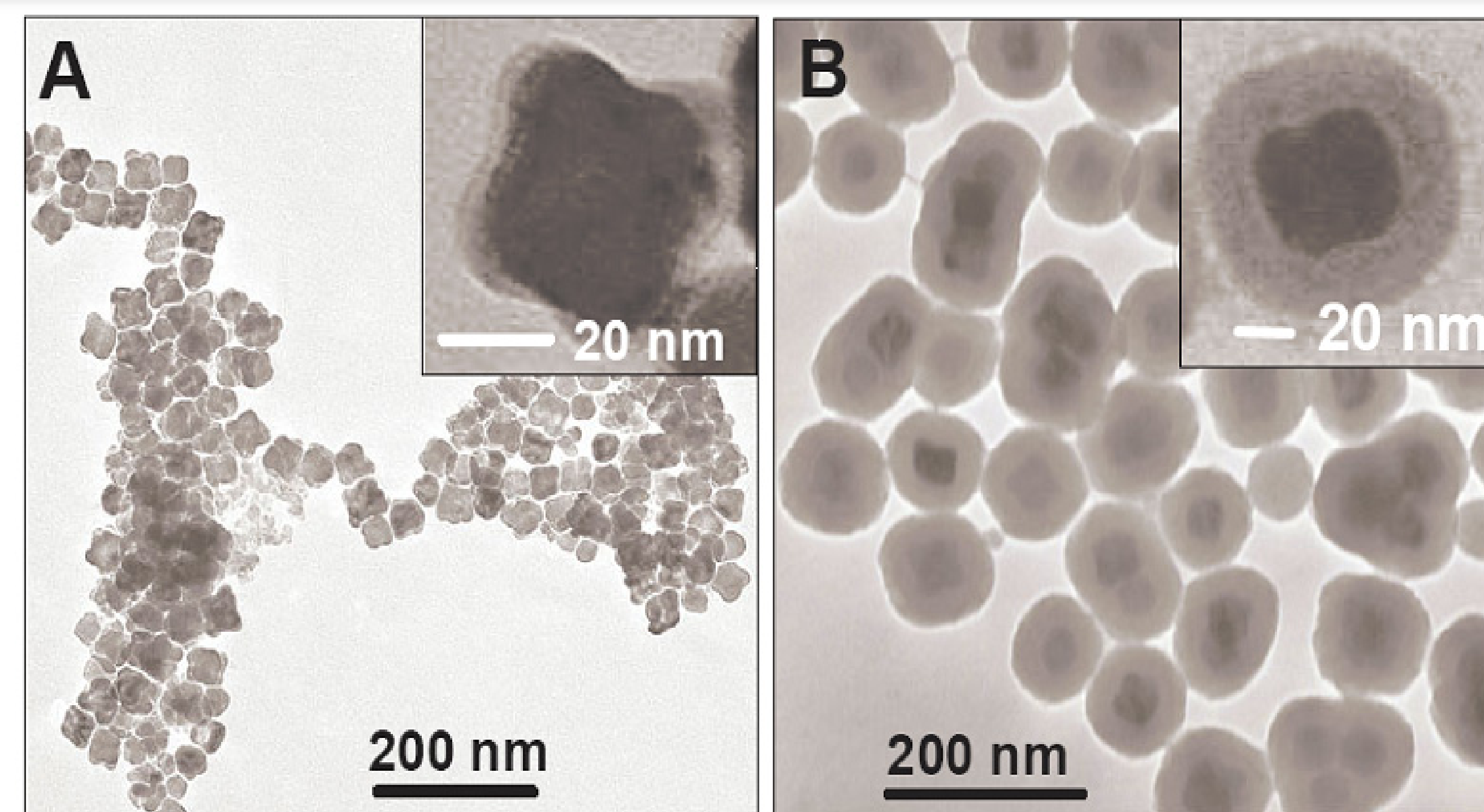


Figure 1. HRTEM images of 60 ± 7nm as synthesised SPIONs (A) and silica coated SPIONs (B)

