SPIO quantification using inversion recovery prepared bSSFP for targeted molecular imaging

C. V. Bowen[1,2], and N. A. Pelot[1,3]

[1]Institute for Biodiagnostics (Atlantic), National Research Council, Halifax, NS, Canada, [2]Physics and Atmospheric Science, Dalhousie University, Halifax, NS, Canada, [3]Physics and Atmospheric Science & Electrical Engineering, Dalhousie University, Halifax, NS, Canada

Introduction: Over the past two decades, pre-clinical MR molecular imaging studies using superparamagnetic iron oxide (SPIO) have significantly advanced the fields of cancer, immunology, regenerative medicine and drug development by providing non-invasive visualization of targeted cells and molecules. However, the vast majority of these studies are limited to showing the presence or absence of iron, rather than quantification of the amount of iron present. This lack of accurate quantification greatly limits these techniques in terms of investigating the comparative efficacy of therapeutic approaches, as well as gradations in immunological response. Most recently, balanced-SSFP (bSSFP) acquisitions have demonstrated exquisite sensitivity to SPIO labels [1], with superior off-resonance artifact resistance as compared to gradient echo acquisitions, suggesting their use for quantitative approaches. In fact, Schmitt et al. [2] showed that T_1, T_2 and proton density maps can be obtained from a single IR-prepped balanced SSFP (IR-bSSFP) acquisition.

In this work, we propose the use of IR-bSSFP for SPIO quantification by R_2 mapping. The two main challenges with quantifying SPIO over a practical dynamic range are the need to measure prohibitively short T_2 values, and the requirement that signal be suitably on-resonance for the Schmitt equations [2] to be valid. The difficulty with characterizing short T_2 values can be reduced with smaller flip angles, since this decreases the apparent recovery rate (R_1*) thus facilitating measurement, given that $R_1^* = R_1\cos^2(\alpha/2) + R_2\sin^2(\alpha/2)$ [2]. With a reduced flip angle, the signal becomes more R_1-weighted, thus decreasing R_1*. The difficulty with B_0 inhomogeneity is notable in pre-clinical animal imaging due to significant susceptibility to off-resonance banding artifacts, thus necessitating the acquisition of multiple acquisitions with different RF phase cycling (or frequencies). This problem was addressed through novel post-processing of RF phase-cycled acquisitions, after which IR-bSSFP quantification was achieved *in vitro* by exploiting the linear relationship between relaxation rate and iron concentration.

Methods: Phantoms were prepared using 20 nm SPIO nanoparticles (Ocean Nanotech, Springdale, AR) over a wide range of iron concentrations (0 to 200 µg/mL in 25 µg/mL increments). The nanoparticles were placed in 5mm diameter NMR tubes after suspension in 4% gelatine doped with 8.4 µmol/L of Mn to better emulate physiological R_1/R_2 values. An array of tubes having different SPIO concentrations were supported in a 50mm diameter, Mn-doped water-filled cylinder for imaging. All scans were performed on a 3T magnet equipped with a 21 cm ID gradient coil (Magnex Scientific, Oxford, UK) interfaced with a Varian DD Console (Varian Inc., Palo Alto, Ca) with an in-house built 52mm diameter quad RF coil. Two-dimensional bSSFP was applied as previously described [3], with IR-prepped trains having half flip angle RF catalyzation (single 10mm slice, 100 echoes, TI/TR/TE=10/5/2.5ms, flip angle 20° or 35°, 50mmx50mm FOV, 128x128 matrix, 128 PE segments, 4 RF phase-cycled acquisitions).

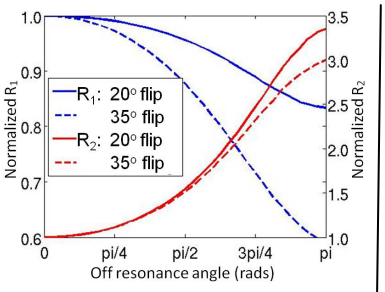

Figure 1: Results of IR-bSSFP Bloch simulation where the R_1 and R_2 values were normalized to their respective on-resonance values. The simulation was performed with R_1/R_2= 2.7/231 s^{-1}, representative of 100 µgFe/mL. Lower iron concentrations led to slightly smaller off-resonance deviations.

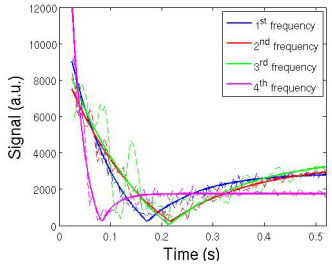

Figure 2: IR-bSSFP time courses for four phase-cycled acquisitions of a single voxel (50 µgFe/mL), where the dashed lines are the data and the solid lines are the fits. The 3rd frequency is rejected due to oscillations. The 2nd frequency has the smallest R_2 value among the remaining three acquisitions, and is therefore identified as being on-resonance.

