Frequency Shift Imaging (FSI) for characterization of cells labeled with superparamagnetic iron-oxide nanoparticles

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Introduction

MRI has been a primary tool used for detecting and tracking the location of cells labeled with superparamagnetic iron-oxide nanoparticles (SPIOs). While most SPIO applications have relied on negative contrast sequences (1), positive contrast imaging of the off-resonance SPIO signal provides clear benefits (2, 3), such as reduction of background tissue signal and possible quantification of labeled cells in the targeted organ. Moreover, SPIO imaging at ultrahigh field strengths, such as 7 Tesla (7T), makes it possible to leverage the greater off-resonance sensitivity afforded by higher field strengths to provide quantitative imaging of smaller cell populations. In this work we introduce frequency shift imaging (FSI), a novel acquisition technique that combines efficient interleaved spectrally selective excitations with fast spiral acquisition to perform comprehensive characterization of the magnetic signature of SPIOs in a reasonable scan time. We demonstrate the performance of the FSI sequence for imaging macrophages labeled with SPIOs and compare the novel sequence with standard negative-contrast acquisitions performed at 7T.

Methods

Cell phantom

Mouse tumor macrophages were grown in DMEM media in two T75 flasks. Both flasks were incubated for 4 hours at 37° Celsius, one with 30 mg Fe/mL ferumoxutol (labeled cells), the other without (control cells). Subsequently, the cells were fixed with 4% PFA and four samples (of 1 million cells each) were made by mixing labeled and unlabeled cells at 0%, 25%, 75% and 100% population ratio. The cell samples were embedded in 2% agar gel for MR imaging.

Sequence Design

A 15 ms-long, 170 Hz bandwidth, self-refocused RF pulse was designed using the Shinnar-Le Roux (SLR) algorithm (**Fig. 1**) in order to provide minimum echo time and high spectral selectivity, as described in (2). Center-out 3D stack-of-spiral readouts were implemented to minimize the echo time in order to reduce signal loss due to transverse relaxation. 15 different frequency points (-1400 to 1400 Hz) were sampled at 200 Hz intervals in interleaved fashion. Interleaving frequency sampling in time allows for fast repetition rates (24 ms between consecutive pulses) while providing sufficient signal recovery (T_R=360 ms between consecutive excitations of the same frequency band).

MR imaging

The stack-of-spiral FSI sequence acquired 30 partitions at isotropic 1.0 mm resolution, using segmented spiral readouts (140 mm field of view, 15 arms per 2D spiral, 5.5 ms per readout). Total acquisition time for interleaved FSI was 10 minutes. In addition, standard negative-contrast sequences were acquired at 1.0 mm isotropic resolution on the same phantom, including a 3D gradient echo (TE/TR 2.4/5.3 ms) and a 2D interleaved spin echo (TE/TR 8.7/1000 ms).

Methods and Results

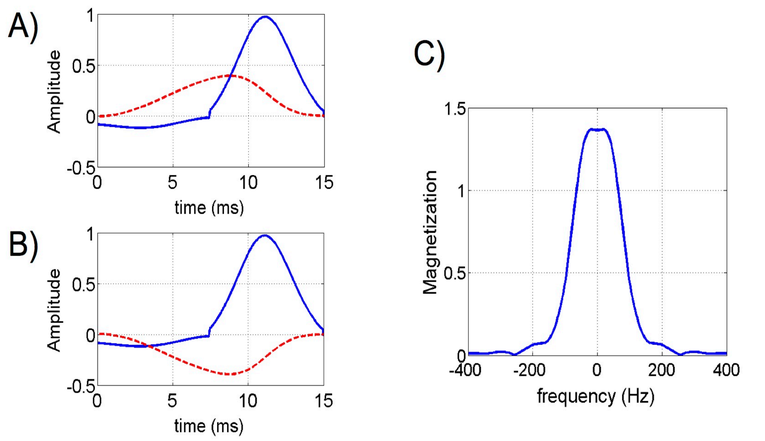

Figure 1: (A-B) waveforms of the self-refocused RF pulses used in this work (continuous line: real part, dashed line: imaginary part). The two pulses are generated to provide opposite echo phase in subsequent acquisition. The spin echo is sampled at the end at the pulse. (C) Bloch simulation showing the frequency dependence of the subtracted spin echo signal.

