

MR imaging on a clinical 3T system of ex-vivo mouse joints with SPION uptake in an arthritis model using 3D difference Ultrashort Echo Time images

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Purpose

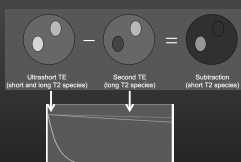
Super paramagnetic iron oxide particles (SPION) accumulate in macrophages in the synovium of arthritic knees (1).

We developed an MRI protocol for fast scanning of multiple mice knee specimens on a clinical 3T MR scanner.

We present its performance for SPION detection in an antigen induced arthritis (AIA) model in mice.

This protocol is based on a 3D difference Ultrashort Echo Time (dUTE) MR sequence that provides quantifiable positive contrast method for iron oxide detection in rodent AIA model in vivo (2).

Context

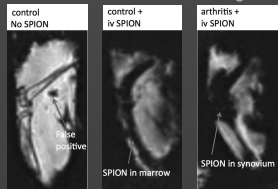


dUTE gives high-resolution 3D anatomical images with suppressed background, reduced artifacts and high iron contrast, allowing automatic quantification of iron oxide

'Multipaw' increases scan efficiency and signal homogeneity: simultaneous scanning of a grid of 42 samples in a wrist coil gives time efficient scanning and better, even SNR over the whole sample compared to small batches in a dedicated surface coil.

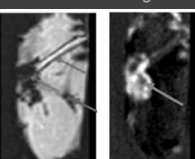
Observations: Images

Normal and arthritic knee imaged using VIBE + SPION



Sagittal slice of mice knee specimens imaged by a VIBE sequence in control (left), after an iv iron oxide (SPION) injection in normal (middle) and arthritic (right) mouse knee. Accumulation of SPION in the synovium is only seen in arthritic knee whereas bone marrow uptake is present in both normal and arthritic knee

Arthritic knee imaged using dUTE + SPION



False positive signal drop on VIBE image (green arrow) not seen in the dUTE image. SPION induced signal drop on VIBE (blue arrow) but signal enhancement on the dUTE sequence. Bone did not show SPION uptake (red arrow) after ia injection in AIA knee

In the figure we see that false positive signal drop on VIBE image is not seen in the dUTE image. Due to reduced "false positive" from artifacts dUTE outperformed VIBE for the quantification of SPION uptake. We have SPION induced signal drop on VIBE but signal enhancement on the dUTE sequence. In addition, bone did not show SPION uptake (red arrow) after ia injection in AIA knee

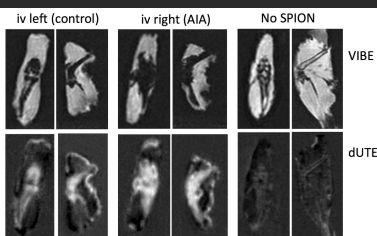
Control knee imaged using dUTE + SPION



Bone marrow SPION uptake after high dose iv injection in a control knee on VIBE and dUTE

Image Fusion of dUTE SPION on VIBE anatomy in a control knee after ia injection, and without SPION

SPIONs VIBE versus dUTE



Coronal and sagittal slices of mice knee specimens imaged by a VIBE sequence (upper) and dUTE (lower) after an iv iron oxide (SPION) injection in control and AIA mice knees. Accumulation of SPION in the synovium is only seen in arthritic knee whereas bone marrow uptake is present in both normal and arthritic knee. The third case shows an AIA knee with no SPION

References and Acknowledgements

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Methods

All particles described in this work are amino-PVA-SPIONs provided by EPFL, Lausanne, and University of Fribourg (3,4).

Animal model: Arthritis was induced in mice using a standard AIA protocol in conformance with the institution ethical committee. SPIONs were injected at day 3 after AIA induction

At timepoints up to day 7 after SPION injection, mice were sacrificed to obtain multiple knee samples

intra-articular (ia) injection of 6µg amino-PVA-SPION

intra-vascular (iv) 1mg of the same amino-PVA-SPION

Both arthritic and control samples (right and left knees) were taken

Ex vivo mouse knees held tightly in sealed plastic tubes arranged in holder with joints aligned forward.