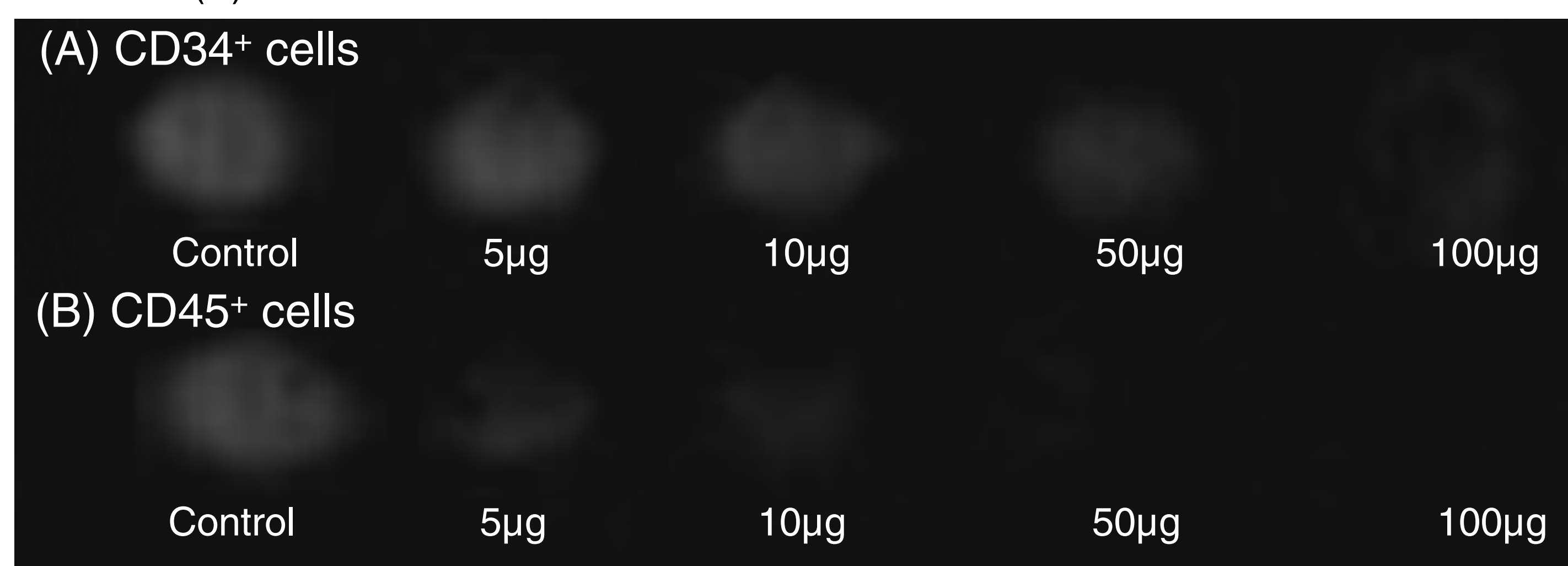


Figure 2. MRI images of CD34⁺ and CD45⁺ cells labelled with silica coated SPIONs (A) CD34⁺ cells at 5µg, 10µg, 50µg, 100µg, (B) CD45⁺ cells at 5µg, 10µg, 50µg, 100µg

