In Vivo 3D ^{19}F Fast Spectroscopic Imaging (F-uTSI) of Angiogenesis on Vx-2 Tumors in Rabbits Using Targeted Perfluorocarbon Emulsions

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Quantitative molecular MR imaging of angiogenesis may fulfill an unmet clinical need for patient stratification by increasing efficacy of anti-angiogenic therapy. In particular, perfluorocarbons targeted to $\alpha_v\beta_3$ have been used to image angiogenesis through paramagnetic markers and also direct detection by ^{19}F MR imaging or spectroscopy [1,2,3]. ^{19}F offers several advantages including absolute quantification, high intrinsic specificity, and no need for pre-contrast imaging. However, one of the major drawbacks of more clinically relevant ^{19}F compounds is the large chemical shift dispersion of the multiple resonances. If imaged by a gradient or spin echo technique, the resulting images will display a large chemical shift artifact in both the read-out direction and the slice selection direction. Fluorine ultra-fast Turbo Spectroscopic Imaging (F-uTSI) has been developed to overcome these drawbacks, without sacrificing sensitivity [4,5]. Additionally F-uTSI offers, without increasing scan time, the advantage of distinguishing various ^{19}F compounds based on chemical shift differences allowing for 'multi-color' imaging. Beyond the preliminary, non-targeted *in vivo* results already shown, herein we demonstrate with *in vivo* tumor models the sensitive detection of angiogenesis with the F-uTSI technique.

Methods

Male New Zealand White rabbits (~2 kg) were implanted in one hind leg with 2-3 mm Vx-2 carcinoma tumors (National Cancer Institute, MD), which grew to 15 mm within 2 weeks. Imaging was performed 3h post-injection of 1.0 ml/kg of $\alpha v\beta_3$-targeted perfluorocrownether $C_{10}F_{20}O_5$ (20 vol%) NP, incorporating Gd-DTPA-bis-oleate in the outer surfactant. The rabbits were anesthetized with xylazine/ketamine *i.m.* anesthesia and maintained with a 20 ml/h *i.v.* ketamine infusion. All animal care and protocols were in accordance with institutional guidelines. The study was performed on a 3T clinical whole-body scanner (Achieva, Philips Healthcare, The Netherlands) using a dual tuned transmit/receive surface coil (7×12 cm) and a dual ^{19}F/^{1}H spectrometer system. The ^{19}F data were collected with the F-uTSI technique [3,4] which is based on acquiring long spin-echo trains, *e.g.* 16-32 echoes per excitation, where each echo corresponds to one point in k-space. In this way, a 3D data set, *e.g.* 32x32x31, can be sampled in 6 minutes. Typical scan time for detecting angiogenesis are between 15-30 minutes depending on target concentration and desired voxel resolution. For the F-uTSI method we optimized the k-space sampling schemes, a Cartesian scheme was compared to a 'pseudo radial' (fig 1). Using this optimized 'pseudo radial' scheme, a total of 1984 echo trains of 16 echoes each were recorded with TR = 192 ms, TE = 5 ms, echo spacing of 5 ms, and acquired spectroscopic voxel size of 5x5x5 mm^3. The center frequency was set on the CF_2 resonances. Spatial reconstruction was done using the standard software available on the scanner. These data were exported and ^{19}F images were created by integrating the signal intensity of the CF_2 resonances using the 3DiCSI software package developed at the Hatch MR Research Center, Columbia University, New York, USA. High-resolution T1-weighted GRE images were recorded for anatomical co-registration (resolution 0.55×0.55×4.0 mm^3, TR/TE=24/6.5 ms, α=35).

Results and Discussion

The F-uTSI sequence has sufficient sensitivity to detect angiogenesis *in vivo*. An example image (overlaid on a T1w anatomic scan) is shown in fig.2. The main advantage of the F-uTSI technique is that, based on integrating defined spectral peaks, the resulting images are free of chemical shift artifacts, therefore, eliminating need of an accurate determination of the (multiple) resonance frequency of the ^{19}F signal. We use a fixed setting which is calculated from the proton resonance frequency. The first preliminary comparisons with FFE based imaging of ^{19}F [6] show that the sensitivity of F-uTSI and FFE are comparable; an additional advantage of the spin-echo F-uTSI being that it is less sensitive to susceptibility. Optimal results in the *in vivo* studies were obtained with a k-space scheme we have dubbed 'pseudo-radial' (fig. 1)

Conclusion

Fluorine ultra-fast Turbo Spectroscopic Imaging is an efficient and sensitive technique for quantitatively detecting minute amounts of ^{19}F contrast agents *in vivo* while overcoming the confounding problems associated with chemical shift. Employing functionalized perfluorocarbon nanoparticles in tumor-bearing rabbit models, angiogenic maps were created with the F-uTSI technique.

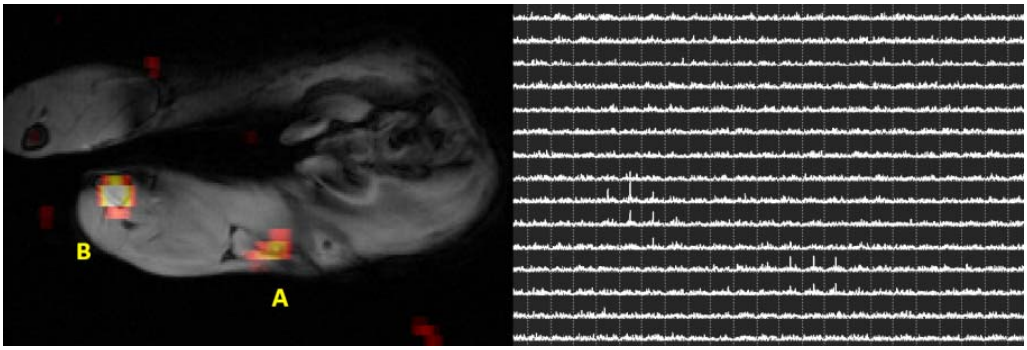

Fig. 2. 3D F-uTSI of angiogenesis. Shown is slice 9 of a 31 slice 3D data set. The ^{19}F data are shown as color overlay on the anatomical image(left panel). Angiogenesis occurs in the major blood vessels leading to the tumor (A). Note that the nanoparticles transiently accumulate in the bone marrow and epiphyseal head (B). The corresponding spectra are shown in the right panel.

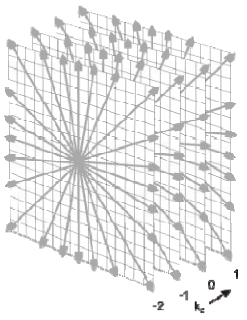

Fig. 1. 'Pseudo radial' k-space sampling. Echo-trains are acquired along radial trajectories from $k_{x,y}$=0 outward; the actual k- values are projected onto the Cartesian grid.[5]. The scheme is extended to 3D with an alternating encoding gradient in k_z direction.

References

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