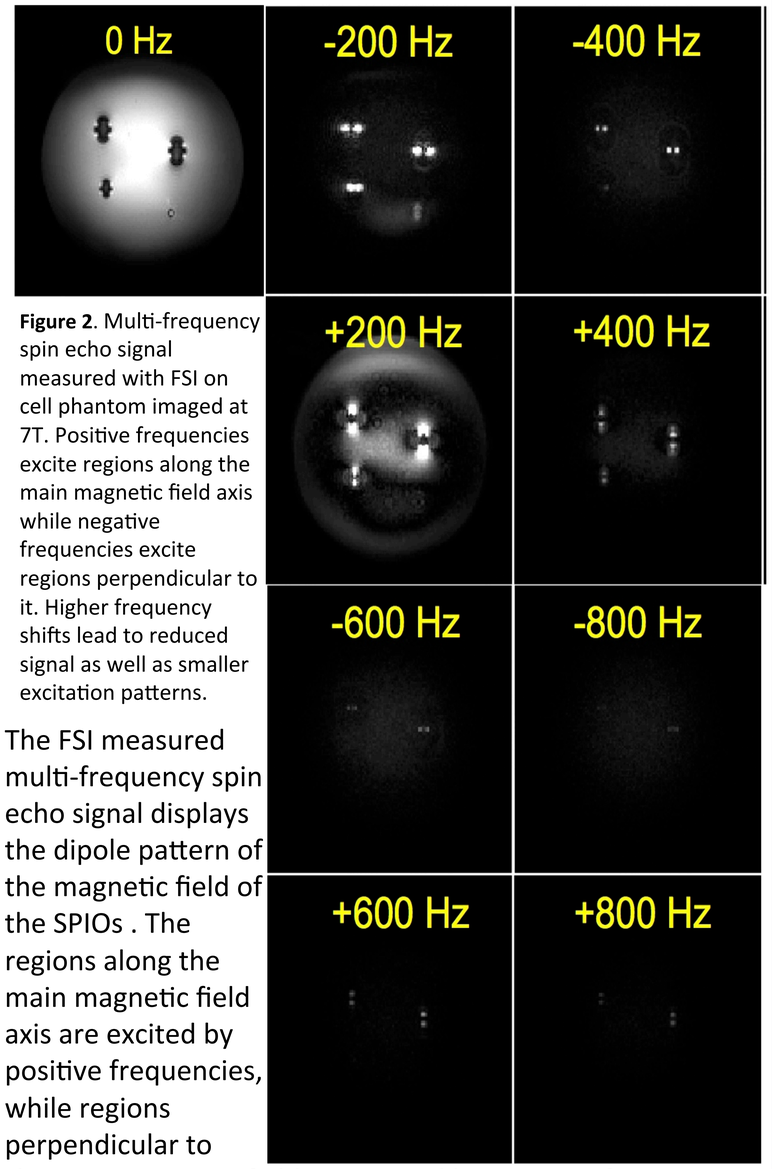

Figure 2. Multi-frequency spin echo signal measured with FSI on cell phantom imaged at 7T. Positive frequencies excite regions along the main magnetic field axis while negative frequencies excite regions perpendicular to it. Higher frequency shifts lead to reduced signal as well as smaller excitation patterns.

The FSI measured multi-frequency spin echo signal displays the dipole pattern of the magnetic field of the SPIOs . The regions along the main magnetic field axis are excited by positive frequencies, while regions perpendicular to the main magnetic field are excited by negative frequencies. This difference in region excitement between positive and negative frequencies is seen in the different orientations of the highlighted dipole patterns. Moreover, lower frequency shifts lead to increased signal and larger excitation patterns, while higher frequency shifts result in the opposite (**Fig 2**). The residual background signal that appears at 200 Hz is a consequence of the large water linewidth at 7T.

Results (continued)

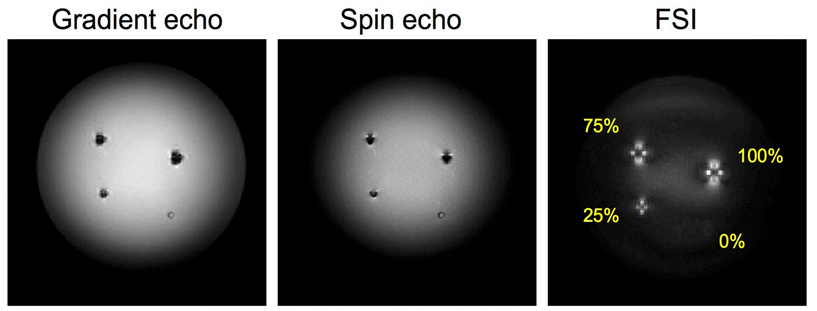

Figure 3. Comparison of gradient echo (left), spin echo (middle) and FSI (right) acquired at 7T on macrophage cultured cell population with 0, 25, 75 and 100% of SPIO-labeled cells. The FSI image was generated using the sum of signals acquired in all frequency bands. While negative contrast and distortions affect standard gradient and spin echo sequences, FSI leads to highly conspicuous signal. Signal intensity and spatial extent of dipole patterns scale with the ratio of labeled cells.

Positive contrast is shown to have increased localization and background suppression compared to negative-contrast sequences (**Fig 3**). The positive contrast technique can provide a framework for quantitative analysis, since it allows for the detection of pattern size, which is visually proportional to the amount of labeled cells.

Conclusions

In this study, we obtained a comprehensive characterization of the SPIO magnetic signature in one acquisition and with fast spiral readout using the novel FSI sequence. We have demonstrated the efficiency of FSI for positive contrast imaging of SPIO labeled macrophages at 7T, as compared to negative contrast methods. Positive contrast imaging suggests a path for quantifying labeled cells in a targeted organ, with significant biomedical applications. For future work, we plan to compare detection limits between 3T and 7T to evaluate the advantages of 7T imaging for SPIOs. We also plan to perform this imaging in *ex vivo* tissue samples and *in vivo* animal models. Our final goal is to develop electromagnetic models to quantify labeled cells content *in vivo*, based on observed multi-frequency patterns.

References

1. Bulte JW, Kraitchman DL. Iron oxide MR contrast agents for molecular and cellular imaging. NMR Biomed. 2004;17(7):484-99.
2. Balchandani P, Yamada M, Pauly J, Yang P, Spielman D. Self-refocused spatial-spectral pulse for positive contrast imaging of cells labeled with SPIO nanoparticles. Magn Reson Med. 2009;62(1):183-92.
3. Cunningham CH, Arai T, Yang PC, McConnell MV, Pauly JM, Conolly SM. Positive contrast magnetic resonance imaging of cells labeled with magnetic nanoparticles. Magn Reson Med. 2005;53(5):999-1005.

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