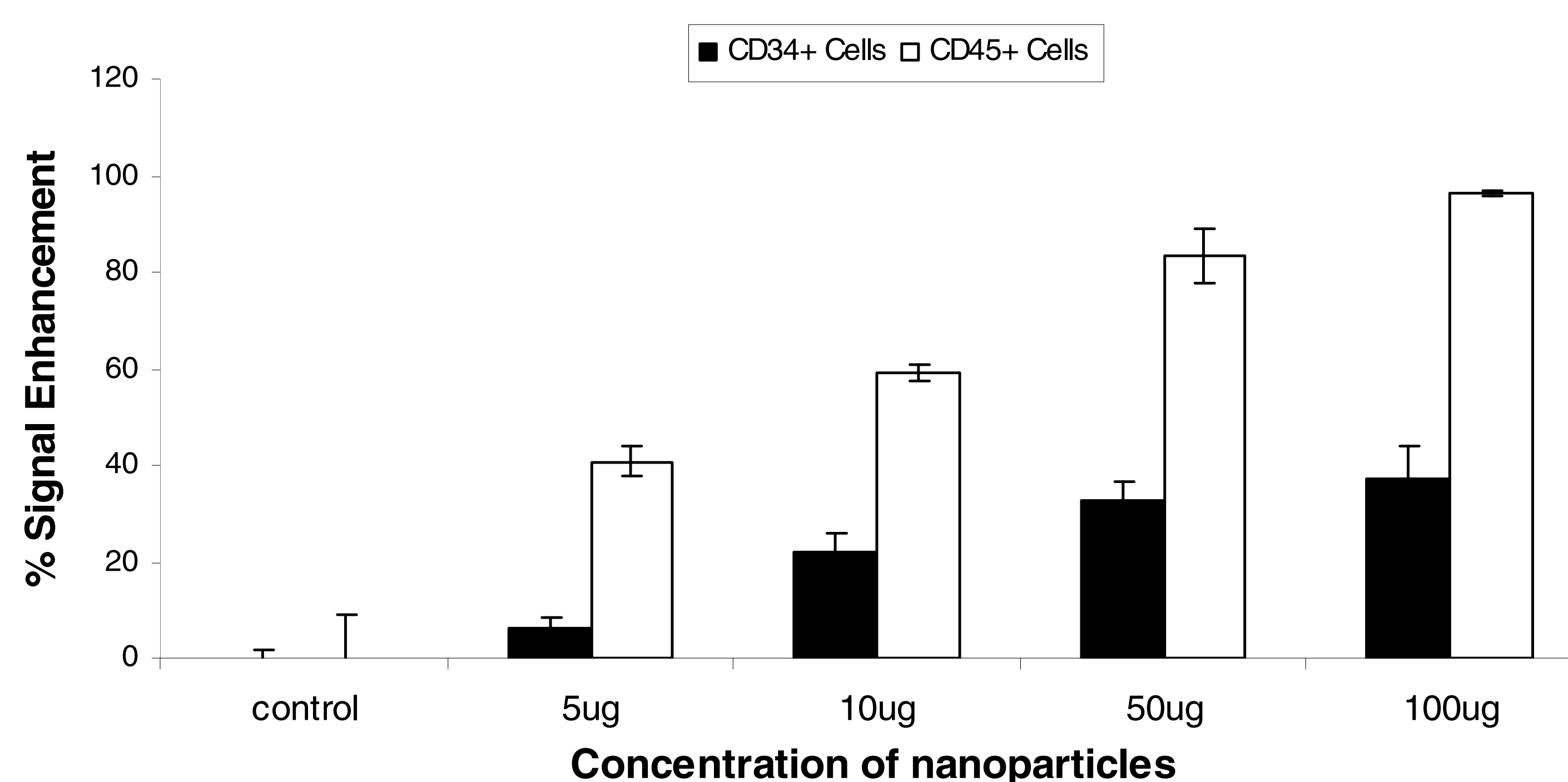


Figure 3. % of signal enhancement of silica coated SPIONs (A) CD34⁺ cells at 5µg, 10µg, 50µg, 100µg, (B) CD45⁺ cells at 5µg, 10µg, 50µg, 100µg