156 knees in total have been scanned with this method in batches of up to 42, with varying doses and timepoints. The results presented have n=3-6 for each route and dose of SPION injection as well as for each timepoint

Magnetic Resonance Imaging:

dUTE sequence parameters were

Siemens 3T Tim Trio clinical scanner with the wrist coil

3D difference Ultrashort Echo Time (dUTE)

3D T1 gradient echo sequences, called 'VIBE' throughout

3D isotropic matrix of 512, 90mm FOV (field of view), = 180µm isotropic resolution, 50000 radial projections, ultrashort TE (echo time) of 0.07ms, TE2 2.46ms, TR (repetition time) 9.6ms, flip angle 10°

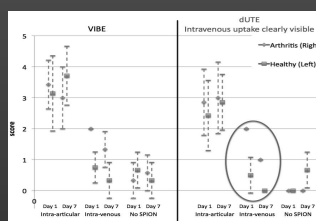
VIBE parameters were: 3D isotropic resolution of 310µm, TR/TE 14.3/5.9ms and flip angle 12°

Whole examination for a single group: 20 minutes repeated for 7 signal averages with 42

knees at a time corresponding to an effective acquisition of <4 min for a single knee for both VIBE and dUTE

Score for presence of SPION to compare groups and MR sequences. Blind scoring was done on a 5-point scale on each knee, after random arrangement in the wrist coil set-up

Observations: Quantification

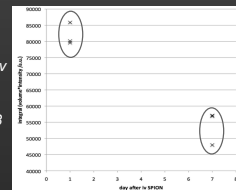


Comparison of SPION accumulation scoring (n=3-6 per group) at day 1 and 7 using both VIBE (left) and dUTE (right) sequences

Due to reduced "false positive" from artifacts dUTE outperformed VIBE for the quantification of SPION uptake

After iv injection of SPION, only dUTE was able to demonstrate a statistically significant uptake at day 1 and following decrease at day 7

Quantification of SPION signal after iv injection on dUTE positive contrast images 1mg dose, 3 animals per group



The graph above demonstrates that dUTE is able to detect a time decrease of the SPION uptake in the synovium of AIA. This could reflect the migration of the macrophage as the inflammation is resolving or a destruction of the SPION and further recycling of the iron secondary to apoptosis of the macrophages

The graphs shows the blinded reader scores for both AIA knees and control (no arthritis) knees. For clarity, the results presented on the figures are from day 1 and day 7 with groups also assessed statistically including 4 hour and 4 days. A Bonferroni ANOVA was carried out over all the cases (20 groups in all) with a significance set at 0.05

After ia injection, both arthritic and control knees demonstrated an important SPION signal not evolving over time (no significant difference between knees and timepoints). VIBE showed non-significance comparing iv and no SPION, including comparison of control and AIA knees. For dUTE the AIA knee after iv showed significant difference from the control, from ia SPION and no SPION

Discussion

- The spatial resolution achieved by our protocol was high enough to identify relevant anatomical landmarks of the mice knee
- Presence of SPION easily detected by a strong signal loss in the bone marrow after iv injection
- Accumulation of iron in the synovium of AIA knee was visible on both VIBE and dUTE
- However, dUTE was more efficient than VIBE when grading the amount of SPION
- After iv injection of SPION, only dUTE was able to demonstrate a statistically significant uptake at day 1 and following decrease at day 7
- On day 1 after iv SPION injection, there was a significant change of signal intensity in the synovium of arthritic knee that was not present in control knee. This SPION signal decreased at day 7 but was still detectable in arthritic knees indicating that the SPION quantification could be efficient to monitor effect of treatment in this arthritic model in mice
- dUTE but not VIBE was able to demonstrate after iv iron injection an uptake of SPION in arthritic knee that decreased after 1 week
- The diagnostic ability of dUTE on a clinical scanner in mouse knee samples is illustrated
- In agreement with previous publications, dUTE was the most efficient MR sequence to quantify small variation of SPION accumulation in the knee
- Applying this protocol on 156 mice knee specimens demonstrated that the signal loss after ia injection of iron was not different between control and arthritic knee and that it did not evolve over a 7 day period. Signal saturation related to a too high dose of injected ia SPION may explain this paradoxical absence of signal clearance

Conclusions and Consequences

We demonstrated a robust, convenient and easily implementable protocol on a clinical MR system for fast assessment SPION uptake in knee specimens of arthritic mice with a strong potential for drug studies.

False positives on conventional signal loss images give overestimate of iron score.

Only SPION is bright on dUTE, with all other tissue signals suppressed.

Using this protocol, iron uptake kinetic in arthritic knee can be diagnosed after iv injection in a mice model of AIA.