Bloch simulations were performed in MATLAB (MathWorks, Natick, MA) to evaluate the B_0 inhomogeneity tolerance of R_1 and R_2 estimates in the range applicable for SPIO applications (Fig. 1). Voxel-wise fits of the IR curves for *each* RF phase-cycled acquisition (i.e., each frequency) were performed in MATLAB to obtain R_1 and R_2 values using the functions described in [2] (Fig. 2). To select the on-resonance frequency index, traditional MIP processing is inappropriate because low flip angles produce an off-resonance peak signal. Instead, the most on-resonance IR curve of the four phase-cycled acquisitions was chosen on a voxel-wise basis using a two stage process. First, the acquisition with the largest residuals was rejected due to oscillatory, non-monoexponential behaviour characteristic of stop-band signal, resulting in an unreliable fit [4]. Second, of the remaining three acquisitions, the smallest R_2 value was deemed on-resonance, in accordance with the Bloch simulation results.

Results and Discussion: Bloch simulations (Fig. 1) illustrated that four phase-cycled acquisitions were required, since these produced R_2 quantification errors limited to 11% (peak off-resonance of $\pi/4$), whereas two phase-cycled acquisitions produced unacceptably large R_2 deviations (50% for $\pi/2$ peak off-resonance). IR curves for a representative voxel (Fig. 2) revealed the oscillatory behaviour of off-resonant stop-band signal, and the significant sensitivity to off-resonance from the remaining three curves. Fig. 3 shows a cross-sectional bSSFP image of the phantom (A), along with maps of the R_2 values (B) resulting from the selected frequency indices (C). Very little contrast is evident between tubes with SPIO (A), since traditional bSSFP techniques have good detection, but poor quantification of SPIO concentration across practical dynamic ranges. However, very homogeneous R_2 maps are achievable with our method (B) despite a significant variation in off-resonance within the phantom (C). Fig. 3D shows the capacity of IR-bSSFP to produce linear R_1 and R_2 calibration curves across a substantial dynamic range of SPIO concentrations, with little variance resulting from off-resonance (small error bars showing standard deviation across voxels within tubes). Quantification was achieved up to concentrations of 75 µgFe/mL with 35° flip angles, and 100 µgFe/mL with 20° flip angles. This demonstrates the potential for dynamic range extension through use of lower flip angles.

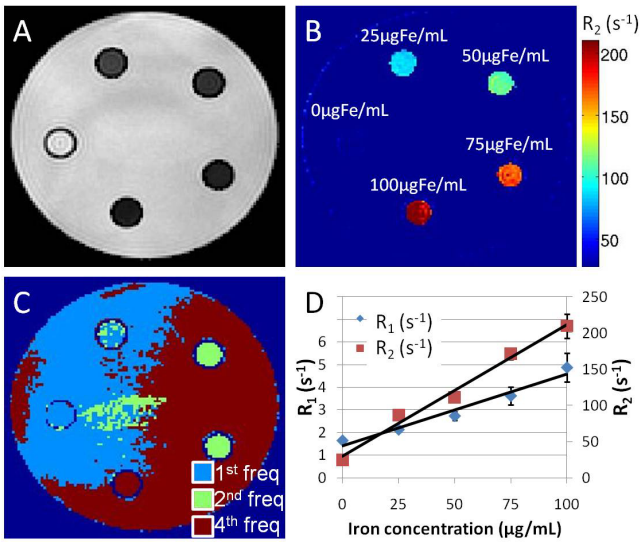

Figure 3: (A) Conventional bSSFP image. (B) R_2 map of 0 to 100 µgFe/mL. (C) Map of selected on-resonance frequencies. (D) IR-bSSFP calibration curves acquired with 20° flip. Relaxation rates were averaged over a 6-by-6 voxel region of interest (ROI) defined within the cross-section of each tube. Error bars indicate standard deviation over the ROI.

Conclusion: We have demonstrated the ability of the IR-bSSFP pulse sequence to quantify SPIO using MRI by employing small flip angles and by addressing issues posed by B_0 inhomogeneity. This technique has potential to significantly advance the use of MRI for pre-clinical studies targeting molecules involved in drugs and disease.

References: [1]Foster-Gareau P et al. Magn Reson Med. 2003;49:968-71. [2]Schmitt P et al. Magn Reson Med. 2004;51:661-7. [3]Oppelt et al. Electromedica. 1986;54:15-8. [4]Deshpande VS et al. Magn Reson Med. 2003;49:151-7.