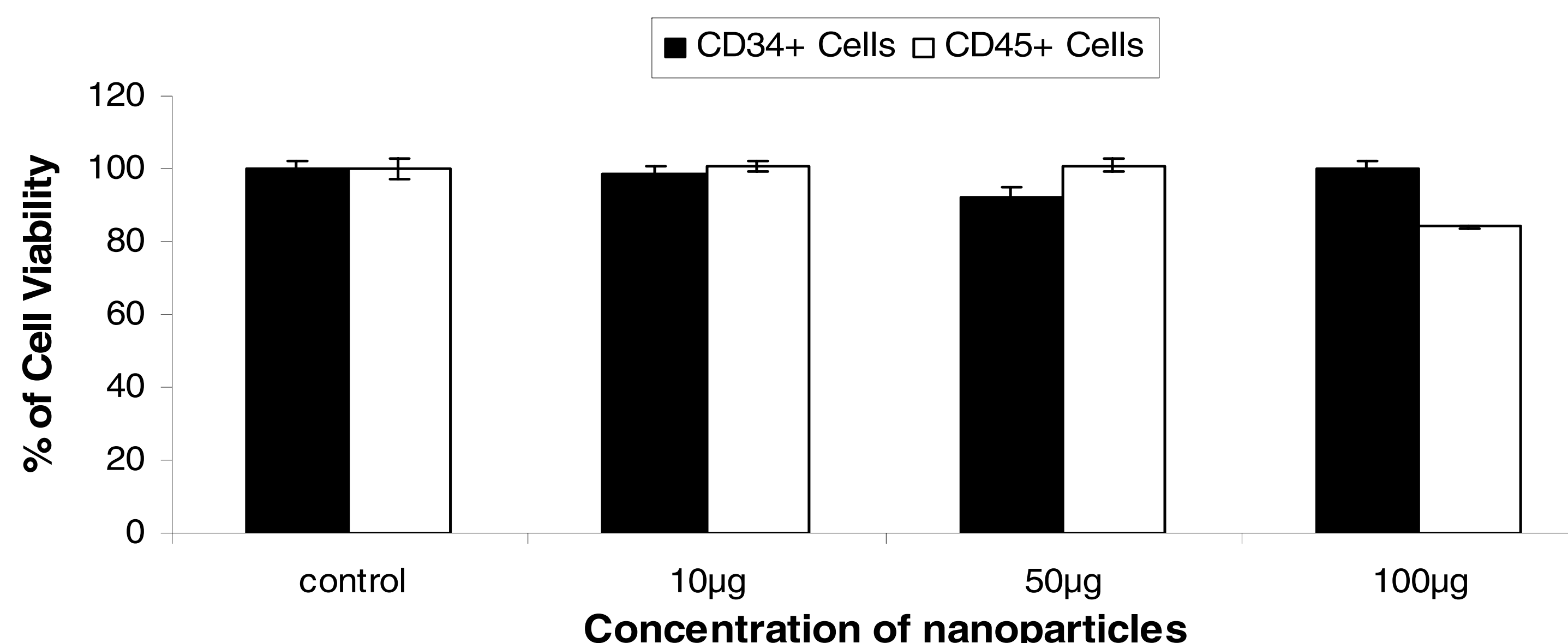


Figure 4. MTT toxicity analysis showing the viability of cells after incubation of silica coated SPIONs at 10µg, 50µg, 100µg

DISCUSSION

Silica coated SPIONs

The TEM analysis demonstrates that the silica coated SPIONs were 60 ± 7 nm. The XRD data indicates the formation of iron oxide coated in silica and the magnetic values are suggestive of very high magnetic nanoparticles (data not shown).

In vitro studies

The MRI studies demonstrates significant contrast enhancement up to 96%. This suggests that silica coated SPIONs are taken up by the cells. The CD45⁺ cells have higher contrast enhancement compared to the CD34⁺ which indicates higher uptake of the nanoparticles.

Cytotoxicity studies

The toxicity results suggest that the silica coated SPIONs are not toxic to both cells at low concentrations but are mildly toxic at concentrations above 50µg. The CD34⁺ cells have lower toxicity compared to the CD45⁺ cells possibly related to the low number of particles internalised as indicated by the contrast enhancement (Figure 2 and Figure 3).

CONCLUSIONS

This pilot study showed that the newly prepared silica coated SPIONs can be used for safe and effective cell labelling of human cells. Due to optimal size and possible higher uptake of the nanoparticles, significant contrast enhancement was visualised *in vitro*. The results indicate that smaller number of cells may be needed for MRI detection. Overall, these results provide preliminary evidence that high efficiency biocompatible labels can improve future cell tracking using MRI and SPIONs as contrast agents.

REFERENCES

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